ABSTRACT

EVANS, CAITLIN E. Evaluation of Unheated, Low Trypsin Inhibitor, Full-Fat Soybeans on Feed Digestibility and Growth Performance of Turkeys at Various Ages. (Under the direction of J. L. Grimes).

The objective of this research was to evaluate new lines of soybeans (SB), naturally selected to contain low anti-nutritional factors such as trypsin inhibitors (TI) and oligosaccharides (OLG) and to determine their potential as a feedstuff for turkeys. These novel SB would, in theory, require little to no processing prior to diet inclusion; in contrast to traditional soybean meal. Consequently, it may be economical to simply incorporate rolled unheated, full-fat SB into poultry feeds as a major source of both protein and energy. Questions, however, arise as to the nutritional availability of energy and amino acids to the bird. A total of four trials were conducted to evaluate and clarify the bioavailability of nutrients when formulating with these novel low trypsin inhibitor (LTI) soybeans.

The initial trial was designed to evaluate 2 novel lines of soybeans: low OLG and low TI (LO) and low TI (LT) for their effect on the growth performance of male turkeys reared from 4 to 19 weeks of age. Poults were reared on pine shavings (PS) or chopped Miscanthus grass (MG). Toms fed diets containing either line of novel SB (LO or LT) had increased BW and FI with a lower overall FCR when compared to birds fed diets with containing commercial SB (CB). Those birds fed the LO or LT also had comparable overall FI when compared to those fed a soybean meal-based diet (SBM), although their BW were lighter the FCR was greater. From this trial it was concluded that LTI SB could be included into turkey rations, but to a limited extent. MG, when used as a litter material through tom market age, resulted in similar growth performance compared to traditional PS, indicating its validity as an acceptable source of poultry bedding.

Trials 2 and 3 evaluated the effect of age on the digestibility of the LTI soybean. Poults began treatments at 0 or 14 days. There were 6 dietary treatments: commercial SB (CB) included at 20 or 40%, commercial soybean meal (SBM), and LTI SB included at 20, 30, or 40%. In Trial 2, increasing LTI from 0 to 40% linearly decreased poult BW and FI from 0 to 14 days, while increasing the FCR when compared to SBM; indicating the TI was still present at levels sufficient to affect the protein digestibility. Performance was improved in trial 3. At day 21, there was only a tendency for reduction in BW with linearly decreasing FI, though no significant difference in FCR. By 28 days, LTI diets remained comparable to SBM in BW, FI, and FCR; indicating that as the poult aged, it was more able to utilize the nutrients in the LTI diets for growth. When comparing CB to SBM or LTI diets, performance was consistently reduced when feeding CB.

Trial 4 evaluated the effect of feed processing on the digestibility of the LTI soybean. Dietary treatments were: soybean meal (SBM), LTI bean included at 20 or 40%. Each of the diets was fed as mash or crumbled pellet. Iincreasing LTI resulted in a linearly reduction of BW with poults consuming crumbles gaining more weight than those consuming mash. LTI inclusion had no effect on FCR at day 21, while crumbled diets had better FCR as compared to mash diets.

Based on the results of these trials, LTI soybeans are a viable ingredient for use in turkey diets. Its effectiveness as an ingredient is improved by feed processing, as shown through improved performance after pelleting. Additionally, the ability of the novel low TI bean to improve PDI without limiting bird growth and performance further increases its value as a feedstuff.

Evaluation of Unheated, Low Trypsin Inhibitor, Full-Fat Soybeans on Feed Digestibility and Growth Performance of Turkeys at Various Ages

by
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BIOGRAPHY

Caitlin E Evans began her undergraduate career at North Carolina State University in the fall of 2007. She was a student research assistant for the majority of her undergraduate career, assisting on several poultry-focused trials ranging from nutrition to incubation. She obtained dual Bachelor's degrees in Animal Science and Zoology with a minor in Agricultural Business. Upon the completion of her undergraduate career, Evans decided to pursue a Master's in Poultry Science working with turkeys. Following the Master's degree, Evans plans to pursue a Ph.D. in Grain Science at Kansas State University located in Manhattan, Kansas, where her focus will be on the role of feed processing on animal nutrition.

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1. INTRODUCTION

Soybeans are the second most planted field crop in the US, after corn (USDA, 2012). In the past several decades there has been a trend of increasing soybean acreage, with over 80 million acres planted in 2014 (Soystats, 2015). Traditionally, soybeans have been classified as an oilseed, comprising approximately 90 percent of the utilized seeds for extraction in the US (USDA, 2012). Reliance on soybeans by the oilseed industry has resulted in a steady supply of the residual meal post extraction. A small percentage of this meal will undergo further processing to create food ingredients including flour, concentrates, isolates and textured protein (Jideani, 2011). The majority, however, will be processed to yield a finished defatted soybean meal of approximately 44 to 48% protein, which can then be used in animal feed (Change et al., 2003; Gillman et al., 2015).

Soybean meal has a favorable amino acid composition that complements many cereal grains used in animal feed, narrowing the deficit in the nutrient requirements in most poultry and swine diets (Liener et al., 1981; Stein et al., 2008; Bruce et al., 2006). Though a high-quality protein source, the defatted soybean meal must undergo further processing to reduce inherent anti-nutritional factors. The toasting or heat treatment of the soybean meal requires high capital investment, but is crucial; as failure to destroy the heat liable anti-nutritional factors in the raw soy results in poor digestibility and thus poor growth (Rackis, 1965). Care, however, must be taken to control the heating process to ensure product quality. Moisture, temperature, time and particle size are all factors in efficient thermal processing (Melcion and van der Poel, 1993; Pacheco-Dominguez, 2015). Failure to control the heating process can lead to irreversible nutrient damage. Of greatest concern is the degradation of heat sensitive amino acids, with the Maillard reaction most commonly observed.

Antinutritional factors present the biggest obstacle to feeding soybeans. Recently, however, new genotypes of soybeans have been selected to contain naturally low levels of the anti-nutritional factors commonly associated with poor growth performance. These improved soybean cultivars may possibly eliminate the necessity of processing, further economizing the use of soybeans in animal feeds (Kumar et al, 2013). In addition to the potential savings in processing cost, the new genotypes retain their natural oil content, resulting in an increased energy value when compared to the traditional de-fatted soybean meal. Depending on the price of other protein and fat sources, the unprocessed, full-fat soybeans could become an economic alternative in least-cost formulation (Swick, 1996).

In an effort to evaluate the new soybean cultivars, it is important to understand what components of the soybean cause the greatest risk to monogastric digestibility. It would appear that the majority of the limitations associated with using raw soybeans can be attributed to the soybean's carbohydrate and protein fractions, as well as a few innate phytochemical compounds.

2. SOYBEAN COMPONENTS AND ANTI-NUTRITIONAL FACTORS

Soybean seeds contains, on average, 21% oil, 35% carbohydrates, and 40% protein (Liu, 1997). In contrast to typical legumes, soybeans are relatively low in carbohydrates, but contain increased amounts of both protein and fat (Lee et al., 2007).

2.1. CARBOHYDRATES

In general, the metabolizable energy (ME) of soybeans in poultry is much lower than its gross energy (GE), which may be due to the poor digestibility of the carbohydrate fraction (Coon et al., 1990; Pierson et al., 1980). Soybean carbohydrates are present in various forms and include glycolipids, monosaccharides, polysaccharides, oligosaccharides, saponins, sterol glucosides, and isoflavones (Eldridge et al., 1979). The majority of these carbohydrate structures pose little risk to monogastric digestibility, with the exception of oligosaccharides.

2.1a. Oligosaccharides

Oligosaccharides are low molecular weight molecules comprised of less than 10 monosaccharide residues connected by glycosidic linkages. They, along with sucrose, comprise over 99% of the sugars in the whole soybean (Hymowitz et al., 1972). Raffinose and stachyose, both soluble α-galacto-ologosaccharides, are highly concentrated in traditional soybean cultivars compared to other legumes (Rackis, 1975); and, when exposed to typical processing conditions, maintain their structure (Leske et al., 1993; Zdunczyk et al., 2011). They are often implicated as causative agents in abdominal discomfort and flatulence when fed to animals at increased levels (De Lumen, 1992; Karr-Lilienthal, 2005; Bainy et al., 2008; Rackis, 1975). This occurs because monogastrics lack the α-galactosidase enzyme in the small intestine, which is responsible for breaking down the α-1,6 galactosyl linkages (Gitzelmann and Auricchio, 1965; Graham et al., 2002). Without the proper digestive enzymes present in the small intestine, the undigested oligosaccharide molecules continue to accumulate in the large intestine. There, exposure to anaerobic microorganisms and subsequent fermentation results in short chain fatty acids and gas production (Karr-Lilienthal et al., 2005; Steggerda, 1968). Additionally, Steggerda (1965) observed increased instances of diarrhea with raffinose and stachyose, which, could not only cause dehydration, but may amplify nutrient absorption issues by increasing the digesta passage rate (Wiggins, 1984). Results of Parsons and others (2000) corroborated these claims and reported an increase in metabolizable energy when chicks were fed soybeans low in oligosaccharides. Thus, it has been suggested that decreasing the amount of oligosaccharide content of the soybean seed through genetic selection, processing, or other means may increase the value and application of the raw ingredient into both human and livestock feed (Hymowitz et al., 1972).

2.2. PROTEINS

The extensive use of soybeans in animal feed can mostly be attributed to the protein composition of the seed. Liener and others (1981) equivocated the high protein quality of soybeans to that of milk protein when used in conjunction with methionine. While soybeans are considered to be the only vegetable food to contain all the amino acids needed in human nutrition (Lonnerdal, 1994), their deficiency in methionine and cysteine prevent the soybean's classification as a complete protein (Delwhiche et al., 2007).

2.2a. Storage Proteins

Storage proteins, mostly consisting of globulins, account for the majority of protein in soybeans. The proteins are classified as 2S, 7S, 11S, or 15S according to the sedimentation coefficient (Singh et al., 2013). Glycinin (11S) and β-conglycinin (7S) account for approximately 80% of these storage proteins (Moriyama et al., 2004), which, in turn, compromise more than 65% of the total protein content of the soybean (Delwiche et al., 2007). Both globulins exhibit allergenic and anti-nutritive effects, with severity dependent on dose, age and species sensitivity. Liu and others (2008) reported impaired immune function, damaged intestinal morphology, and reduced nutrient digestibility in rats as a result of glycinin. Similar results were observed in young swine, with antigenic proteins causing intestinal villi atrophy and GI tract damage, reducing protein digestion and suppressing growth performance (Li et al., 1990; Friesen et al., 1993). Zhao and others (2008) suggested the majority of the antigenic effects observed with soy proteins should be contributed to β-conglycinin due to an increased resistance to proteolytic enzymes when compared to other soy proteins. Though impervious to enzymes, Castro (2014) reported that β-conglycinin was more susceptible to heat than glycinin, however, both globulins could be efficiently denatured to yield superior nutritional value.

2.2b. Lectin

Soy lectins are carbohydrate-binding glycoproteins that have an affinity to binding to terminal N-acetyl-D-glucosamine and D-galactose (Schulze et al., 1995). They can be classified as agglutinating or non-agglutinating based on their degree of denaturation (Irish et al., 1999). Agglutinating lectins are non-denatured and maintain a quaternary structure with multiple carbohydrate-binding sites, while non-agglutinating lectins are partially denatured and have a single carbohydrate binding site (Fasina et al., 2003; Maenz et al., 1999). The additional binding sites on the agglutinating lectins not only enables them to bind to carbohydrates and cells like the non-agglutinating lectins, but also allows them to agglutinate cell membranes. Furthermore, lectins are sometimes referred to as hemagglutin due to their binding capabilities with erythrocytes of higher species.

Biologically, lectins provide protection for the plant from outside antagonists like insects or phytopathogenic organisms. Lectins' defensive role in the plant is well-suited with its high resistance to proteolysis and stability over a wide range of pH (Vasconcelos and Oliveira, 2004), which may contribute to digestibility issues in monogastric animals. Pusztai (1994) reported that undigested lectins bind to the erythrocytes along the brush border membrane of the intestine, damaging morphology and disrupting normal nutrient absorption. Researchers speculate that this intestinal enterocyte damage is the primary cause of growth depression in monogastrics fed high levels of lectin (Pusztai et al. 1979). In contrast, Fasina and others (2006) reported enhanced intestinal development in turkeys fed semi-purified diets supplemented with low or high levels of lectin. They cited the intestine's ability to adapt to dietary changes by altering length, absorptive area and rate of enterocyte turnover as a possible explanation. Though increased villus:crypt ratio was reported, Fasina and others (2006) did recognize the deleterious effects of soy lectin on lymphoid organs, indicating that lectins may lead to a compromised immune system. Douglas and others (1999) reported that soy lectin reduces the growth performance of chicks, though to a lesser extent than trypsin inhibitor. Lectin levels can be reduced or eliminated by heating (Vasconcelos and Oliveira, 2004) or through genetic selection (Douglas et al., 1999; Palacios, 2004).

2.2c. Protease Inhibitors

Protease inhibitors are a class of proteins that disrupt digestive enzymes, impeding protein hydrolysis and digestion. Their presence in legumes is believed to serve as protection against grazing, in addition to acting as a storage protein supporting plant growth (Hwang et al., 1978). Trypsin inhibitors are a group of serine proteases that have been researched extensively and been shown to reduce soybean nutrient utilization in poultry, swine, and mice (Liener, 1981; Rackis, 1965). Of the four identified trypsin inhibitors, the Kunitz and Bowman-Birk inhibitors are the best known and most studied. The Kunitz inhibitor, isolated in 1944, has 181 amino acids and possesses two disulfide bonds (Kunitz, 1944). It exclusively binds and inhibits the digestive enzyme trypsin. The Bowman-Birk inhibitor, identified in 1944 and later purified in 1961, has 71 amino acids with 7 disulfide bonds (Birk et al., 1961). It binds and inhibits both trypsin and chymotrypsin enzymes (Liener, 1994). The presence of multiple disulfide bonds in the Bowman-Birk inhibitor make it highly resistant to degradation and require temperatures in excess of 100ºC to denature. In contrast, the Kunitz inhibitor is much more susceptible to degradation by heat.

In addition to reducing protein digestion through proteolytic enzyme inhibition, trypsin inhibitors also cause pancreas enlargement or hypertrophy, leading to increased secretions. This may indicate that the cause of growth suppression is not entirely due to difficulties in protein digestion, but also a loss of endogenous amino acids resulting from hyper secretions in the pancreas (Liener, 1981). The mechanism behind this biological response is not fully understood, however studies have indicated that trypsin inhibitor is not fully responsible for pancreatic hypertrophy. In a study by Kakade and others (1974) rats fed an inhibitor free soybean extract still had suppressed growth and enlarged pancreas weights. Subsequent in vitro digestion studies lead the authors to conclude that trypsin inhibitors accounted for only 40% of growth inhibition in raw soybeans, with other antinutritional factors accounting for the remaining 60%.

2.3. PHYTOCHEMICALS

2.3a. Saponins

Saponins are a diverse group of glycoside compounds containing a steroidal or triterpenoid aglycone linked to one or more carbohydrate chains (Dourado, 2011). They occur naturally in plants and are present in relatively high concentrations in soybeans (Hu et al., 2002). More specifically, soy saponins are divided into 2 groups – A and B (Gu et al., 2002; Nahashon and Kilonzo-Nthenge, 2011). Group A, typically found in the germ, contain 2 sugar chains and are associated with an undesirable astringent or bitter taste, which may act as a feed deterrent (Hubert et al., 2005). In contrast, Group B, found in the germ or cotyledon, contain a singular sugar chain and is associated with positive health benefits like immune system stimulation and cholesterol binding (Nahashon and Kilonzo-Nthenge, 2011; Oakenfull and Sidhu, 1990). Initial saponin type and concentration can be attributed to soybean cultivar, maturity, and growing region, however, processing and storage conditions can lead to changes in composition and concentration. Saponin structure can be altered by both heat and pH changes (Rickert et al., 2004). There is also some evidence that saponins interact with soy storage proteins, glycinin and β-conglycinin, reducing their sensitivity to chymotrypsin (Shimoyamada et al., 1998).

2.3b. Phytic Acid

Phytic acid (inositol hexakisphosphate, IP6) is the storage form of phosphorous in plant seeds. It accounts for 67-78% of the total phosphorous content in the mature soybean (Raboy et al., 1984). Phosphorous, when bound with phytic acid, has reduced digestibility in monogastric animals. Additionally, phytic acid is generally viewed as an antinutritional factor due to its ability to chelate minerals, amino acids, and starch (Liener, 1981; Wang and Wixon, 1999). Once bound, minerals like calcium, magnesium, zinc, copper, and iron are more difficult to absorb in the small intestine (Liener, 1981). This can lead to poor mineral availability in monogastric animal feeds as well as protein and starch digestibility issues (Liener, 1994; Wang and Wixon, 1999). Phytase enzymes can be included in diets to help ameliorate the negative effects of phytic acid in soybeans. These are an economic way to improve phosphorous availability, while releasing the chelated minerals, protein and starch.

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MANUSCRIPT I:

The effects of *Miscanthus* grass as a bedding source and the dietary inclusion of unheated, low-trypsin inhibitor soybeans on the performance of tom turkeys reared to market age.

ABSTRACT. A study was conducted to evaluate bedding source and the inclusion of rolled, unheated soybeans on the growth performance of male turkeys when reared to market age. Pine shavings are the most common poultry bedding material, however recently, increased marketplace competition has significantly reduced its availability and necessitated the need for economical alternatives. Additionally, the development of new varieties of soybeans with low levels of antinutritional factors have created an opportunity for dietary inclusion with minimal processing. The objective of this study was to determine the efficacy of Miscanthus grass as an alternative to pine shavings litter and to evaluate two novel, low trypsin inhibitor soybean varieties as a potential ingredient for turkeys. Treatments were arranged in a 2 × 4 factorial design with main effects of litter type (Miscanthus grass or pine shavings) and dietary treatment. Diets were defined based on the soy protein source: LO (novel, unheated low oligosaccharide and trypsin inhibitor soybean), LT (novel, unheated low trypsin inhibitor soybean), SBM (commercial solvent extracted soybean meal), or CB (unheated conventional soybeans). Bedding type did not affect BW gain or feed intake at market, however, a tendency for improved FCR was observed when turkeys were raised on Miscanthus grass ($P = 0.073$). Turkey toms fed diets containing low trypsin inhibitor soybeans had improved BW gain and feed intake with a lower overall FCR compared to turkeys fed diets containing conventional raw whole soybeans. Therefore, there was better digestibility of the novel soybean lines. Nutrient utilization of the novel low trypsin inhibitor soybeans, however, remained inferior to diets containing solvent extracted soybean meal, which yielded the heaviest BW and lowest FCR with similar feed intake to the low trypsin inhibitor soybean diets.

Keywords: turkey, soybean, trypsin inhibitor, alternative litter, *Miscanthus* grass

INTRODUCTION

Recent marketplace competition for pine shavings (PS), a common poultry bedding material, has increased the need for viable alternatives. *Miscanthus* grass (MG), traditionally used in the biofuel industry, offers one such alternative. This perennial grass is largely appealing due to its low field management requirements and high yield, averaging 20 tons of dry (90% dry matter based on harvest reports) bedding material per 1 acre harvested. Previous work has been done testing MG's efficacy as a bedding with broilers; however, little is known about its effectiveness when used as a bedding source for tom turkeys reared to market age.

Soybean products provide the major source of protein in many livestock diets including poultry. The heavy reliance on their use is due to the quality and composition of the amino acid profile, which complements cereal grains like corn (Stein et al., 2008). Unfortunately, conventional varieties of soybeans contain several different antinutritional factors (ANF) that have been shown to significantly reduce nutrient utilization and depress growth (Liener and Kakade, 1980). Of these ANF, trypsin inhibitors (TI) are considered to be the most problematic for digestion, though they can be reduced or even destroyed by heating (Herkelman et al., 1993 and Perez-Maldonado et al., 2003). This, however, requires the soybeans to undergo extensive processing including conditioning, flaking, and toasting. Not only is the additional processing expensive, it can also destroy nutrients if not carefully controlled.

Recently, novel high-yield, non-genetically modified lines of soybeans have entered the market that are significantly lower in TI activity when compared to current commercial soybeans. These new soybean varieties lack most or all of one or more of the four protease (trypsin, chymotrypsin) inhibitors commonly present in commercial soybean varieties (Rackis, 1965). Consequently, it may be economical to simply incorporate these rolled unheated, full-fat, low TI soybeans into poultry feeds as a major source of both protein and energy (oil), without the costs associated with processing at a commercial crushing facility. Questions, however, arise as to the nutrient utilization of this raw product when compared to the traditional commercial toasted soybean meal.

MATERIALS AND METHODS

Bird Husbandry

A total of 672 male turkey poults[1], sourced from a commercial hatchery, were weighed on day of hatch and randomly assigned to 48 litter floor-pens with 14 birds each. Poults were reared in a single curtain-sided house on the North Carolina State University Turkey Research Facility (Raleigh, NC) from July to November. Pens were blocked in groups of 12 based on location within the house. Each pen measured 3.4 m^2 and was equipped with one tube feeder and one bell drinker[2] to provide *ad libitum* access to both feed and water for the duration of the study. Heat lamps located in each pen were used for the first 5 wk with adjustments based on ambient temperature and bird comfort. House temperature was maintained at 32° C for the first three days and then gradually reduced by 5° C each week until outside ambient temperature was reached. Incandescent lighting was provided during the first 72 h followed by natural lighting and day length for the remainder of the trial. All handling procedures and euthanasia protocols were carried out in accordance with the guidelines approved by the Institutional Animal Care and Use Committee of North Carolina State University.

[1] Hybrid Converter, Hendrix Genetics Company, Ontario, Canada.
[2] Plasson Livestock, Menashe, Israel.

Litter Type

Initially, all concrete-floor pens received a 5 cm base of PS, with the remaining 20 cm of bedding material comprised of either MG or PS. A single source of *Miscanthus* × *giganteus*, a sterile hybrid grass, was harvested after field drying to approximately 90% dry matter. Once harvested, the MG was chopped to 25 mm using a commercial tub grinder (Model TG5000, Vermeer Equipment, Pella, IA). Litter type was evenly distributed among dietary treatments, resulting in 24 pens of each litter treatment. Pen litter condition was visually evaluated biweekly and observations recorded. When necessary, pens were caked and top-dressed with their respective bedding type.

Dietary Treatments

All diets were formulated to meet or exceed breeder recommendations. Diets were formulated with one of four different soy protein sources: solvent extracted soybean meal (SBM), unheated conventional soybeans (CB), novel unheated low oligosaccharide (OLG) and TI soybeans (LO), or novel unheated low TI soybeans (LT). Genetic lines of soybeans included e2011×812×731 (CB), e3032S Trivecta (LO), and e3311 (LT) (eMerge Genetics, Des Moines, IA). The full-fat, unheated soybeans were analyzed for proximate analysis prior to formulation (Table 1). Dietary treatments were randomly assigned to 48 pens based on 4 locational blocks of 12 pens each. The feeding program was segmented into seven phases to meet the changing nutrient requirements of the toms: Starter 1, Starter 2, Grower 1, Grower 2, Grower 3, Finisher 1, and Finisher 2 (Table I-2). Diets were formulated to be iso-caloric and iso-nitrogenous within each feeding phase. Poults were allowed *ad libitum* access to feed, with Starter 2 and later phases provided on a kilogram per bird basis. A commercial-type crumbled pellet was fed to all poults during the Starter 1 phase from placement to four weeks of age to provide an acclimation period prior to the onset of coarse, mash dietary treatments in phase 2. Once dietary treatments began, total feed allotment per pen of birds was adjusted based on any mortalities or culls. Upon the completion of a feeding phase, the next phase was begun.

Feed Manufacturing

All feed was manufactured in accordance with current Good Manufacturing Practices (CGMP) at the North Carolina State University Feed Mill Educational Unit (Raleigh, NC). All grains and soybeans were ground using a double high roller mill (Model C128829, RMS, Harrisburg, SD) equipped with 305 mm diameter rolls measuring 406 mm long. Furnished with two 15 hp motors, the approximate roll differentials were 1.5:1 on the top roll set and 2:1 on the bottom. The roll gap or distance between roll sets was adjusted manually for cereal grains and whole beans to yield finished diets with an estimated particle size of 700 microns. Whole corn was ground using a 0.64 mm gap between the top set of rolls and 0.56 mm gap between the bottom set, while wheat was ground with a gap setting of 0.20 mm between both the top and bottom set of rolls. Both the LO and LT novel unheated full-fat experimental beans were ground with 0.20 mm and 0.15 mm roll gaps, respectively. In order to compensate for different seed characteristics, the CB unheated full-fat commercial soybean was ground with 0.20 mm roll gaps. Grind settings remained constant throughout the manufacture of all feeding phases in the trial. A 3.6 m^3 counterpoise mixer (Model TRDB126-0604, Hayes and Stolz, Ft. Worth, TX) was used to mix each treatment for the standard time of 270 seconds. Finished feed was then transported in bulk to the trial site.

Feed Analysis

Finished feed samples were analyzed to determine proximate analysis, complete amino acid profile, trypsin-inhibitor level, and urease activity by the Agriculture Experiment Station

Chemical Laboratories at the University of Missouri (Columbia, MO). The particle size of major ingredients and dietary treatments were determined according to dry sieving method ASAE S319.2 with a 10 min run time (ASAE, 1995). Analyses included the addition of sieve agitators and 0.5 g of dispersing agent[3] per 100 g of sample.

Live Performance

Individual bird and feeder weights were collected at 4, 6, 9, 12, 15, and 19 wk of age to determine BW, BW gain (BWG), feed intake (FI), and FCR. Mortalities were recorded daily and feed allotments adjusted accordingly. FCR was calculated for each weigh period and adjusted based on period mortalities by dividing the pen FI by the pen BWG plus BW of dead birds or culls from the specified period.

Statistical Analysis

All data were analysed as a 2 × 4 factorial in a randomized block design with 6 replicates per treatment. Main effects were defined as the litter type (MG or PS) and the source of soy protein (LO, LT, SBM, or CB). The measured growth parameters were analysed using the Fit Model procedure in JMP 10 (SAS Institute, Cary, NC) with least square means separated with Tukey adjustment. Treatment main effects and interactions were considered significant at $P \leq 0.05$. The experimental unit was defined as the pen.

RESULTS

Feed Analysis

Results for the nutrient analysis of starter, grower, and finisher phase feeds were as expected and are presented in Tables I-3-I-5. Analyzed TI values remained on trend throughout all dietary phases. In all phases the lowest TI levels were observed in the SBM diet, followed by

[3] Silicon Dioxide, model SSA-58, Gilson, Lewis Center, OH.

intermediate levels in the two novel soybean diets (LO and LT), and the greatest values in the CB diet. Particle size analysis of the commercial soybean meal used in the SBM diets resulted in a slightly greater geometric mean diameter (Dgw) value than those of the rolled soy protein sources (Table I-6). This likely contributed to the increased particle size in the SBM complete diets beginning with the Starter 2 phase (Tables I-7-I-9).

Live Performance

There was a total of 6.1% mortality across all treatments, with 1.9% occurring within the first 4wk, prior to the start of dietary treatments. Birds were culled as necessary based on bird health or mis-sexing. Cull rate was 4.3% for the duration of the trial. Treatment interactions and main effects of litter type and diet on live performance are presented in Tables I-10-I-13. The only observed interaction occurred at 19 weeks for BW and BWG. This interaction was driven by the birds receiving the LO and LT diets housed on PS having similar BW and BWG when compared to the other treatments which had differing weights based on diet regardless of litter type. While statistically different, there is no known biological reason as to why this response was observed.

Litter Type Main Effect on Live Performance

There were no observed differences in pen management when rearing poults on MG versus PS. The frequency of caking and top-dressing remained similar between both litter types. At 4 wk of age, poults housed on PS weighed significantly more and had marginally increased FI when compared to poults on the MG. Litter type showed no further impact on BW or BWG of poults reared through 19 weeks. While there was no overall consistent effect on feed consumption, poults raised on MG had significantly lower feed conversion compared to those housed on pine shavings at 6 weeks and marginally improved feed conversion at 9 weeks of age.

Additionally, at the conclusion of the trial, the *Miscanthus* grass birds were approaching significantly lower overall FCR compared to those reared on the pine shavings ($P = 0.073$).

Dietary Treatment Main Effect on Live Performance

There were no differences observed in body weight at placement or at 4 weeks of age when the dietary treatments began. At 6 weeks of age a clear pattern emerged with the SBM birds weighing significantly more than birds fed either of the experimental beans (LO and LT), where treatment birds fed these two beans were not significantly different from each other. Birds fed the unprocessed commercial beans (CB) weighed significantly less than all others. This trend in bird body weight was observed at 6, 9, 12, and 19 wk of age. The only deviation occurred at 15 weeks when the birds fed the LO diet had greater average body weight than those fed the LT diet, thus changing from a three-tier structure to a four-tier based on significance. FI was significantly depressed by the inclusion of the CB soybean, with birds fed CB never reaching the Finisher 2 feeding phase. Furthermore, from 9-15 weeks the birds fed the experimental bean diets (LO and LT) consumed less than those fed the SBM diet. However, during the last period from 15-19 weeks, the birds responded with similar feed consumption to those fed the SBM diet. Significant effects on FCR based on dietary treatments were observed as early as six weeks. The birds on the SBM diet had the lowest FCR throughout the trial, though, at times, one or both of the novel beans (LO and LT) resulted in comparable FCR.

DISCUSSION

Litter Type

Miscanthus grass under the conditions of this trial proved to be a viable bedding source for growing tom turkeys to market age. At the end of the trial, there were no differences in BW, BWG, FI, or FCR when compared to poults reared on pine shavings. These results are similar to those reported by Smith, who found no difference in BW or FCR in turkey hens reared to 13 wk on chopped Bermuda grass or pine shavings (Smith, 2002). Additional work with broilers has also resulted in similarities in growth performance when rearing on chopped grass materials versus pine shavings (Davis et al., 2010; Davis et al., 2015, Davis et al., 2019; Hulet et al., 2010; Nakaue et al., 1978; and Nakaue et al., 1995). With lack of effect on growth performance, the greatest deterrent for commercial use of grass bedding may be perceived difficulties with material management and handling. These fears, however, remained unsubstantiated in this trial; and contrary to observations by Malone (1992), there were no notable differences in pen management when rearing on pine shavings or *Miscanthus* grass. In both instances, pens were caked and top dressed as needed with no obvious differences in frequency of de-caking or litter quality observed. It is possible that the grass caking issues reported by Malone were a result of the chop size and preparation of the grass as opposed to the material itself. Nakaue and others (1995) noted the importance of correct preparation of grasses in achieving a viable litter source. They found that while the preparation method had little effect on growth, significant differences in litter quality, i.e. caking, could result from improper processing of the litter prior to use. Thus, ensuring that the *Miscanthus* grass in this trial was chopped to an average 2.5 cm in length was crucial in maintaining its ability to successfully control moisture and minimize caking within the pen. While this study clearly showed the effectiveness of *Miscanthus* grass as a bedding source for tom turkeys reared to 19 weeks, the value of a litter is also dependent on its end use. Grimes and colleagues (2002) stated that for a litter source to be successful it must have an additional purpose after its use. Subsequently, *Miscanthus* grass has shown potential as a source for biochar production and as a substrate for manure composting prior to land application (Janus et al., 2015 and Leth et al., 2001). Based on the results of this trial, *Miscanthus* grass appears to be a promising alternative bedding source that warrants additional research in a commercial setting under typical management conditions.

Dietary Treatment

In this trial, two novel varieties of soybeans low in ANF were tested for growing turkeys. The relatively high protein and energy requirements of turkeys made them an ideal model for this trial and allowed for maximum dietary inclusion of the unheated soybeans. Levels of ground soybeans, initially included at 42.5% in the starter diet, were decreased through grower (40%) and finisher (34% and 31%, respectively) phases to meet the changing needs of the tom. The natural oil content of the unheated soybeans provided additional benefits as an energy source when formulating, resulting in less supplemental fat addition. The full-fat, unheated soybeans were assumed an energy value of 3300 Kcal/kg based on the NRC value listed for heat processed, full-fat soybeans (NRC, 1994). Dietary treatments began at 4 wk once proper gut enzyme activity was established. Kroghdahl and Sell (1988) reported trypsin and lipase pancreas activities were not consistent in turkeys until 28 days, with stabilized intestinal secretions at 21 days. Mature poults reduced possible bias and substantiated that observed differences were likely a result of digestibility issues rather than impaired development of the GI tract.

Even with matured gut functions, young poults did not fully utilize the unheated soybean diets. Toms receiving the CB diet and subsequently the highest levels of TI, were the lightest with the highest FCR. The depressed FI on CB diets as well as poor digestibility were likely both contributing factors to the reduced performance, especially considering few pens on this treatment reached the finisher phases containing the lowest inclusion levels of ground soybeans. In contrast, the reduced levels of ANF in the LT and LO diets did result in improved growth when compared to the commercial unheated soybean in the CB diet. Comparatively, the LT and

LO diets yielded heavier birds with lower FCR suggesting improved digestibility with the novel lines of unheated soybeans. No consistent performance differences were observed between the LT and LO diets throughout the trial; and analysed TI levels remained similar across phases. Consequently, the reduced soybean OLG levels in the LO diet appeared to have little effect on digestibility as also seen by Irish and colleagues (Irish et al., 1995). The lack of improvement contrasts works by Coon et al. (1990) and Parsons et al. (2000) who both agreed that the removal of OLG from soybeans increased its ME. Since OLG levels were not directly measured in this trial a definite conclusion cannot be offered to explain the lack of growth response. While the LT and LO diets had marked improvement over the CB diet, levels of ANF were likely still too high to optimize growth when compared to the SBM diet containing solvent extracted soybean meal. Poults consuming the SBM diet had the greatest BW and lowest FCR throughout the entire trial; however, as expected, summer temperatures and mash diets led to reduced performance when compared to breed standards.

The results of this trial are similar to work reported with broilers where select soybeans with low TI exhibited increased nutritive value when compared to conventional soybeans, yet remained inferior to processed meal (Herkelman et al., 1993; Perez-Maldonado et al., 2003, and Han et al., 1991). The literature offers a multitude of explanations and theories on the reduced digestibility of raw soybeans compared to meal; however, within the confines of this trial using novel lines of soybeans, the observed responses can likely be attributed to the remaining ANF in the unheated soybeans, the high inclusion levels used, and the age of the turkeys. It is probable that the large inclusion levels coupled with residual ANF in the unheated soybeans were too great for young turkeys to handle early in the trial. Mian and Garlich (1995) recommended that TI levels not exceed 3 mg TIU per g of diet for turkeys between 6 and 8 wk of age, whereas the initial starter 2 diets in this trial contained between 6.7 to 9.5 mg TIU per g of diet. In addition to TI levels, the younger poults may have also struggled to handle the increased levels of other ANF, like lectin, which would have been compounded by the high inclusion levels (Kakade et al., 1973 and Friedman et al., 1991). It is suggested that lowering the inclusion levels of the unheated low TI soybeans in the starter and grower phases may have allowed birds to maintain comparable performance to the SBM diet. Limiting inclusion of the unheated soybeans to 25% of the diet may have garnered similar results to those reported by Han and others (1991), where unheated low TI soybeans performed similar to the control in broilers.

CONCLUSIONS AND APPLICATIONS

1. Miscanthus grass, when used as a litter material through tom market age, resulted in similar growth performance compared to traditional pine shavings, indicating its validity as an acceptable source of poultry bedding.
2. Novel lines of unheated soybeans naturally low in ANF, like trypsin inhibitor, improve turkey performance when compared to traditional commercial lines.
3. Additional work is needed to determine the optimum inclusion levels of unheated low ANF soybeans in turkey diets.

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Table I-1. Analyzed nutrient content of unheated, whole soybeans used to manufacture dietary treatments fed to male turkeys from 4 to 19 weeks of age.

Nutrient	LO[1]	LT[2]	CB[3]
	----------- % of Total -----------		
Crude Protein	39.73	39.34	38.99
Moisture	8.12	7.95	8.18
Crude Fat	9.61	8.57	7.17
Crude Fiber	18.19	17.59	17.79
Ash	5.10	4.70	3.50
Total Lysine	2.54	2.67	2.61
Tryptophan	0.56	0.57	0.56
Cystine	0.53	0.50	0.51
Methionine	0.53	0.52	0.53
Aspartic Acid	4.53	4.42	4.41
Threonine	1.50	1.47	1.49
Serine	1.99	1.95	1.92
Glutamic acid	7.35	7.30	7.00
Proline	1.98	1.98	1.95
Glycine	1.66	1.64	1.64
Alanine	1.65	1.63	1.63
Valine	1.90	1.89	1.88
Isoleucine	1.83	1.80	1.81
Leucine	2.99	2.94	2.91
Tyrosine	1.35	1.31	1.33
Phenylalanine	1.99	1.97	1.96
Histidine	1.05	1.04	1.00
Arginine	3.00	3.01	2.89
Trypsin inhibitor, TIU/g	18,200	10,300	29,800

[1] LO represents an unheated low trypsin inhibitor and oligosaccharide soybean variety.
[2] LT represents an unheated low trypsin inhibitor soybean variety.
[3] CB represents an unheated commercial soybean variety.

Table I-2. Composition of dietary treatments by phase fed to male turkeys from hatch to 19 weeks of age.

Dietary Phase	Starter 1	Starter 2		Grower 1		Grower 2		Grower 3		Finisher 1		Finisher 2	
Feed per bird (kg)[1]	----	13.2		30.8		44.0		55.0		66.0		55.0	
Diet[2]	----	SBM	RB	SBM	RB	SBM	RB	SBM	RB	SBM	RB	SBM	RB
Ingredients	% of Total Diet												
Corn	40.00	27.20	22.90	30.60	26.80	34.30	30.40	36.10	32.00	40.20	37.00	42.30	39.20
Wheat	0.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00
Soybean meal	37.50	32.38	0.00	30.50	0.00	30.40	0.00	30.30	0.00	26.00	0.00	23.80	0.00
Whole soybeans	0.00	0.00	42.50	0.00	40.00	0.00	40.00	0.00	40.00	0.00	34.00	0.00	31.20
Poultry meal	10.00	8.00	8.00	6.00	6.00	2.00	2.00	0.00	0.00	0.00	0.00	0.00	0.00
Soy oil	6.50	6.63	0.95	7.02	1.55	8.00	2.58	8.65	3.23	8.74	4.13	9.15	5.03
Limestone	1.85	1.80	1.70	1.80	1.73	1.73	1.65	1.60	1.55	1.63	1.55	1.45	1.40
Monocalcium P, 21%	2.40	2.30	2.25	2.38	2.30	2.10	2.00	2.00	1.95	2.00	1.95	1.93	1.85
L-lysine HCl	0.45	0.48	0.38	0.50	0.40	0.30	0.20	0.19	0.10	0.25	0.18	0.21	0.15
DL-methionine	0.43	0.35	0.38	0.36	0.38	0.25	0.25	0.21	0.23	0.21	0.23	0.19	0.20
Sodium chloride	0.23	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Sodium bicarbonate	0.10	0.10	0.10	0.10	0.10	0.18	0.18	0.20	0.20	0.23	0.23	0.23	0.23
Choline chloride, 60%	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Mineral premix[3]	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Vitamin premix[4]	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Selenite premix[5]	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
L-threonine	0.10	0.10	0.10	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Calculated Analysis													
Crude protein	29.50	27.10	27.10	25.00	25.00	22.30	22.30	20.90	20.90	19.00	19.00	18.10	18.10
Crude fat	9.10	9.00	10.60	9.20	10.60	9.80	11.20	10.30	11.70	10.50	11.70	10.90	12.10
ME, kcal/kg	3131	3234	3238	3274	3271	3355	3355	3406	3403	3447	3450	3500	3500

[1]Feed allotted to birds per feeding phase in kilograms.
[2] Control SBM diet formulated with solvent extracted soybean meal, while WB indicates diets LO, LT, and CB containing unheated rolled soybeans.
[3] Each kilogram of mineral premix (0.1% inclusion) supplies the following per kg of complete feed: Iron 20 mg; Copper 2.5 mg; Manganese 30 mg; Zinc 30 mg; Iodine 1.25 mg, Cobalt 1.0 mg per kilogram of complete feed.
[4] Each kilogram of vitamin premix (0.1% inclusion) supplies the following per kg of complete feed: Vit A 13,200 IU; Vit D3 3,960 IU; Vit E 66 IU; Vit B1 4 mg; Vit B2 13 mg; Vit B 6 8 mg; Vit B12 40 µg; Vit K3 4 mg; Nicotinic acid 110 mg; Pantothenic acid 22 mg; Folic acid 2.2 mg; Biotin 254 µg;
[5] $NaSeO_3$ premix provides 0.3 mg Se per kilogram of complete feed.

Table I-3. Analysed nutrient content of dietary treatments fed to male turkeys during the Starter phases.

Phase	Starter 1	Starter 2			
Diet[1]	---	LO	LT	SBM	CB
Nutrient	% of Total				
Crude Protein	30.79	29.85	29.91	28.29	29.94
Moisture	10.89	9.92	10.41	11.69	11.32
Crude Fat	9.17	12.98	11.47	8.07	11.12
Crude Fiber	1.92	3.19	3.02	2.29	3.09
Ash	9.11	7.77	7.58	7.95	8.19
Calcium	2.08	1.68	1.45	1.51	1.71
Phosphorous	1.26	1.22	1.09	1.14	1.21
Taurine	0.09	0.08	0.08	0.08	0.08
Hydroxyproline	0.27	0.18	0.26	0.17	0.23
Aspartic Acid	2.87	2.82	2.72	2.67	2.86
Threonine	1.18	1.17	1.13	1.08	1.16
Serine	1.29	1.28	1.16	1.16	1.23
Glutamic Acid	4.74	4.97	4.90	4.79	5.09
Proline	1.87	1.84	1.89	1.81	1.92
Lanthionine	0.00	0.00	0.00	0.00	0.00
Glycine	1.54	1.51	1.64	1.46	1.64
Alanine	1.51	1.38	1.40	1.36	1.41
Cysteine	0.37	0.39	0.38	0.39	0.41
Valine	1.44	1.25	1.42	1.32	1.41
Methionine	0.79	0.72	0.80	0.86	0.85
Isoleucine	1.35	1.18	1.19	1.21	1.27
Leucine	2.39	2.22	2.18	2.19	2.24
Tyros	0.95	0.99	0.96	0.94	0.99
Phenylalanine	1.41	1.39	1.37	1.36	1.43
Hydroxylysine	0.06	0.06	0.07	0.06	0.06
Ornithine	0.02	0.01	0.02	0.02	0.02
Lysine	2.10	1.96	2.03	1.96	2.04
Histidine	0.72	0.70	0.69	0.68	0.71
Arginine	1.96	2.00	1.99	1.83	2.05
Tryptophan	0.37	0.24	0.23	0.29	0.23
Trypsin Inhibitor, TIU/g	---	7017	6725	1580	9488
Urease Activity[2]	---	2.33	2.31	0.27	2.37

[1] Common starter 1 diet followed by treatments: LO and LT (ground unheated low trypsin inhibitor soybeans), SBM (soybean meal), and CB (ground unheated commercial soybean).

[2] Urease activity presented in net pH increase.

Table I-4. Analysed nutrient content of dietary treatments fed to male turkeys during the Grower phases.

Phase	Grower 1				Grower 2				Grower 3			
Diet[1]	LO	LT	SBM	CB	LO	LT	SBM	CB	LO	LT	SBM	CB
Nutrient	% of Total											
Crude Protein	24.54	24.35	23.33	24.95	24.80	25.16	25.64	24.58	24.44	21.93	23.36	22.94
Moisture	11.31	11.77	13.19	12.44	10.29	10.19	10.72	10.71	10.97	11.07	11.69	10.85
Crude Fat	10.04	9.25	7.29	9.81	8.04	7.84	8.68	8.04	9.61	8.57	7.17	9.13
Crude Fiber	2.85	2.82	2.15	2.85	5.27	4.98	2.80	4.03	3.27	3.46	2.50	3.26
Ash	6.68	6.44	6.93	6.79	7.52	7.58	8.08	7.82	6.26	6.77	6.60	6.75
Calcium	1.26	1.32	1.30	1.44	1.36	1.38	1.52	1.47	1.12	1.26	1.10	1.27
Phosphorous	0.92	0.90	0.97	1.09	1.00	0.92	1.01	1.01	0.82	0.99	0.75	1.01
Taurine	0.07	0.07	0.07	0.07	0.07	0.06	0.06	0.06	0.17	0.17	0.17	0.16
Hydroxyproline	0.16	0.19	0.15	0.21	0.19	0.14	0.15	0.20	0.03	0.03	0.02	0.03
Aspartic Acid	2.35	2.21	2.18	2.23	2.35	2.38	2.62	2.32	2.19	2.11	2.23	2.24
Threonine	0.85	0.81	0.79	0.81	0.88	0.88	0.96	0.85	0.82	0.80	0.85	0.85
Serine	0.97	0.93	0.87	0.92	1.09	1.02	1.13	0.97	1.01	0.99	1.02	1.03
Glutamic Acid	4.09	3.96	3.97	3.91	4.53	4.46	4.74	4.29	4.16	4.16	4.17	4.13
Proline	1.50	1.47	1.45	1.44	1.36	1.34	1.41	1.32	1.31	1.34	1.35	1.32
Lanthionine	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Glycine	1.19	1.20	1.20	1.22	1.17	1.08	1.18	1.18	0.92	0.90	0.95	0.94
Alanine	1.14	1.12	1.13	1.13	1.15	1.12	1.23	1.13	1.01	1.00	1.06	1.04
Cysteine	0.33	0.31	0.31	0.31	0.33	0.36	0.37	0.35	0.37	0.35	0.39	0.35
Valine	1.23	1.20	1.21	1.22	1.15	1.18	1.28	1.18	1.01	1.00	1.06	1.05
Methionine	0.67	0.62	0.59	0.68	0.61	0.67	0.66	0.64	0.53	0.53	0.57	0.56
Isoleucine	1.04	0.99	1.01	1.01	1.00	1.05	1.12	1.03	0.91	0.89	0.95	0.93
Leucine	1.90	1.82	1.81	1.82	1.94	1.99	2.14	1.89	1.80	1.78	1.83	1.83
Tyros	0.80	0.77	0.72	0.72	0.86	0.86	0.91	0.84	0.73	0.72	0.75	0.75
Phenylalanine	1.16	1.12	1.10	1.13	1.18	1.21	1.24	1.18	1.08	1.07	1.11	1.11
Hydroxylysine	0.04	0.04	0.04	0.05	0.06	0.04	0.02	0.06	0.03	0.03	0.05	0.03
Ornithine	0.02	0.02	0.02	0.02	0.02	0.01	0.01	0.01	0.01	0.01	0.01	0.01
Lysine	1.70	1.70	1.71	1.68	1.55	1.61	1.72	1.59	1.29	1.26	1.37	1.33
Histidine	0.61	0.59	0.56	0.58	0.64	0.63	0.68	0.62	0.57	0.56	0.58	0.57
Arginine	1.70	1.66	1.53	1.64	1.68	1.74	1.78	1.66	1.51	1.50	1.51	1.56
Tryptophan	0.18	0.19	0.28	0.16	0.16	0.17	0.28	0.15	0.17	0.17	0.26	0.21
Trypsin Inhibitor, TIU/g	6467	6311	912	9975	3248	3023	976	4115	6072	5478	916	9704
Urease Activity[2]	2.02	1.98	0.04	2.00	2.07	2.11	0.10	1.97	2.00	1.99	0.00	1.97

[1] Common starter 1 diet followed by treatments: LO and LT (ground unheated low trypsin inhibitor soybeans), SBM (soybean meal), and CB (ground unheated commercial soybean).

[2] Urease activity presented in net pH increase.

Table I-5. Analysed nutrient content of dietary treatments fed to male turkeys during the Finisher phases.

Phase	Finisher 1				Finisher 2			
Diet[1]	LO	LT	SBM	CB	LO	LT	SBM	CB
Nutrient	% of Total							
Crude Protein	21.76	21.36	20.67	21.13	19.69	20.44	19.97	19.88
Moisture	11.08	11.28	12.87	11.15	11.14	11.61	11.70	11.70
Crude Fat	8.21	8.00	9.35	8.40	9.14	9.87	8.55	9.16
Crude Fiber	3.89	3.48	2.22	3.02	3.13	2.74	2.10	2.55
Ash	5.57	6.06	7.05	6.98	5.86	5.69	6.09	5.79
Calcium	1.42	1.09	1.06	1.25	1.07	1.05	1.24	1.13
Phosphorous	0.91	0.82	0.78	0.83	0.82	0.88	0.91	0.82
Taurine	0.17	0.18	0.18	0.18	0.18	0.18	0.18	0.18
Hydroxyproline	0.03	0.03	0.02	0.03	0.03	0.03	0.02	0.02
Aspartic Acid	2.01	1.90	1.81	1.89	1.83	1.84	1.69	1.87
Threonine	0.76	0.73	0.71	0.72	0.70	0.71	0.66	0.71
Serine	0.97	0.89	0.88	0.88	0.87	0.88	0.80	0.87
Glutamic Acid	3.94	3.72	3.66	3.69	3.65	3.68	3.41	3.61
Proline	1.27	1.21	1.19	1.20	1.19	1.20	1.13	1.19
Lanthionine	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Glycine	0.85	0.82	0.79	0.82	0.79	0.80	0.76	0.80
Alanine	0.99	0.96	0.92	0.92	0.90	0.92	0.91	0.92
Cysteine	0.35	0.30	0.31	0.30	0.28	0.31	0.29	0.32
Valine	0.94	0.95	0.88	0.92	0.90	0.89	0.85	0.91
Methionine	0.58	0.52	0.41	0.50	0.45	0.45	0.40	0.48
Isoleucine	0.83	0.83	0.79	0.82	0.79	0.78	0.75	0.80
Leucine	1.74	1.70	1.62	1.61	1.61	1.61	1.57	1.64
Tyros	0.70	0.67	0.64	0.65	0.65	0.64	0.61	0.65
Phenylalanine	1.01	0.98	0.94	0.97	0.95	0.95	0.89	0.96
Hydroxylysine	0.03	0.03	0.03	0.03	0.05	0.05	0.03	0.05
Ornithine	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
Lysine	1.24	1.17	1.21	1.23	1.13	1.18	1.08	1.14
Histidine	0.54	0.51	0.48	0.50	0.50	0.51	0.46	0.50
Arginine	1.39	1.34	1.21	1.32	1.28	1.31	1.14	1.30
Tryptophan	0.15	0.16	0.25	0.16	0.17	0.17	0.24	0.18
Trypsin Inhibitor, TIU/g	5401	7152	759	9600	7960	8062	535	9361
Urease Activity[2]	1.96	2.01	0.00	1.91	1.99	1.94	0.00	1.85

[1] Common starter 1 diet followed by treatments: LO and LT (ground unheated low-trypsin soybeans), SBM (soybean meal), and CB (ground unheated commercial soybean).

[2] Urease activity presented in net pH increase.

Table I-6. Particle size analyses of grains and soy protein source included in male turkey diets fed from hatch to 19 weeks of age.

Ingredient		Corn	Wheat	SBM[1]	LO[2]	LT[3]	CB[4]
U. S. Sieve, #	Sieve Size, µm	------------------------- % retained on sieve -------------------------					
4	4760	0.06	0.01	0.06	0.01	0.03	0.03
6	3360	5.91	0.10	0.10	0.04	0.49	0.43
8	2380	5.88	4.15	1.72	0.57	1.28	1.58
12	1680	22.42	16.96	15.62	3.93	4.79	8.82
16	1190	34.12	24.10	30.20	22.57	24.75	26.69
20	840	13.12	12.05	19.98	25.64	24.30	22.40
30	590	5.56	9.72	13.57	21.47	18.84	15.93
40	420	3.83	8.17	8.45	11.33	10.52	9.12
50	297	1.65	5.61	4.30	4.99	5.26	4.51
70	210	1.02	3.22	2.08	2.56	2.77	2.76
100	149	1.16	1.98	1.00	2.41	1.51	1.35
140	105	1.47	2.12	0.72	1.06	1.94	1.99
200	74	1.00	1.75	0.50	0.95	0.86	2.04
270	53	1.33	0.90	0.36	1.82	1.38	1.28
Pan	37	6.06	9.15	1.35	0.67	1.27	1.07
Dgw, µm[5]		976	684	957	750	772	809
Sgw[6]		2.92	3.27	2.06	2.08	2.18	2.27

[1] Soybean meal was included in diets at the manufacturer particle size and not ground on site.
[2] LO represents an unheated low trypsin and oligosaccharide soybean variety.
[3] LT represents an unheated low trypsin soybean variety.
[4] CB represents an unheated commercial soybean variety.
[5] Geometric mean diameter calculated according to ASAE S319.2.
[6] Geometric mean standard deviation calculated according to ASAE S319.2.

Table I-7. Particle size analyses of complete diets fed to male turkeys during the Starter phases.

	Phase[1]	Starter 1	Starter 2			
	Diet[2]	---	LO	LT	SBM	CB
U. S. Sieve, #	Sieve Size, µm	---------------- % retained on sieve ----------------				
4	4760	0.08	0.05	0.02	0.06	0.01
6	3360	5.38	0.21	0.35	0.43	0.21
8	2380	12.63	1.74	2.46	2.67	1.27
2	1680	17.93	9.34	8.96	12.55	9.44
16	1190	16.85	24.45	23.09	26.42	24.53
20	840	10.84	18.63	18.68	17.74	18.94
30	590	11.77	14.89	14.70	13.54	14.68
40	420	9.54	9.33	9.75	10.15	10.09
50	297	6.55	5.98	5.88	7.28	6.29
70	210	3.49	3.53	3.72	5.04	3.88
100	149	3.05	2.71	3.01	2.76	3.75
140	105	0.69	2.91	2.66	0.75	2.20
200	74	0.47	2.23	2.06	0.23	1.60
270	53	0.28	1.53	1.66	0.05	0.61
Pan	37	0.44	2.46	3.00	0.33	2.52
Dgw, µm[3]		1067	713	700	877	727
Sgw[4]		2.37	2.55	2.61	2.06	2.44

[1] Common Starter 1 pelleted crumble fed to 4 weeks of age followed by mash dietary treatments starting with Starter 2 phase.
[2] Treatments: LO and LT (ground unheated low-trypsin soybeans), SBM (soybean meal), and CB (ground unheated commercial soybean).
[3] Geometric mean diameter calculated according to ASAE S319.2.
[4] Geometric mean standard deviation calculated according to ASAE S319.2.

Table I-8. Particle size analyses of complete diets fed to male turkeys during the Grower phases.

Phase		Grower 1				Grower 2				Grower 3			
Diet[1]		LO	LT	SBM	CB	LO	LT	SBM	CB	LO	LT	SBM	CB
U. S. Sieve, #	Sieve Size, µm	-- % retained on sieve --											
4	4760	0.01	0.02	0.01	0.03	0.01	0.03	0.01	0.05	0.02	0.01	0.01	0.16
6	3360	0.15	0.13	0.20	0.12	0.06	0.13	0.02	0.17	0.16	0.05	0.03	0.07
8	2380	1.25	1.02	1.85	1.59	0.82	0.78	1.25	0.96	0.71	0.03	0.09	0.97
12	1680	5.81	3.44	8.34	5.34	5.14	3.96	8.29	5.33	4.01	1.17	2.27	4.81
16	1190	14.85	12.62	22.24	14.83	17.18	13.04	22.98	15.42	16.75	13.74	20.36	18.01
20	840	18.24	19.61	20.40	18.51	19.62	20.26	20.81	18.94	20.41	20.65	22.15	19.27
30	590	19.04	20.84	15.55	17.79	19.50	20.97	15.55	18.77	22.17	22.15	18.59	20.24
40	420	13.06	13.73	11.19	12.14	13.72	14.36	11.43	13.20	15.45	16.62	14.12	14.69
50	297	7.79	7.93	7.67	7.85	9.03	8.14	8.16	8.76	8.94	10.11	10.35	8.55
70	210	4.65	5.10	4.91	4.98	5.10	4.20	4.73	5.11	5.88	7.13	5.95	5.31
100	149	4.22	3.18	2.78	3.48	7.10	3.09	4.98	8.73	2.78	5.28	2.96	2.44
140	105	3.90	3.41	2.38	4.09	1.05	3.01	0.88	2.26	3.51	2.19	2.05	3.44
200	74	3.47	1.92	1.39	3.04	1.00	2.17	0.44	1.51	0.83	0.40	0.93	1.83
270	53	3.37	4.65	0.93	5.86	0.59	1.44	0.14	0.59	0.03	0.01	0.09	0.16
Pan	37	0.18	2.41	0.16	0.34	0.08	4.42	0.19	0.19	0.04	0.14	0.05	0.05
Dgw, µm[2]		586	539	751	558	662	560	771	623	670	610	679	677
Sgw[3]		2.47	2.56	2.19	2.62	2.09	2.54	2.04	2.21	2.00	1.92	1.96	2.09

[1] Treatments: LO and LT (ground unheated low-trypsin soybeans), SBM (soybean meal), and CB (ground unheated commercial soybean).
[2] Geometric mean diameter calculated according to ASAE S319.2.
[3] Geometric mean standard deviation calculated according to ASAE S319.2.

Table I-9. Particle size analyses of complete diets fed to male turkeys during the Finisher phases.

Phase		Finisher 1				Finisher 2			
Diet[1]		LO	LT	SBM	CB	LO	LT	SBM	CB
U. S. Sieve, #	Sieve Size, µm	% retained on sieve							
4	4760	0.02	0.06	0.19	0.01	0.02	0.01	0.02	0.09
6	3360	0.04	0.12	0.70	0.04	0.14	0.17	0.16	0.16
8	2380	0.54	0.67	2.72	0.93	0.74	0.71	1.52	0.88
12	1680	4.52	4.64	12.78	5.20	4.61	4.75	9.57	5.10
16	1190	22.00	18.87	19.65	20.86	21.41	18.43	29.30	21.94
20	840	20.14	20.52	20.00	20.93	21.55	20.13	20.84	21.00
30	590	18.02	19.57	13.30	18.47	19.34	18.63	14.53	18.12
40	420	12.30	12.94	9.60	12.72	13.56	13.12	10.39	13.21
50	297	7.37	7.89	6.47	7.74	8.85	8.50	6.71	8.60
70	210	4.64	5.94	2.63	4.40	6.04	7.78	3.54	7.76
100	149	2.63	5.63	0.66	2.08	2.86	5.00	1.13	0.87
140	105	3.34	1.02	0.75	2.79	0.51	1.36	1.02	1.40
200	74	2.61	0.63	0.16	2.10	0.20	0.92	0.33	0.52
270	53	1.49	0.49	0.24	1.66	0.02	0.08	0.19	0.15
Pan	37	0.33	1.01	0.14	0.07	0.15	0.40	0.74	0.18
Dgw, µm[2]		666	673	973	691	746	671	875	744
Sgw[3]		2.26	2.13	1.92	2.20	1.90	2.08	1.99	1.96

[1] Treatments: LO and LT (ground unheated low-trypsin soybeans), SBM (soybean meal), and CB (ground unheated commercial soybean).
[2] Geometric mean diameter calculated according to ASAE S319.2.
[3] Geometric mean standard deviation calculated according to ASAE S319.2.

Table I-10. The effects of unheated, low trypsin inhibitor soybeans and litter type on mean body weight (BW) of male turkeys reared to 19 weeks of age.

Effect	N	Age in weeks 4	6	9	12	15	19
Diet[1]		kg body weight per bird					
LO	12	0.91	1.77 [b]	3.86 [b]	7.05 [b]	11.11 [b]	16.75 [b]
LT	12	0.91	1.73 [b]	3.81 [b]	6.85 [b]	10.71 [c]	16.31 [b]
SBM	12	0.89	1.87 [a]	4.37 [a]	8.22 [a]	12.88 [a]	19.14 [a]
CB	12	0.90	1.61 [c]	3.19 [c]	5.53 [c]	8.87 [d]	13.57 [c]
SEM		0.02	0.03	0.06	0.09	0.13	0.19
Litter Type[2]							
MG	24	0.89 [b]	1.75	3.81	6.89	10.92	16.54
PS	24	0.92 [a]	1.74	3.81	6.94	10.87	16.34
SEM		0.01	0.03	0.05	0.07	0.11	0.16
Diet × Litter Type							
LO × MG	6	0.89	1.75	3.90	7.11	11.21	17.06 [b]
LO × PS	6	0.93	1.79	3.82	6.99	11.01	16.44 [bc]
LT × MG	6	0.88	1.71	3.80	6.75	10.59	16.05 [c]
LT × PS	6	0.93	1.75	3.82	6.95	10.82	16.56 [bc]
SBM × MG	6	0.87	1.88	4.34	8.15	12.87	19.27 [a]
SBM × PS	6	0.90	1.86	4.41	8.30	12.90	19.00 [a]
CB × MG	6	0.89	1.65	3.21	5.53	8.99	13.77 [d]
CB × PS	6	0.90	1.57	3.17	5.53	8.75	13.37 [d]
SEM		0.02	0.03	0.07	0.11	0.17	0.24
Source of Variation		*P- values*					
Diet		0.416	<0.001	<0.001	<0.001	<0.001	<0.001
Litter Type		0.022	0.792	0.859	0.383	0.657	0.171
Diet × Litter Type		0.556	0.192	0.614	0.295	0.305	0.045

[1] Dietary treatments began at 4 weeks: LO and LT (ground unheated low-trypsin soybeans), SBM (soybean meal), and CB (ground unheated commercial soybean).

[2] Litter treatments began at placement and consisted of Miscanthus grass (MG) or pine shavings (PS).

[a,b,c,d] Means in a column with different superscripts are significantly different ($P \leq 0.05$).

Table I-11. The effect of unheated, low trypsin inhibitor soybeans and litter type on mean body weight gain (BWG) of male turkeys reared to 19 weeks of age.

Effect	N	Age in weeks 0-4	4-6	6-9	9-12	12-15	15-19	0-19
Diet[1]		---------- kg body weight gain per bird ----------						
LO	12	0.85	0.84 [b]	2.07 [b]	3.19 [b]	4.06 [b]	5.64 [b]	16.68 [b]
LT	12	0.85	0.81 [b]	2.16 [b]	3.06 [b]	3.85 [b]	5.60 [b]	16.24 [b]
SBM	12	0.83	0.97 [a]	2.49 [a]	3.84 [a]	4.67 [a]	6.25 [a]	19.08 [a]
CB	12	0.83	0.68 [c]	1.58 [c]	2.33 [c]	3.34 [c]	4.70 [c]	13.51 [c]
SEM		0.02	0.02	0.08	0.07	0.10	0.16	0.19
Litter Type[2]								
MG	24	0.82 [b]	0.84	2.11	3.08	4.03	5.62	16.48
PS	24	0.85 [a]	0.81	2.05	3.13	3.93	5.47	16.27
SEM		0.01	0.02	0.06	0.06	0.08	0.13	0.16
Diet × Litter Type								
LO × MG	6	0.84	0.84	2.14	3.21	4.10	5.84	16.99 [b]
LO × PS	6	0.87	0.84	2.01	3.12	4.01	5.43	16.37 [bc]
LT × MG	6	0.82	0.81	2.26	2.98	3.84	5.46	15.99 [c]
LT × PS	6	0.87	0.80	2.06	3.13	3.87	5.74	16.50 [bc]
SBM × MG	6	0.81	0.99	2.45	3.80	4.73	6.40	19.21 [a]
SBM × PS	6	0.84	0.95	2.53	3.88	4.60	6.10	18.94 [a]
CB × MG	6	0.83	0.70	1.58	2.32	3.45	4.78	13.71 [d]
CB × PS	6	0.84	0.66	1.59	2.35	3.22	4.61	13.31 [d]
SEM		0.02	0.03	0.09	0.08	0.13	0.20	0.24
Source of Variation		---------- *P- values* ----------						
Diet		0.465	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Litter Type		0.018	0.131	0.302	0.254	0.164	0.219	0.175
Diet × Litter Type		0.618	0.710	0.355	0.586	0.658	0.216	0.045

[1] Dietary treatments began at 4 weeks: LO and LT (ground unheated low-trypsin soybeans), SBM (soybean meal), and CB (ground unheated commercial soybean).

[2] Litter treatments began at placement and consisted of Miscanthus grass (MG) or pine shavings (PS).

[a,b,c,d] Means in a column with different superscripts are significantly different ($P \leq 0.05$).

Table I-12. The effect of unheated, low trypsin inhibitor soybeans and litter type on mean feed intake (FI) of male turkeys reared to 19 weeks of age.

Effect			N	Age in weeks 0-4	4-6	6-9	9-12	12-15	15-19	0-19
Diet[1]				kg feed intake per bird						
LO			12	1.49	1.74	5.52 [a]	8.27 [b]	12.61 [b]	20.32 [a]	49.99 [a]
LT			12	1.47	1.73	5.47 [a]	8.24 [b]	12.04 [b]	20.30 [a]	49.34 [a]
SBM			12	1.48	1.78	5.35 [a]	8.78 [a]	13.64 [a]	19.86 [a]	50.94 [a]
CB			12	1.51	1.62	4.69 [b]	6.73 [c]	10.45 [c]	18.10 [b]	43.15 [b]
SEM				0.05	0.06	0.18	0.17	0.28	0.39	0.72
Litter Type[2]										
		MG	24	1.45	1.69	5.16	7.87	12.20	19.61	48.04
		PS	24	1.52	1.74	5.35	8.14	12.17	19.68	48.68
SEM				0.04	0.05	0.15	0.15	0.24	0.33	0.59
Diet × Litter Type										
LO	×	MG	6	1.42	1.72	5.55	8.18	12.61	20.36	49.88
LO	×	PS	6	1.56	1.75	5.48	8.36	12.62	20.28	50.10
LT	×	MG	6	1.43	1.67	5.36	8.06	11.95	19.86	48.37
LT	×	PS	6	1.51	1.79	5.57	8.43	12.13	20.73	50.31
SBM	×	MG	6	1.43	1.77	5.29	5.55	13.61	19.86	50.55
SBM	×	PS	6	1.52	1.79	5.42	9.01	13.67	19.87	51.33
CB	×	MG	6	1.52	1.63	4.44	6.70	10.63	18.35	43.34
CB	×	PS	6	1.49	1.62	4.93	6.74	10.27	17.86	42.97
SEM				0.06	0.09	0.24	0.22	0.36	0.51	0.90
Source of Variation				*P- values*						
Diet				0.892	0.101	<0.001	<0.001	<0.001	<0.001	<0.001
Litter Type				0.061	0.332	0.174	0.051	0.893	0.798	0.231
Diet × Litter Type				0.473	0.783	0.556	0.659	0.827	0.437	0.466

[1] Dietary treatments began at 4 weeks: LO and LT (ground unheated low-trypsin soybeans), SBM (soybean meal), and CB (ground unheated commercial soybean).

[2] Litter treatments began at day 0 of placement and consisted of Miscanthus grass (MG) or pine shavings (PS).

[a,b,c] Means in a column with different superscripts are significantly different ($P \leq 0.05$).

Table I-13. The effect of unheated, low trypsin inhibitor soybeans and litter type on mean feed conversion ratio (FCR)[1] of male turkeys reared to 19 weeks of age.

Effect	N	Age in weeks 0-4	4-6	6-9	9-12	12-15	15-19	0-19
Diet[2]		kg feed intake: kg body weight gain						
LO	12	1.63	2.16 ^{bc}	2.64 ^{a}	2.51 ^{a}	3.15 ^{a}	3.71 ^{a}	3.02 ^{b}
LT	12	1.61	2.31 ^{ab}	2.61 ^{a}	2.61 ^{a}	3.14 ^{a}	3.71 ^{a}	3.01 ^{b}
SBM	12	1.65	1.93 ^{c}	2.14 ^{b}	2.21 ^{b}	2.88 ^{b}	3.34 ^{b}	2.65 ^{c}
CB	12	1.68	2.46 ^{a}	2.90 ^{a}	2.66 ^{a}	3.15 ^{a}	3.90 ^{a}	3.19 ^{a}
SEM		0.06	0.08	0.10	0.08	0.08	0.10	0.03
Litter Type[3]								
MG	24	1.63	2.13 ^{b}	2.51	2.51	3.03	3.64	2.88
PS	24	1.65	2.30 ^{a}	2.64	2.48	3.13	3.70	2.94
SEM		0.05	0.07	0.09	0.07	0.07	0.08	0.03
Diet × Litter Type								
LO × MG	6	1.58	2.13	2.59	2.49	3.10	3.61	2.98
LO × PS	6	1.67	2.19	2.70	2.53	3.20	3.82	3.06
LT × MG	6	1.61	2.13	2.53	2.62	3.13	3.72	2.99
LT × PS	6	1.62	2.49	2.69	2.59	3.15	3.70	3.03
SBM × MG	6	1.63	1.89	2.16	2.22	2.84	3.24	2.61
SBM × PS	6	1.67	1.97	2.13	2.20	2.92	3.44	2.68
CB × MG	6	1.71	2.39	2.74	2.72	3.06	3.97	3.19
CB × PS	6	1.66	2.54	3.05	2.60	3.24	3.83	3.20
SEM		0.07	0.11	0.13	0.11	0.10	0.13	0.05
Source of Variation		*P- values*						
Diet		0.654	<0.001	<0.001	<0.001	0.005	<0.001	<0.001
Litter Type		0.604	0.013	0.091	0.642	0.108	0.395	0.073
Diet × Litter Type		0.781	0.336	0.494	0.875	0.820	0.293	0.746

[1] Feed conversion ratio defined as pen FI divided by pen BWG plus any mortality or culls within the period.
[2] Dietary treatments began at 4 weeks: LO and LT (ground unheated low-trypsin soybeans), SBM (soybean meal), and CB (unheated commercial soybean).
[3] Litter treatments began at placement and consisted of Miscanthus grass (MG) or pine shavings (PS).
[a,b,c] Means in a column with different superscripts are significantly different ($P \leq 0.05$).

MANUSCRIPT II:

The effect of unheated, low-trypsin inhibitor soybeans on the digestibility and growth performance of tom turkeys reared to 14 and 28 days of age.

ABSTRACT: Two studies were conducted to evaluate the effect of poult age on the optimal inclusion rate of a novel low trypsin inhibitor soybean in turkey starter diets. Poults were either raised from hatch to 2 weeks of age (Trial 1) or from 2 to 4 weeks of age (Trial 2). Male Hybrid Converter turkey poults (288) were randomly assigned in groups of 6 (Trial 1) and 5 (Trial 2) to 48 Alternative Design battery cages. Poults were provided ad libitum access to water and dietary treatments. All finished feeds were iso-caloric and iso-nitrogenous with treatments defined by the inclusion of solvent extracted soybean meal (SBM), the unheated low trypsin inhibitor (LTI) soybean at 20, 30, or 40%, or the conventional unheated soybean (CB) at 20 or 40%. Poults and feeders were weighed weekly to determine body weight (BW) and feed intake (FI) to calculate the feed conversion ratio (FCR). At day 14 and day 28 (Trial 1 and Trial 2, respectively) excreta were collected and pooled by pen for the determination of apparent metabolizable energy (AMEn) and apparent lipid digestibility (ALD). At the conclusion of each trial, poults were euthanized via cervical dislocation and their pancreas excised. Pancreas weights were expressed relative to poult body weight (PRW). Additionally, at the conclusion of Trial 2, ileal contents were collected and pooled by pen for the determination of apparent ileal digestibility (AID) of various amino acids. Overall, increased inclusion of the LTI bean from 0 to 40% linearly decreased poult BW and FI from 0 to 14 days, while increasing the FCR and P_{RW} when compared to SBM. The depressed performance and enlarged pancreases of the poults indicated that the TI was still present at levels sufficient to affect the protein digestibility. Performance was improved in trial 2 with the older poults. At day 21, there was only a tendency for reduction in BW with linearly decreasing FI, though no significant difference in FCR. By day 28, LTI diets

remained comparable to SBM in BW, FI, and FCR, though P_{RW} remained greater in relation to increasing LTI inclusion. P_{RW} increased in proportion to the trypsin inhibitor content of the feed. Total apparent amino acid digestibility decreased as feed trypsin inhibitor content increased. These results would indicate that as the poult approached 21 days of age, it was more able to utilize the nutrients in the LTI diets for growth. When comparing CB to SBM or LTI diets, performance was consistently reduced when feeding CB.

Keywords: turkey, soybean, trypsin inhibitor, poult age, anti-nutritional factors

INTRODUCTION

Soybeans, typically fed as a defatted meal as a result of oil extraction, are utilized world-wide as a protein source in animal nutrition. It is estimated that over 55% of the US supply of soybeans are consumed by the poultry industry alone (Soystats, 2015). Unlike other major ingredients such as corn, current soybean varieties require extensive processing prior to use in animal feeds (Gillman et al., 2015). Processing involves conditioning, flaking, solvent extraction of fat and toasting of the high protein meal. The conditioning and flaking process releases oils for fat extraction, while toasting with moist heat at high temperature denatures the anti-nutritional factors (ANF) associated with the defatted raw soybeans. The resulting low-fat product is a 48% protein soybean meal for use in poultry feeds.

The added cost associated with the increased processing of the residual meal, ie toasting, is considered a necessity. Failure to destroy heat liable ANFs like protease inhibitors such as trypsin inhibitor (TI) lead to reduced protein digestion, resulting in poor growth performance (Rackis, 1965). McIsaac and others (2005) demonstrated that it was possible to feed a full-fat ground soybean to turkey hens without diminishing performance if that soybean was roasted prior. This confirmed that soybeans could successfully be utilized in turkey diets once the ANF compounds were eliminated or reduced, in this instance by heat.

Gene selection offers an alternative method to soybean heat treatment for reducing the ANF level in soybeans. Improved cultivars are being developed to contain naturally low levels of the ANFs commonly associated with poor growth performance, with much of the focus surrounding the reduction of TI protease. These novel lines could potentially eliminate the necessity of further processing and economize the use of soybeans in animal feeds (Kumar et al, 2013). Additionally, the retained natural oil content in the bean increases the energy value when compared to the traditional de-fatted soybean meal. Therefore, depending on the price of other protein and fat sources, the unprocessed, full-fat soybeans could become an economic alternative in least-cost formulation (Swick, 1996).

Attempts to feed unheated, low TI soybeans to poultry have resulted in varied success, with bean inclusion level and bird age among the main sources of debate. Han and Parson (1991) observed no significant difference in weight gain or FCR at day 19 between broilers receiving 25 and 50% soy protein from a low TI bean and those receiving 100% soy protein from commercial solvent extracted soybean meal. Further increases, however, of the low TI bean to 75 or 100% of the soy protein repressed growth performance, which was likely a result of chick age. It is believed that young birds may be especially susceptible to ANFs in soybeans as their gut function continues to develop (Mian and Garlich, 1995; Kroghdal and Sell, 1988).

Thus, the objective of this study was to determine the effect of feeding increasing levels of unheated, low TI soybeans on the digestibility and growth performance of young turkey poults at 14 and 28 days of age.

MATERIALS AND METHODS

Bird Husbandry

Trial 1. Male Hybrid Converter[4] turkey poults were sourced from a single commercial hatch[5] for use in both Trial 1 and Trial 2. Upon arrival at the research facility (North Carolina State University, Raleigh, NC), 288 poults were randomly selected and distributed into groups of 6 for placement. Poults were weighed and assigned to 48 Alternative Design (Siloam Springs, AR) battery cages. Cages were randomly allotted to 1 of 6 dietary treatments resulting in 8 replicates per treatment. Feed and water were consumed *ad libitum*. Batteries were located in a

[4] Hendrix Genetics, Ontario, Canada
[5] Sleepy Creek Hatchery, Goldsboro, NC

climate-controlled room and equipped with dual nipple water drinkers, feeding trough, supplemental heater, and collection pan located beneath the floor of each cage. Artificial light was provided for 24 hours for the first 7 days; and then adjusted to provide 23 hours of light and 1 hour of dark for the remainder of the study. Heat was provided for bird comfort and adjusted based on bird behaviour. Temperatures were monitored twice a day to yield averages of 32-35^0 C for the first 7 days followed by 26-34^0 C for the next 7 days.

Trial 2. Male Hybrid Converter turkey poults remaining after the placement of Trial 1 were randomly placed in litter floor pens of a curtain-sided facility and fed a common crumbled starter diet until 2 weeks of age when Trial 2 began. All floor pens were equipped with a tube feeder, bell drinker[6] and supplemental heat lamp, which was adjusted based on ambient temperature and bird comfort. House temperature was maintained at 32^0 C for the first 3 days and then gradually lowered by 0.5^0 C per day until outside ambient temperature was reached. Incandescent light was provided during the first 72 hours followed by natural lighting. At day 14, 240 poults were transferred, weighed and randomly assigned to 48 Alternative Design (Siloam Springs, AR) battery cages. Battery cages, management, and dietary treatments remained the same as in Trial 1; however, poult numbers were reduced to 5 poults per pen to ensure adequate feeder space at 4 weeks of age. Poults consumed feed and water *ad libitum*.

Dietary Treatments

All diets were formulated to meet or exceed the breeder recommendations for young toms (Hybrid). Soy oil was used to maintain similar caloric density of diets across treatments, with decreasing levels needed as inclusion of the unheated soybeans increased. To create dietary treatments solvent extract soybean meal was replaced with unheated soybeans on a protein basis.

[6] Plasson Livestock, Menashe, Israel

An assumed energy value of 3300 Kcal/kg was designated for the unheated soybeans based on the NRC value given to heat processed, full-fat soybeans (NRC, 1994). Maximum inclusion rate of the unheated soybean was limited to 40% based on the natural oil content. Soybean varieties e3311 and e37×812 (eMerge Genetics, Des Moines, IA) were chosen for testing. Bean e3311 contained naturally low levels of Kunitz TI (10,300 TIU/g) and hereafter is referred to as LTI. Bean e37×812, referred to as CB, was a conventional commodity-type soybean containing typical levels of ANFs like TI (29,800 TIU/g). Six dietary treatments were tested: 20, 30, or 40% unheated LTI, 20 or 40% unheated CB, and a diet containing only commercial solvent extracted soybean meal (SBM), which served as a control (Table 1). Dietary treatments remained the same for both Trial 1 and Trial 2 and were fed from day 1 to 14 and day 14 to 28, respectively. A common crumbled starter diet was fed from hatch until 2 weeks of age in Trial 2, while poults in Trial 1 immediately began mash dietary treatments. Water and dietary treatments were consumed *ad libitum*. Cellulose[7] was included to balance nutrient density across treatments, while diatomaceous earth or silicone dioxide[8] was added as an indigestible marker at 1.7% of the diet.

Feed Manufacturing

Feed was manufactured in accordance with Current Good Manufacturing Practices (CGMPs) at the North Carolina State University Educational Feed Mill (Raleigh, NC). Corn and unheated soybeans (LTI and CB) were ground using a double high roller mill (RMS, Harrisburg, SD) furnished with 305 mm diameter rolls measuring 406 mm long. Equipped with two 15-hp motors, the approximate roll differentials were calculated as 1.5:1 on the top and 2:1 on the bottom. The roll gap or distance between roll sets was adjusted manually for whole corn and whole beans to yield finished diets with an estimated particle size of 700 microns. Whole corn

[7] Solka-Floc®, International Fiber Corporation, North Tonawanda, NY
[8] Celite®, World Minerals, Inc., Santa Barabara, CA

was ground using a 0.64 mm gap between the top set of rolls and 0.56 mm gap between the bottom set, while the unheated LTI and CB soybeans were both ground with 0.20 mm and 0.15 mm gaps, respectively. Diets were mixed in a 3.6 m^3 counterpoise mixer (Hayes and Stolz, Fort Worth, TX) for the standard time of 270 sec. Dry ingredients were allowed to mix for 90 sec prior to the addition of the liquid soy oil.

Live Performance

Individual poult and feeder weights were collected at day 7 and 14 or day 14 and 28 (Trial 1 and Trial 2, respectively) to acquire body weight (BW), body weight gain (BWG), and feed intake (FI). Mortalities were recorded daily. Feed conversion ratio (FCR) was calculated for each weigh period and adjusted based on mortalities by dividing the pen FI by the pen BWG plus the BW of any mortalities from the period.

Sample Collection and Chemical Analysis

Feed. Representative feed samples were obtained for each dietary treatment and analyzed by the University of Missouri Agricultural Experiment Station (Columbia, MO) for proximate analysis, trypsin inhibitor, urease index, and complete amino acid profile. A subsample of all dietary treatments was taken and stored at -20° C until processed and analyzed for digestibility measurements as outlined below.

The particle size of complete diets were determined according to dry sieving method ASAE S319.2 (ASAE, 1995). Analysis included 13 sieves, the addition of sieve agitators and 0.5 g of dispersing agent[9] per 100 g of sample with a 10 min run time. Sieve sizes ranged from 3350 to 53 µm.

[9] Silicon Dioxide, model SSA-58, Gilson, Lewis Center, OH

Bulk density of dietary treatments was calculated using the method described by Clementson and others (2010). Briefly, sample was poured into a funnel-shaped hopper centered over a 1 L measuring cup. The sample was allowed to freely flow into the measuring cup and excess material was systematically leveled off. The filled measuring cup was then weighed and the bulk density calculated by dividing the mass of the sample in the measuring cup by the volume of the measuring cup.

To estimate the fat availability of the dietary treatments, 5 g of sample was placed in a cellulose extraction thimble measuring 20 mm in diameter and 50 mm long. The sample was rinsed with 25 ml of petroleum ether for 6 consecutive washes. The sample rested for 30 min between each wash to allow the ether to completely drain from the thimble and the sample temperature to return to ambient. The sample was stirred in the thimble prior to washes 3 and 6. After each wash, the eluate of ether and fat was collected into a beaker of known weight and placed in a forced-air drying oven at 105° C for 1 hr. After evaporating off the ether, the remaining oil residue was quantified by weighing the beaker and subtracting the empty weight. The weight of the recovered fat was then divided by the initial sample weight and multiplied by 100. Results were cumulative, with Wash 6 comprised of the total fat recovery of the sample. The fat recovery was determined on as-fed samples and samples that had been ground through a 0.5 mm screen.

Excreta. At 2 and 4 weeks of age (Trial 1 and 2, respectively) fresh excreta samples were collected once per day for 3 days from a pan located beneath each battery cage, with care taken to avoid feed or feather contaminants. Composited samples were frozen and stored at -20° C until processed. Homogenized pen samples were lyophilized (Labconco, Kansas City, MO) and finely ground to pass through a 0.5 mm screen prior to analysis. Excreta and feed samples were then analyzed for gross energy, nitrogen, and fat content. Gross energy content was determined on an adiabatic bomb calorimeter (IKA Works, Inc., Wilmington, NC) with benzoic acid as the calibration standard, while nitrogen content was measured via combustion analysis (LECO Corporation, St. Joseph, MI) with EDTA as the calibration standard. Crude fat content was determined using petroleum ether and Soxhlet extraction. Lastly, all samples were analyzed for recovery of the indigestible marker based on the acid-insoluble ash procedure described in Vogtmann et al. (1975) for the determination of apparent metabolizations.

Pancreas Collection. At the conclusions of Trials 1 and 2 on day 14 or 28, respectively, all birds were euthanized via cervical dislocation, weighed and their pancreas excised. Pancreases were carefully removed from the duodenal loop and individually weighed. Resulting weights were then expressed as a percentage of the poult's body weight.

Ileal Digesta Collection. On day 28 of Trial 2, all poults were euthanized and their small intestine excised. Ileal contents, beginning 1 cm posterior to the Meckel's diverticulum and ending 1 cm prior to the ileal-cecal junction, were collected and pooled by pen. Composite samples were stored at -20°C prior to lyophilization (Labconco, Kansas City, MO). Samples were finely ground to pass through a 0.5 mm screen and sent to the University of Missouri Agricultural Experiment Station (Columbia, MO) for determination of amino acid concentrations. Additionally, the pooled ileal contents were analyzed for indigestible marker recovery according to Vogtmann et al. (1975), the same method utilized for feed and excreta samples.

Calculations

Retained nitrogen (N_{Ret}), nitrogen-corrected apparent metabolizable energy (AME_n), apparent lipid digestibility (ALD), and apparent ileal digestibility of amino acids (AID) were calculated using the following equations as described by Kong and Adeola (2013):

$$N_{Ret} = N_{feed} - \frac{(N_{excreta} * AiA_{feed})}{AiA_{excreta}}$$

$$AME_n = GE_{feed} - \left(\frac{GE_{excreta} * AiA_{feed}}{AiA_{excreta}}\right) - (8.22 * N_{Ret})$$

$$ALD(\%) = 100 - \left[\left(\frac{AIA_{feed} * C_{excreta}}{AIA_{excreta} * C_{feed}}\right) * 100\right]$$

$$AID(\%) = 100 - \left[\left(\frac{AiA_{feed} * AA_{sample}}{AiA_{sample} * AA_{feed}}\right) * 100\right]$$

where AiA refers to the concentration of the index compound (Celite®) in the feed, excreta, or ileal digesta sample; N refers to the nitrogen content of the feed or excreta; GE refers to the gross energy of the feed or excreta; C refers to the lipid concentration of the feed or excreta; and AA refers to the concentration of an amino acid in the feed or ileal digesta sample.

Animal Ethics

All handling procedures and euthanasia protocols were carried out in accordance with the guidelines approved by the Institutional Animal Care and Use Committee of North Carolina State University.

Statistical Analysis

All data were analysed using the GLIMMIX procedure in SAS version 9.4 (SAS Institute Inc., Cary, NC). Treatments were arranged in a randomized block design and included: SBM, 20% LTI, 30% LTI, 40% LTI, 20% CB, and 40% CB. Pre-planned linear and quadratic contrasts were performed to further characterize interactions as a result of increasing LTI or CB inclusion, with additional and contrast to evaluate the interaction between soybean type (LTI and CB) and level (20% and 40%). A final orthogonal contrast was used to test 20% and 40% LTI versus 20% and 40% CB inclusion. Treatment effects were considered significant at $P \leq 0.05$ and marginally significant at $0.10 \leq P \geq 0.05$. The experimental unit was defined as the pen (n=8 for Trials 1 and 2) for all parameters except relative pancreas weight, where bird (n=48 and n=40 for Trials 1 and 2, respectively) served as the experimental unit.

RESULTS

Dietary Treatments

Complete diets were analyzed for nutrient composition, particle size and bulk density with results reported in Table II-2. Diets remained similar in crude protein and crude fat across all treatments, with comparable physical attributes such as particle size and bulk density. As expected, TI increased proportionally to the inclusion level of both the unheated soybeans (LTI and CB); however, even when at a higher inclusion level, diets containing the LTI soybean with an estimated 10,300 TIU/g had lower dietary TI when compared to either diet containing the CB soybean with an estimated 29,800 TIU/g. Urease activity followed a similar trend and increased as the inclusion level of the unheated soybeans rose in the diet. Though, like TI, the urease activity was lower in the LTI diets regardless of inclusion level when compared to the CB diets. As mentioned, analyzed crude fat was similar among dietary treatments, however, recovered fat from petroleum ether washes may indicate that there is a difference in the availability of this fat (Table II-3). On an as-fed basis, the SBM diet had the greatest initial fat recovery after Wash 1. The next greatest recoveries were observed with 20% inclusion of either unheated soybean (LTI or CB), followed by the 30% LTI diet. The lowest initial fat recoveries were diets containing either 40% LTI or CB. Grinding the samples prior to rinsing with ether yielded numerically greater fat recovery both after the initial Wash 1 and cumulatively after Wash 6.

Live Performance

Trial 1. There was no significant difference in poult BW among dietary treatments at placement (P = 0.230; Table 4). At day 7, there was no interaction between soybean type and inclusion level (P = 0.309); however, poult BW linearly decreased as LTI and CB inclusion increased in their respective diets (P = 0.002 and P < 0.001, respectively). At day 14, there was a tendency for an interaction between soybean type and inclusion level (P = 0.061), with a greater reduction in poult BW when consuming 40% CB diets compared to 40% LTI diets. Additionally, as at day 7, poult BW linearly decreased as LTI and CB inclusion increased in their respective diets (P = 0.009 and P < 0.001, respectively). Poults consuming LTI diets were heavier than those on CB diets regardless of inclusion level (P < 0.001). These observations were echoed in the poult BWG results (Table II-5), however, there was no interaction between soybean type and inclusion level at any point. Initial FI data (Table II-6) on day 7 again showed a linear reduction as LTI and CB inclusion were increased (P = 0.024 and P < 0.001, respectively). By day 14, this linear reduction in intake resulting from LTI inclusion disappeared, though CB continued to repress intake (P = 0.113 and P < 0.001, respectively). The improvement in period intake of the LTI diets was not substantial enough to affect cumulative trial FI, resulting in observed linear FI reductions for both LTI and CB diets from day 0 to 14 (P = 0.016 and P < 0.001, respectively). There was an interaction between soybean type and inclusion level at day 14 (P = 0.04) and overall from day 0 to 14 (P = 0.014), with LTI diets having a lower FCR at 40% compared to CB diets at the same inclusion level. While there was no linear impact (P = 0.264; Table II-7) on FCR at day 7, at day 14, increasing LTI inclusion tended to increase the FCR (P = 0.061), resulting in an overall linear increase in FCR from day 0 to 14 (*P* = 0.020). Increasing CB inclusion linearly increased FCR at both 7 and 14 days (*P* < 0.001 and *P* < 0.001, respectively), resulting in an increased cumulative FCR from day 0 to 14 (*P* < 0.001). The LTI diets continued to outperform the CB diets regardless of period and inclusion level.

Trial 2. There was no significant difference in poult BW among dietary treatments at the start of the trial on day 14 (*P* = 0.632; Table II-4). At day 21, increasing LTI inclusion tended to reduce BW (*P* =0.056); however, by day 28, BW was similar for SBM and LTI diets (*P* = 0.291). BW was linearly decreased as CB level in the diet increased regardless of period. The LTI diets had greater BW than CB diets regardless of inclusion level at day 21 and 28 (*P* < 0.001 and P= 0.002, respectively). These observations were echoed in the cumulative poult BWG results (Table II-5); however, there was only a marginal reduction in BWG from day 21 to 28 when feeding CB compared to LTI diets (*P* = 0.055). While increased LTI inclusion repressed FI from d 14 to 21 (*P* = 0.010; Table II-6), FI increased from day 14 to 28, remaining comparable to SBM for during both the 7 day period and cumulatively from 14 to 28 days (*P* = 0.347 and *P* = 0.104, respectively). Increased inclusion of CB soybeans linearly decreased period and cumulative FI (*P* < 0.001, *P* < 0.001 and *P* < 0.001 for 21, 28, and 14 to 28 d, respectively). An interaction between soybean type and level was observed at day 28 as a result of the reduced FI of the 40% CB bean diet in comparison to the 40% LTI diet (*P* = 0.031). The LTI diets had significantly higher 21, 28, and cumulative 14 to 28 day FI compared to CB diets regardless of inclusion level (*P* = 0.037, *P* = 0.020 and *P* = 0.004, respectively). FCR remained unaffected by increased LTI inclusion at 21, 28, and 14 to 28 days (*P* = 0.223, *P* = 0.169 and *P* = 0.633, respectively). Increasing CB inclusion linearly increased FCR at day 21 (*P* < 0.001), but remained similar to SBM at day 28 and cumulatively from day 14 to 28 (*P* = 0.124 and *P* =

0.431, respectively). This was again observed when comparing CB to LTI diets, where like SBM, LTI had lower FCR at day 21 ($P < 0.001$), but not at day 28 or cumulatively ($P = 0.726$ and $P = 0.273$, respectively).

Relative Pancreas Weight

Trial 1. As LTI increased in the diet there was a linear increase in P_{RW} at day 14 ($P < 0.001$; Table II-8), while increased CB resulted in a quadratic response ($P < 0.001$). P_{RW} was increased at a greater rate with CB inclusion than LTI (interaction, $P < 0.001$). LTI diets yielded lower P_{RW} when compared to CB diets regardless of inclusion level ($P < 0.001$).

Trial 2. Dietary treatments affected day 28 P_{RW} in a similar manner as in Trial 1. A linear increase in P_{RW} was observed as LTI increased ($P < 0.001$) and a quadratic response was detected as CB increased ($P = 0.007$). P_{RW} was increased at a greater rate with CB inclusion than LTI (interaction, $P < 0.001$). Regardless of inclusion level, LTI diets had lower P_{RW} when compared to CB diets ($P < 0.001$).

Retained Nitrogen

Trial 1. At day 14, the 30% LTI diet had the greatest N_{RET} resulting in a quadratic effect as LTI increased ($P = 0.019$; Table 8). N_{RET} also responded quadratically to increasing levels of CB in the diet ($P = 0.027$). There was an interaction between soybean type and inclusion level on N_{RET} ($P < 0.001$), where increasing bean inclusion from 20% to 40% increased N_{RET} in the LTI diets, but reduced N_{RET} in the CB diets. On average, however, LTI diets had greater N_{RET} compared to CB diets ($P < 0.001$).

Trial 2. At day 28, there was no linear or quadratic response observed for increasing LTI inclusion ($P = 0.852$ and $P = 0.950$, respectively). The greatest N_{RET} was again observed with the 30% LTI diet followed by the SBM control. N_{RET} was linearly decreased with increasing levels of CB in the diet ($P < 0.001$). There was an interaction between soybean type and inclusion level on N_{RET} ($P < 0.001$), where increasing bean inclusion from 20% to 40% increased N_{RET} in the LTI diets, but reduced N_{RET} in the CB diets.

Apparent Metabolizable Energy

Trial 1. At day 14, there was no linear or quadratic effect on AMEn as LTI inclusion was increased ($P = 0.775$ and $P = 0.563$, respectively). The 30% LTI diet had the largest AMEn at day 14, followed by the SBM and 40% LTI diets (Table 8). Increasing inclusion of CB linearly decreased the AMEn ($P < 0.001$). There was an interaction between soybean type and inclusion level on AMEn ($P < 0.001$), where increasing bean inclusion from 20% to 40% increased AMEn in the LTI diets, but reduced AMEn in the CB diets.

Trial 2. At day 28, increased LTI inclusion linearly reduced AMEn ($P < 0.001$); however, numerically, the greatest AMEn was again observed in the 30% LTI diet. Increasing CB inclusion also linearly reduced AMEn ($P < 0.001$). There was an interaction between soybean type and inclusion level on AMEn ($P < 0.001$), where increasing bean inclusion from 20% to 40% increased AMEn in the LTI diets, but reduced AMEn in the CB diets.

Apparent Lipid Digestibility

Trial 1. At day 14, there was an interaction between soybean type and inclusion level on ALD ($P = 0.021$; Table 8). Increasing CB bean inclusion from 20% to 40% reduced fat digestibility at a greater rate than when LTI bean inclusion was increased. Increasing either LTI or CB inclusion linearly reduced ALD in their respective diets ($P < 0.001$ and $P < 0.001$, respectively).

Trial 2. There was no interaction between soybean type and inclusion level on ALD at day 28 ($P = 0.473$); however, increasing either LTI or CB inclusion linearly reduced ALD in their respective diets ($P < 0.001$ and $P < 0.001$, respectively).

Apparent Ileal Digestibility of Amino Acids

Trial 2. Amino acid digestibility was determined based on the ileal contents of the poults at day 28 with indispensable amino acid results reported in Table II-9 and dispensable amino acid results in Table II-10. Regardless of the amino acid, the overall trend in digestibility was similar. The SBM diet generally had the best digestibility, followed by the novel LTI bean diets, and then the CB diets. Often times, 20 and 30% LTI diets had similar digestibility to that of SBM, while the 40% had more fluctuation depending on the specific amino acid. As a result, increasing the LTI in the diets typically reduced the AID linearly. Increased CB inclusion also typically led to a linear decrease in AID, with the lower 20% inclusion level improving digestibility over that of the 40%.

DISCUSSION

Previous research with broilers has shown that while unheated soybeans low in TI lead to better growth performance compared to unheated commercial soybean cultivars, they continue to be nutritionally inferior to conventional soybean meal (Chohan et al., 1993; Han et al., 1991; Herkelman et al., 1993; Palacios et al., 2004; Yen et al., 1974). In contrast, however, the results of these trials agree with a hypothesis by Batal and Parsons (2003) that when ANF levels are not excessive, birds, especially as they age, may be better able to adapt and utilize nutrients. Thus, garnering similar growth performance to that of a SBM based diet when fed at an older age.

Comparing the differences in LTI and SBM in these trials, increased inclusion of the LTI bean from 0 to 40% linearly decreased poult BW and FI from 0 to 14 days, while increasing the

FCR and P_{RW}. The depressed performance and enlarged pancreases of the poults indicate that the TI was still present at levels sufficient to affect the protein digestibility. Dietary TI levels were measured at 1.1, 3.1, 5.2, and 5.1 mg TIU per g of diet for the SBM, 20%, 30% and 40% LTI diets, respectively. Mian and Garlich (1995) reported that turkeys were able to tolerate 2.5 to 3.2 mg of TIU per g of diet for the first 3 weeks, however, their findings were based on TI levels created through heating of soy flakes at 107 °C for between 5 to 25 min. It is possible that during the heating process to achieve the TI level gradients, other ANFs, such as heat-liable lectin, may have also been reduced and further improved diet digestibility. Thus, poult TI tolerance levels may be lower when feeding unheated soybeans than those described by Mian and Garlich (1995); or, alternatively, the levels of other ANFs, besides TI, may have remained too great in the LTI bean and inhibited growth. Kakade and other (1974) concluded that TI account for only 40% of the growth inhibition of raw soybeans. Herkelman and others (1993) observed similar growth performance to trial 1 in 16-day old broilers consuming 33% unheated LTI soybeans, which had repressed BW gain and FI with an increased FCR and P_{RW} when compared to a SBM diet. Dietary TI levels, however, were not reported, so the authors can only conclude that, similar to our trials, levels of ANFs were too great in the unheated soybeans for the young poults to digest.

Performance was improved in trial 2 with the older poults. At day 21, there was only a tendency for reduction in BW with linearly decreasing FI, though no detectable difference in FCR. By day 28, LTI diets remained comparable to SBM in BW, FI, and FCR, though P_{RW} remained greater in relation to increasing LTI inclusion. These results would indicate that as the poult approached 21 days of age, it was more able to utilize the nutrients in the LTI diets for growth. This is likely due to the maturing of the poult's digestive system, which according to

Kroghdahl and Sell (1988) does not have stable intestinal secretions prior to day 21 or pancreatic activity until day 28. The hypersecretion by the pancreas, as shown by increased P_{RW}, denotes the poult's response to increased levels of TI and its attempt at neutralizing the inhibitors (Chernick et al., 1948; Mian and Garlich, 1995). Furthermore, the older birds in trial 2 exhibited lower P_{RW} in response to the diets than in trial 1, which may signify that as the poult ages, it has an increased ability to digest diets with increased levels of ANFs like TI. This would support the conclusions of several other studies, where the detrimental effects associated with high TI diets were diminished as bird age increased (Baker, 2000; Batal and Parsons, 2003; Leeson et al., 1987; MacIsaac et al., 2005).

When comparing CB to SBM or LTI diets, performance was consistently reduced when feeding CB. The only exception was at day 28 where FCR remained similar among all treatments. This was likely due to increased dietary TI levels in the CB diets, measuring 6.9 and 9.4 mg TIU per g of diet for the 20 and 40% inclusion level, respectively, which is a 33 and 81% increase in TI when compared to the same 20 and 40% inclusion of the LTI bean. Poults consuming 20 and 40% CB diets gained 18.4 and 32.1% less BW at day 14 and 5.5 and 13.6% less at day 28 as compared to birds consuming the LTI bean at the same levels. The reduced performance of CB beans compared to LTI beans has been well documented and agrees with the findings of these studies (Han et al., 1991; Hosatani et al., 2016; Palacios et al., 2004; Perez-Maldonado et al., 2003; Yen et al., 1937).

During both trial 1 and 2, the 30% LTI diet had the greatest dietary AMEn, which was unexpected and a deviation from the overall pattern shown in the live performance data. Duplicate samples were analysed to ensure accuracy of the assay. Remaining AMEn results concur with the poult performance in trial 1, though AMEn values associated with the 20% LTI diet in trial 2 appear to be low when compared to other diets like 20% CB, which had greater AMEn.

The deviation from the expected AMEn values for the diets could be linked to differences in fat digestibility. Fat digestibility decreased linearly with increasing levels of both LTI and CB. This was likely caused by decreasing additions of supplemental oil as unheated soybean inclusion increased, with 7.15% added to SBM, representing the greatest, and decreasing to 0.6% in the 40% LTI diet and none in the 40% CB diet, representing the lowest. Increased levels of supplemental oil in the finished feed likely slowed gut passage rate and was ultimately easier for the poults to access during digestion when compared to the natural oil in the unheated soybeans. Sequential petroleum ether washes of the diets to recover fat showed increased lipid recovery during the first wash as supplemental fat was increased, providing further insight into the access of the poult to fat in the diets. It was likely that roller milling the unheated soybeans in these trials was not sufficient enough to rupture all of the oil glands, resulting in decreased availability of both the oil and amino acids from protein. The results of milling would be more prevalent in the higher inclusion levels without the supplemental oil. Herkelman and others (1993) also suggested that there is reduced utilization of fat by chicks in soybean diets as compared to meal and oil diets, which, again, was attributed to unruptured oil cells.

In addition to increased ANFs, Parsons and others (1991) observed a decrease in amino acid availability with unheated soybeans, which they held partially responsible for the poor protein utilization in chicks. In their study, unheated LTI soybeans had generally better amino acid bioavailability when compared to CB soybeans, though both were lower than SBM. They attributed these differences to increased loss of endogenous protein due to increased pancreatic enzyme secretions and decreased dietary digestibility of amino acids due to reduced proteolytic activity as described by Liener (1977). These findings concur with those observed in these trials, in which the greatest total amino acid digestibility was observed in the SBM diet and was linearly decreased as either LTI or CB inclusion increased, with CB inclusion having the greatest impact on reduced amino acid digestibility. Even with the difference in amino acid availability, the LTI diet remained comparable in performance to the SBM at day 28.

CONCLUSIONS AND APPLICATIONS

1. Under the constraints of this trial, turkeys 4 weeks of age can tolerate soybean TI concentrations of 5.1 mg TIU per g of diet when consuming a low TI soybean cultivar.
2. Turkey poults have a greater tolerance for TI as they mature.
3. Roller milling unheated whole soybeans for use in turkey diets may not be sufficient enough to rupture all of the oil glands, reducing the availability of fat in the diet.

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Table II-1. Composition of mash dietary treatments with varying levels of unheated soybeans fed to male turkeys from hatch to 28 days of age (Trials 1 and 2).

Diet[1]	SBM	20% LTI	30% LTI	40% LTI	20% CB	40% CB
Ingredients	---- % of Total Diet ----					
Corn	30.00	30.00	30.00	30.00	30.00	30.00
SBM[1]	52.70	36.99	29.14	21.29	36.99	21.29
LTI[1]	0.00	20.00	30.00	40.00	0.00	0.00
CB[1]	0.00	0.00	0.00	0.00	20.59	40.92
Soy oil	7.15	3.87	2.25	0.60	3.36	0.00
Limestone	1.70	1.70	1.70	1.70	1.70	1.70
Dicalcium P	3.00	3.00	3.00	3.00	3.00	3.00
Sodium chloride	0.30	0.30	0.30	0.30	0.30	0.30
L-lysine HCl	0.22	0.22	0.22	0.22	0.22	0.22
DL-methionine	0.35	0.35	0.35	0.35	0.35	0.35
Sodium bicarbonate	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix[2]	0.10	0.10	0.10	0.10	0.10	0.10
Vitamin premix[3]	0.10	0.10	0.10	0.10	0.10	0.10
Selenite premix[4]	0.05	0.05	0.05	0.05	0.05	0.05
Solka-Floc®	2.363	1.353	0.823	0.323	1.273	0.00
Santoquin-6®	0.017	0.017	0.017	0.017	0.017	0.017
Celite®	1.700	1.700	1.700	1.700	1.700	1.700
Calculated Analysis						
Crude protein, %	28.6	28.6	28.6	28.6	28.6	28.6
Crude Fat, %	9.0	9.0	9.0	9.0	9.0	9.0
ME, kcal/kg	3014	3014	3014	3014	3014	3014

[1] SBM = solvent extracted soybean meal; LTI = unheated LTI soybean (20, 30 or 40%); CB = unheated conventional soybean (20 or 40%).

[2] Each kilogram of mineral premix (0.1% inclusion) supplies the following per kg of complete feed: Iron 20 mg; Copper 2.5 mg; Manganese 30 mg; Zinc 30 mg; Iodine 1.25 mg, Cobalt 1.0 mg per kilogram of complete feed.

[3] Each kilogram of vitamin premix (0.1% inclusion) supplies the following per kg of complete feed: Vit A 13,200 IU; Vit D3 3,960 IU; Vit E 66 IU; Vit B1 4 mg; Vit B2 13 mg; Vit B 6 8 mg; Vit B12 40 µg; Vit K3 4 mg; Nicotinic acid 110 mg; Pantothenic acid 22 mg; Folic acid 2.2 mg; Biotin 254 µg.

[4] $NaSeO_3$ premix provides 0.3 mg Se per kilogram of complete feed.

Table II-2. Nutrient analysis[1] of mash diets with varying levels of unheated soybeans fed to male turkeys from hatch to 28 days of age (Trials 1 and 2).

Diet[2]	SBM	20% LTI	30% LTI	40% LTI	20% CB	40% CB
Nutrient	------------	------------	------------ % of Total	------------	------------	------------
Crude protein	33.55	32.35	32.27	32.58	31.56	31.72
Crude fat	8.49	9.06	8.71	7.87	9.26	9.32
Crude fiber	3.85	3.79	4.50	4.42	4.22	4.31
Ash	9.11	9.67	10.36	10.77	10.36	10.49
Calcium	1.18	1.43	1.57	1.65	1.55	1.67
Phosphorous	1.02	1.09	1.10	1.13	1.05	1.07
Sodium	0.15	0.18	0.19	0.19	0.19	0.20
Trypsin Inhibitor, TIU/g	1095	3145	5150	5126	6861	9367
Urease Activity[3]	0.00	0.77	1.46	1.54	1.71	2.00
Dgw, µm [5]	915	895	892	779	830	775
Sgw[6]	1.95	1.98	2.06	2.27	2.15	2.23
Bulk density, kg/m^3	674	642	642	620	656	620

[1] Results presented on a dry matter basis.
[2] SBM = solvent extracted soybean meal; LTI = unheated LTI soybean (20, 30 or 40%); CB = unheated conventional soybean (20 or 40%).
[4] Urease Activity expressed as net pH increase.
[5] Geometric mean diameter calculated according ASAE Standard S319.2.
[6] Geometric mean standard deviation calculated according ASAE Standard S319.2.

Table II-3. Cumulative fat recovery of mash diets with varying levels of unheated soybeans fed to male turkeys from hatch to 28 days of age (Trials 1 and 2).

Diet[1]	SBM	20% LTI	30% LTI	40% LTI	20% CB	40% CB
	---------------------- Cumulative % Fat Recovered ----------------------					
As Fed[2]						
Wash 1	3.70	3.00	2.16	1.76	2.97	1.54
Wash 2	5.16	4.98	4.97	4.57	5.27	3.67
Wash 3	5.44	5.30	5.55	5.19	5.93	4.50
Wash 4	5.54	5.37	6.08	5.69	6.35	4.96
Wash 5	5.62	5.44	6.29	5.89	6.55	5.33
Wash 6[3]	5.72	5.80	6.49	6.01	6.66	5.58
Ground[4]						
Wash 1	4.93	4.03	4.21	3.48	4.20	3.39
Wash 2	6.39	5.56	6.91	5.69	6.74	5.97
Wash 3	6.63	5.79	7.49	6.57	7.24	6.65
Wash 4	6.79	5.70	7.70	6.88	7.56	7.00
Wash 5	6.90	5.97	7.97	7.08	7.77	7.15
Wash 6[3]	6.97	6.01	8.15	7.21	7.89	7.23

[1] SBM = solvent extracted soybean meal; LTI = unheated LTI soybean (20, 30 or 40%); CB = unheated conventional soybean (20 or 40%).
[2] Diets analysed as-fed with particle sizes from 775 to 915 µm.
[3] Recovered fat per wash is cumulative, wash 6 represents the total recovered fat after petroleum ether washes 1 through 6.
[4] Diets analysed after being ground to pass through a 0.5 mm screen.

Table II-4. Effect of unheated soybean inclusion level on mean body weight (BW) of male turkeys from 0-14 days and 14-28 days.

	Trial 1			Trial 2		
Age, days	0	7	14	14	21	28
Diet[1]	g body weight per bird					
SBM	56.92	147.67[a]	384.28[a]	542.00	956.82[a]	1598.70[a]
20% LTI	55.50	142.42[ab]	349.75[ab]	548.63	938.67[ab]	1570.15[a]
30% LTI	56.61	136.71[abc]	359.59[ab]	545.63	936.85[ab]	1590.78[a]
40% LTI	56.94	131.61[bc]	328.29[b]	547.88	923.67[ab]	1545.00[a]
20% CB	58.13	124.65[c]	312.36[b]	544.63	891.32[bc]	1509.95[ab]
40% CB	55.92	106.88[d]	240.08[c]	547.25	849.67[c]	1409.25[b]
SEM[2]	0.82	4.05	14.24	3.08	12.29	32.30
Source of Variation	Probability, $P <$					
Diet	0.230	<0.001	<0.001	0.632	<0.001	<0.001
Soybean × Inclusion[3]	0.309	0.330	0.061	0.570	0.263	0.205
LTI Response[4]						
Linear	0.948	0.002	0.009	0.200	0.056	0.291
Quadratic	0.216	0.546	0.918	0.475	0.962	0.752
CB Response[5]						
Linear	0.391	<0.001	<0.001	0.215	<0.001	<0.001
Quadratic	0.095	0.548	0.991	1.000	0.411	0.869
LTI vs. CB[6]	0.332	<0.001	<0.001	0.437	<0.001	0.002

[1] SBM = solvent extracted soybean meal; LTI = unheated LTI soybean (20, 30 or 40%); CB = unheated conventional soybean (20 or 40%).
[2] Standard error of the mean.
[3] Interaction between soybean type (LTI and CB) and inclusion level (20 and 40%).
[4] Response determined using increasing inclusion levels of the LTI soybean (0% SBM, 20%LTI, 30% LTI, and 40% LTI).
[5] Response determined using increasing inclusion levels of the CB soybean (0% SBM, 20% CB, and 40% CB).
[6] Contrast of 20% and 40% inclusion level of LTI soybeans versus 20% and 40% inclusion of CB soybean.
[a,b,c,d] Means in a column with different superscripts are significantly different ($P < 0.05$).

Table II-5. Effect of unheated soybean inclusion level on mean body weight gain (BWG) of male turkeys from 0-14 days and 14-28 days.

	Trial 1			Trial 2		
Age, days	7	14	0-14	21	28	14-28
Diet[1]	g body weight gain per bird					
SBM	90.75[a]	236.61[a]	327.35[a]	414.65[a]	666.25[a]	1056.53[a]
20% LTI	86.92[a]	224.08[ab]	311.73[ab]	404.12[ab]	631.47[ab]	1021.65[a]
30% LTI	80.11[ab]	222.88[ab]	302.98[ab]	391.20[abc]	653.92[a]	1045.13[a]
40% LTI	76.24[ab]	196.68[b]	271.34[bc]	375.78[bc]	621.32[ab]	997.10[a]
20% CB	66.53[b]	187.71[b]	254.25[c]	362.78[c]	618.62[ab]	965.13[ab]
40% CB	51.23[c]	132.94[c]	184.16[d]	302.28[d]	559.57[b]	861.85[b]
SEM[2]	3.96	9.50	12.28	8.43	21.92	31.79
Source of Variation	Probability, *P* <					
Diet	<0.001	<0.001	<0.001	<0.001	0.007	<0.001
Soybean × Inclusion[3]	0.499	0.138	0.181	0.263	0.205	0.185
LTI Response[4]						
Linear	0.003	0.006	0.001	<0.001	0.182	0.236
Quadratic	0.505	0.280	0.224	0.305	0.957	0.802
CB Response[5]						
Linear	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Quadratic	0.280	0.7883	0.909	0.638	0.808	0.869
LTI vs. CB[6]	<0.001	<0.001	<0.001	<0.001	0.055	0.002

[1] SBM = solvent extracted soybean meal; LTI = unheated LTI soybean (20, 30 or 40%); CB = unheated conventional soybean (20 or 40%).
[2] Standard error of the mean.
[3] Interaction between soybean type (LTI and CB) and inclusion level (20 and 40%).
[4] Response determined using increasing inclusion levels of the LTI soybean (0% SBM, 20%LTI, 30% LTI, and 40% LTI).
[5] Response determined using increasing inclusion levels of the CB soybean (0% SBM, 20% CB, and 40% CB).
[6] Contrast of 20% and 40% inclusion level of LTI soybeans versus 20% and 40% inclusion of CB soybean.
[a,b,c,d] Means in a column with different superscripts are significantly different (P < 0.05).

Table II-6. Effect of unheated soybean inclusion level on mean feed intake (FI) of male turkeys from 0-14 days and 14-28 days.

	Trial 1			Trial 2		
Age, days	7	14	0-14	21	28	14-28
Diet[1]	g feed intake per bird					
SBM	115.34^{a}	289.77^{a}	415.35^{a}	762.18^{a}	1191.10^{a}	1840.75^{a}
20% LTI	114.00^{a}	275.96^{a}	389.95^{ab}	748.68^{a}	1144.43^{a}	1817.90^{a}
30% LTI	06.40^{ab}	268.14^{ab}	374.56^{ab}	743.88^{a}	1132.35^{a}	1771.95^{a}
40% LTI	103.03^{ab}	267.46^{ab}	370.49^{abc}	718.43^{ab}	1148.83^{a}	1731.23^{ab}
20% CB	99.82^{ab}	248.46^{ab}	348.25^{bc}	737.08^{a}	1137.10^{a}	1694.46^{ab}
40% CB	91.02^{b}	224.51^{b}	315.52^{c}	682.13^{b}	971.33^{b}	1550.28^{b}
SEM[2]	4.29	12.75	15.41	11.95	36.87	51.19
Source of Variation	Probability, $P <$					
Diet	<0.001	0.002	<0.001	<0.001	0.003	0.002
Soybean × Inclusion[3]	0.779	0.460	0.618	0.272	0.031	0.562
LTI Response[4]						
Linear	0.024	0.113	0.016	0.010	0.347	0.104
Quadratic	0.416	0.755	0.754	0.360	0.520	0.609
CB Response[5]						
Linear	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Quadratic	0.485	0.503	0.300	0.293	0.223	0.987
LTI vs. CB[6]	0.002	0.002	<0.001	0.037	0.020	0.004

[1] SBM = solvent extracted soybean meal; LTI = unheated LTI soybean (20, 30 or 40%); CB = unheated conventional soybean (20 or 40%).
[2] Standard error of the mean.
[3] Interaction between soybean type (LTI and CB) and inclusion level (20 and 40%).
[4] Response determined using increasing inclusion levels of the LTI soybean (0% SBM, 20%LTI, 30% LTI, and 40% LTI).
[5] Response determined using increasing inclusion levels of the CB soybean (0% SBM, 20% CB, and 40% CB).
[6] Contrast of 20% and 40% inclusion level of LTI soybeans versus 20% and 40% inclusion of CB soybean.
[a,b,c] Means in a column with different superscripts are significantly different (P < 0.05).

Table II-7. Effect of unheated soybean inclusion level on mean feed conversion ratio (FCR) of male turkeys from 0-14 days and 14-28 days.

	Trial 1			Trial 2		
Age, days	7	14	0-14	21	28	14-28
Diet[1]	g feed intake: g body weight gain					
SBM	1.30^{b}	1.16^{b}	0.97^{b}	1.85^{c}	1.81	1.75
20% LTI	1.31^{b}	1.24^{b}	1.11^{b}	1.95^{bc}	1.85	1.78
30% LTI	1.34^{b}	1.19^{b}	1.05^{b}	1.90^{bc}	1.74	1.70
40% LTI	1.39^{b}	1.35^{b}	1.16^{b}	1.93^{bc}	1.78	1.74
20% CB	1.53^{b}	1.34^{b}	1.15^{b}	2.06^{b}	1.77	1.77
40% CB	1.79^{a}	1.71^{a}	1.45^{a}	2.26^{a}	1.74	1.80
SEM[2]	0.06	0.06	0.05	0.04	0.05	0.04
Source of Variation	Probability, $P <$					
Diet	<0.001	<0.001	<0.001	<0.001	0.605	0.600
Soybean × Inclusion[3]	0.109	0.037	0.014	0.006	0.747	0.333
LTI Response[4]						
Linear	0.264	0.061	0.020	0.223	0.462	0.589
Quadratic	0.534	0.522	0.867	0.336	0.735	0.732
CB Response[5]						
Linear	<0.001	<0.001	<0.001	<0.001	0.310	0.374
Quadratic	0.851	0.209	0.314	0.997	0.945	0.868
LTI vs. CB[6]	<0.001	<0.001	0.002	<0.001	0.228	0.571

[1] SBM = solvent extracted soybean meal; LTI = unheated LTI soybean (20, 30 or 40%); CB = unheated conventional soybean (20 or 40%).
[2] Standard error of the mean.
[3] Interaction between soybean type (LTI and CB) and inclusion level (20 and 40%).
[4] Response determined using increasing inclusion levels of the LTI soybean (0% SBM, 20%LTI, 30% LTI, and 40% LTI).
[5] Response determined using increasing inclusion levels of the CB soybean (0% SBM, 20% CB, and 40% CB).
[6] Contrast of 20% and 40% inclusion level of LTI soybeans versus 20% and 40% inclusion of CB soybean.
[a,b,c] Means in a column with different superscripts are significantly different ($P < 0.05$).

Table II-8. Effect of unheated soybean inclusion level on retained nitrogen (N_{Ret}), apparent metabolizable energy (AMEn), apparent lipid digestibility (ALD), and relative pancreas weight (P_{RW}) of male turkeys at 14 and 28 days of age.

	Trial 1: Day 14				Trial 2: Day 28			
Diet[1]	N_{Ret}, %	AMEn[2]	ALD, %	P_{RW}[3], %	N_{Ret}, %	AMEn[2]	ALD, %	P_{RW}[3], %
SBM	3.13[bc]	3353[b]	91.29[a]	0.37[e]	2.57[b]	3251[b]	94.17[a]	0.28[e]
20% LTI	2.86[de]	3083[c]	85.59[bc]	0.45[d]	2.19[c]	2900[d]	90.41[bc]	0.34[d]
30% LTI	3.54[a]	3601[a]	86.80[b]	0.50[c]	2.98[a]	3486[a]	92.58[ab]	0.37[cd]
40% LTI	3.31[b]	3226[bc]	81.81[c]	0.52[c]	2.36[bc]	2968[cd]	87.74[c]	0.41[c]
20% CB	3.07[bc]	3254[b]	84.66[bc]	0.71[b]	2.34[bc]	3066[c]	90.24[bc]	0.51[b]
40% CB	2.72[e]	3071[c]	76.00[d]	0.89[a]	2.16[c]	2887[d]	88.68[c]	0.68[a]
SEM[4]	0.06	43	1.28	0.01	0.08	42	0.92	0.01
Source of Variation	Probability, $P <$							
Diet	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Soybean × Inclusion[5]	<0.001	<0.001	0.021	<0.001	0.009	<0.001	0.473	<0.001
LTI Response[6]								
Linear	<0.001	0.775	<0.001	<0.001	0.852	0.016	<0.001	<0.001
Quadratic	0.019	0.563	0.830	0.541	0.950	0.766	0.373	0.464
CB Response[7]								
Linear	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Quadratic	0.027	0.351	0.337	<0.001	0.782	0.931	0.211	0.007
LTI vs. CB[5]	0.001	0.836	0.004	<0.001	0.681	0.185	0.615	<0.001

[1] SBM = solvent extracted soybean meal; LTI = unheated LTI soybean (20, 30 or 40%); CB = unheated conventional soybean (20 and 40%).
[2] AMEn expressed as kcal/kg.
[3] Relative pancreas weight (P_{RW}) calculated by dividing pancreas weight by body weight (Trial 1 n=48; Trial 2 n=40).
[4] Standard error of the mean.
[5] Interaction between soybean type (LTI and CB) and inclusion level (20 and 40%).
[6] Response determined using increasing inclusion levels of the LTI soybean (0% SBM, 20%LTI, 30% LTI, and 40% LTI).
[7] Response determined using increasing inclusion levels of the CB soybean (0% SBM, 20% CB, and 40% CB).
[8] Contrast of 20% and 40% inclusion level of LTI soybeans versus 20% and 40% inclusion of CB soybean.
[a,b,c,d] Means in a column with different superscripts are significantly different ($P < 0.05$).

Table II-9. Effect of unheated soybean inclusion level on apparent ileal digestibility of indispensable amino acids in male poults at 28 days (Trial 2).

	Total AA[1]	Arg	His	Ile	Leu	Lys	Met	Phe	Thr	Trp	Val
Diet[2]	%										
SBM	71.65[a]	79.94[a]	72.78[a]	71.06[a]	71.15[a]	74.52[a]	79.27[a]	73.68[a]	60.16[a]	75.69[a]	66.34[a]
20% LTI	65.84[ab]	74.97[ab]	66.80[ab]	63.82[ab]	64.36[ab]	69.03[ab]	73.52[abc]	67.30[ab]	53.96[ab]	51.82[b]	58.82[ab]
30% LTI	66.85[ab]	74.79[ab]	66.83[ab]	64.36[ab]	66.14[a]	68.85[ab]	75.45[ab]	68.34[ab]	53.72[ab]	67.83[a]	58.82[ab]
40% LTI	58.73[b]	68.35[b]	58.89[b]	55.13[bc]	56.93[b]	61.52[bc]	66.93[dc]	60.22[b]	44.21[b]	56.88[b]	48.11[bc]
20% CB	58.95[b]	69.03[b]	59.42[b]	54.45[c]	57.06[b]	62.39[b]	71.42[bdc]	60.73[b]	45.01[b]	54.91[b]	48.05[c]
40% CB	47.81[c]	59.27[c]	46.38[c]	39.46[d]	42.93[c]	51.67[c]	64.67[d]	48.66[c]	31.25[c]	37.35[c]	30.58[d]
SEM[3]	2.14	2.29	2.34	2.51	2.38	2.73	2.37	2.29	3.10	2.48	3.01
Source of Variation	Probability, $P <$										
Diet	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Soybean × Inclusion[4]	0.313	0.370	0.228	0.158	0.104	0.475	0.969	0.203	0.483	<0.001	0.209
LTI Response[5]											
Linear	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Quadratic	0.320	0.343	0.335	0.331	0.323	0.384	0.313	0.371	0.311	0.002	0.265
CB Response[6]											
Linear	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Quadratic	0.730	0.773	0.946	0.745	0.994	0.781	0.808	0.841	0.830	0.517	0.891
LTI vs. CB[7]	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.274	<0.001	<0.001	<0.001	<0.001

[1] Average of all amino acids.
[2] SBM = solvent extracted soybean meal; LTI = unheated LTI soybean (20, 30 or 40%); CB = unheated conventional soybean (20 or 40%).
[3] Standard error of the mean.
[4] Interaction between soybean type (LTI and CB) and inclusion level (20 and 40%).
[5] Response determined using increasing inclusion levels of the LTI soybean (0% SBM, 20%LTI, 30% LTI, and 40% LTI).
[6] Response determined using increasing inclusion levels of the CB soybean (0% SBM, 20% CB, and 40% CB).
[7] Contrast of 20% and 40% inclusion level of LTI soybeans versus 20% and 40% inclusion of CB soybean.
[a,b,c,d] Means in a column with different superscripts are significantly different (P < 0.05).

Table II-10. Effect of unheated soybean inclusion level on apparent ileal digestibility of dispensable amino acids in male poults at 28 days (Trial 2).

Diet[1]	Ala	Asp	Cys	Glu	Gly	Hyl	Hyp	Orn	Pro	Ser	Tyr
	%										
SBM	68.65^{a}	71.13^{a}	46.42^{a}	78.54^{a}	66.47^{a}	91.17^{a}	58.84^{a}	47.02^{c}	72.47^{a}	69.08^{a}	74.79^{a}
20% LTI	62.09^{ab}	6.21^{abc}	33.24^{bc}	74.36^{abc}	60.21^{ab}	89.52^{ab}	38.65^{c}	45.43^{c}	67.90^{ab}	63.14^{ab}	68.77^{ab}
30% LTI	63.50^{ab}	67.67^{ab}	40.40^{ab}	75.49^{ab}	60.77^{ab}	89.59^{a}	50.89^{ab}	53.38^{b}	69.74^{a}	61.85^{abc}	69.11^{a}
40% LTI	53.87^{b}	60.45^{bc}	30.41^{bc}	69.80^{bc}	51.36^{b}	87.87^{ab}	31.76^{c}	40.99c	61.72^{b}	55.33^{bdc}	60.61^{b}
20% CB	54.47^{b}	59.29^{c}	40.33^{ab}	69.55^{c}	51.42^{b}	85.47^{ab}	55.25^{a}	46.02^{c}	62.60^{b}	53.98^{dc}	61.37^{b}
40% CB	40.61^{c}	49.56^{d}	25.74^{c}	62.10^{d}	38.76^{c}	82.25^{ab}	43.26^{bc}	68.75^{a}	49.51^{c}	46.14^{d}	49.50^{c}
SEM[2]	2.38	2.11	2.79	1.80	2.65	1.83	3.24	1.82	1.98	2.69	2.05
Source of Variation	Probability, $P <$										
Diet	<0.001	<0.001	<0.001	<0.001	<0.001	0.007	<0.001	<0.001	<0.001	<0.001	<0.001
Soybean × Inclusion[3]	0.217	0.270	0.034	0.310	0.414	0.656	0.373	<0.001	0.050	0.995	0.322
LTI Response[4]											
Linear	<0.001	<0.001	0.002	<0.001	<0.001	0.197	<0.001	0.257	<0.001	<0.001	<0.001
Quadratic	0.287	0.337	0.617	0.379	0.261	0.870	0.916	0.010	0.202	0.480	0.225
CB Response[5]											
Linear	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Quadratic	0.951	0.603	0.164	0.632	0.650	0.533	0.194	<0.001	0.412	0.159	0.714
LTI vs. CB[6]	<0.001	<0.001	0.652	<0.001	<0.001	0.009	<0.001	<0.001	<0.001	<0.001	<0.001

[1] SBM = solvent extracted soybean meal; LTI = unheated LTI soybean (20, 30 or 40%); CB = unheated conventional soybean (20 or 40%).
[2] Standard error of the mean.
[3] Interaction between soybean type (LTI and CB) and inclusion level (20 and 40%).
[4] Response determined using increasing inclusion levels of the LTI soybean (0% SBM, 20%LTI, 30% LTI, and 40% LTI).
[5] Response determined using increasing inclusion levels of the CB soybean (0% SBM, 20% CB, and 40% CB).
[6] Contrast of 20% and 40% inclusion level of LTI soybeans versus 20% and 40% inclusion of CB soybean.
[a,b,c,d] Means in a column with different superscripts are significantly different (P < 0.05).

MANUSCRIPT III:

The effect of feed processing of novel unheated, low trypsin inhibitor soybeans on the performance of young female turkeys reared from hatch to 21 days of age.

ABSTRACT: The following study was designed to evaluate the effects of feed processing on the inclusion level of a novel low trypsin inhibitor soybean (LTI) in turkey diets from hatch to twenty-one days. Female Hybrid Converter turkey poults (336) were randomly assigned in groups of 7 to 48 Alternative Design battery cages. Treatments were arranged as a 3×2 factorial with 3 inclusion levels of the LTI soybean: 0, 20, and 40% and two feed forms: mash or crumbled pellet. Poults consumed feed and water *ad libitum* for the duration of the study. At day 7, 14 and 21 poults and feeders were weighed to determine body weight (BW) and feed intake (FI) to calculate the feed conversion ratio (FCR). At day 21, excreta were collected and pooled by pen for the determination of apparent metabolizable energy (AMEn) and apparent lipid digestibility (ALD). All birds were euthanized via cervical dislocation and their ileum and pancreas excised. Ileal contents were collected and pooled by pen for the determination of apparent ileal digestibility (AID) of various amino acids. Pancreas weights, expressed as a percentage of body weight (P_{RW}), were calculated. Reduced BW gain at 7 and 14 days with increasing inclusion of the LTI bean resulted in a linearly reduction of BW at 7, 14, and 21 days. Poults consuming crumbles gained more weight ($P \leq 0.05$) with greater average BW than poults consuming mash at 7, 14, and 21 days. LTI inclusion and feed form did not affect FI, except at day 14 where increasing LTI resulted in a linear decrease in feed consumption. There was a linear interaction in FCR between LTI inclusion and feed form at day 7 and 14, which was driven by the poorer FCR with 0% LTI inclusion in crumbles and 40% LTI in mash. LTI inclusion had no affect on FCR at day 21 or overall from day 0 to 21, while crumbled diets had better FCR as compared to mash diets. There were linear interactions between LTI inclusion and

feed form in AMEn, ALD and P_{RW}. Increased LTI inclusion resulted in decreased AMEn and ALD with an increased P_{RW}, though pelleting improved AMEn and ALD and decreased R_{RW} compared to mash diets.

Keywords: turkey, unheated soybeans, trypsin inhibitor, anti-nutritional factors

INTRODUCTION

Full fat soybeans (FFSB) offer an excellent source of both protein and fat for poultry diets. They have naturally high oil and protein content with a favorable amino acid profile for growth (Waldroup, 1982). Soybeans, however, contain several anti-nutritional factors (ANF) that can inhibit the ability of poultry to fully utilize nutrients. Trypsin inhibitors (TI), Kunitz and Bowman-Birk, are often considered to be the most problematic of these ANFs (Liener, 1981; Rackis, 1965). They bind and inhibit the digestive enzymes trypsin and chymotrypsin, thus reducing protein digestibility (Liener, 1994); while also increasing pancreatic secretions and hypertrophy leading to a loss of endogenous amino acids (Leeson and Summer; Liener, 1981). It has been well documented that increased levels of TI lead to depressed growth rates, feed intake, and feed efficiency in poultry (Liener, 1981; Rackis, 1965, Yen et al., 1973,). Fortunately, Kunitz (KTI) and Bowman-Birk (BBTI) can be denatured by heat, but at an additional cost that may reduce FFSB's competitiveness in least cost formulation.

Alternatively, new soybean cultivars are being generated that lack or have reduced levels of KTI, which could potentially eliminate the cost of heat treatment before feeding to poultry. Current research, however, has shown that while unheated KTI soybeans are nutritionally superior to conventional soybean lines, they remain substandard to conventional soybean meal (Han et al., 1991, Herkelman et al., 1993; Zhang et al., 1991). This is likely due to the remaining Bowman-Birk TI. Successful further reductions in TI have been observed on KTI soybeans with steam treatment (Batal and Parsons, 2003; Herkelman et al., 1993), roasting (McIsaac, 2005) and extrusion (Palacios et al., 2004; Perilla et al., 1997; Zhang et al., 1993). Little, however, is known about the effect of steam pelleting on KTI soybeans. Thus, the objective of this study was to determine whether steam pelleting could reduce residual TI levels and allow for greater inclusion of low TI soybeans in turkey diets.

MATERIALS AND METHODS

Birds Husbandry

Female Hybrid Converter[10] turkey poults were sourced from a commercial hatchery[11] and raised from hatch to 3 weeks of age. Poults (336) were randomly distributed into groups of 7 and weighed prior to placement in 48 Alternative Design (Siloam Springs, AR) battery cages. Cages were randomly allotted to 1 of 6 dietary treatments resulting in 8 replicates per treatment. Feed and water were consumed *ad libitum*. Batteries were located in a climate-controlled room and equipped with dual nipple water drinkers, feeding trough, supplemental heater, and collection pan located beneath the floor of each cage. Artificial light was provided for 24 hours for the first 7 days; and then adjusted to provide 23 hours of light and 1 hour of dark for the remainder of the study. Heat was provided for bird comfort and adjusted based on bird behaviour. Temperatures were monitored twice a day to yield averages of 32-35^0 C for the first 7 days followed by 26-34^0 C for the next 7 days.

Dietary Treatments

All diets were formulated to meet or exceed the breeder recommendations for young toms (Hybrid). Soy oil was used to maintain similar caloric density of diets across treatments, with decreasing levels needed as inclusion of the unheated soybean increased. To create dietary treatments solvent extract soybean meal was replaced with the unheated soybean on a protein basis (Table 1). An assumed energy value of 3300 Kcal/kg was designated for the unheated soybeans based on the NRC value given to heat processed, full-fat soybeans (NRC, 1994). Maximum

[10] Hendrix Genetics, Ontario, Canada
[11] Sleepy Creek Hatchery, Goldsboro, NC

inclusion rate of the unheated soybean was limited to 40% based on the natural oil content. Soybean variety e3311 (eMerge Genetics, Des Moines, IA) was the unheated soybean tested and contained naturally low levels of Kunitz TI (10,300 TIU/g) and hereafter is referred to as LTI. There were 3 diet formulations tested based on increasing levels of LTI inclusion: 0, 20 and 40%, with 0% containing only solvent extracted soybean meal (SBM). Dietary treatments were consumed *ad libitum* as mash or a crumbled pellet. Cellulose[12] was included in diets to balance nutrient density across treatments, while diatomaceous earth or silicone dioxide[13] was added as an indigestible marker at 1.7% of the diet.

Feed Manufacturing

Feed was manufactured in accordance with current Good Manufacturing Practices (CGMP) at the North Carolina State University Educational Feed Mill (Raleigh, NC). Corn and unheated soybeans were ground using a double high roller mill (RMS, Harrisburg, SD) furnished with 305 mm diameter rolls measuring 406 mm long. Equipped with two 15-hp motors, the approximate roll differentials were calculated as 1.5:1 on the top and 2:1 on the bottom. The roll gap or distance between roll sets was adjusted manually for whole corn and whole beans to yield finished diets with an estimated particle size of 700 microns. Whole corn was ground using a 0.64 mm gap between the top set of rolls and 0.56 mm gap between the bottom set, while the unheated LTI soybean was ground with 0.20 mm and 0.15 mm gaps, respectively. Diets were mixed in a 3.6 m^3 counterpoise mixer (Hayes and Stolz, Fort Worth, TX) for the standard time of 270 sec. Dry ingredients were allowed to mix for 90 sec prior to the addition of the liquid soy oil.

The 0, 20 and 40% LTI dietary treatments were mixed in a 3.6^3 counterpoise mixer (Hayes and Stolz, Fort Worth, TX) for the standard time of 270 sec. Dry ingredients were

[12] Solka-Floc®, International Fiber Corporation, North Tonawanda, NY
[13] Celite®, World Minerals, Inc., Santa Barabara, CA

allowed to mix for 90 sec prior to the addition of the liquid soy oil. After mixing, batches were divided into 2 separate aliquots: one to be fed as mash and the other to undergo further processing into a crumbled pellet. This was done to reduce variation in diets due to deviations in scaling or mixing. To create crumbles, mash feed was conditioned at 82^0 C for approximately 30 sec and pelleted (PM1112-2, CPM, Crawfordsville, Indiana) using a 4 mm × 35 mm die with a L:D of 8.

Live Performance

Individual poult and feeder weights were collected at day 7, 14, and 21 to acquire body weight (BW), body weight gain (BWG), and feed intake (FI). Mortalities were recorded daily. Feed conversion ratio (FCR) was calculated for each weigh period and adjusted based on mortalities by dividing the pen FI by the pen BWG plus the BW of any mortalities from the period.

Sample Collection and Chemical Analysis

Feed. Representative feed samples were obtained for each dietary treatment and analyzed by the University of Missouri Agricultural Experiment Station (Columbia, MO) for proximate analysis, trypsin inhibitor, urease index, and complete amino acid profile. A subsample of all dietary treatments was taken and stored at -20° C until processed and analyzed for digestibility measurements as outlined below.

The particle size of complete diets were determined according to dry sieving method ASAE S319.2 (ASAE, 1995). Analysis included 13 sieves, the addition of sieve agitators and 0.5 g of dispersing agent[14] per 100 g of sample with a 10 min run time. Sieve sizes ranged from 3350 to 53 µm.

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[14] Silicon Dioxide, model SSA-58, Gilson, Lewis Center, OH

Bulk density of dietary treatments was calculated using the method described by Clementson and others (2010). Briefly, sample was poured into a funnel-shaped hopper centered over a 1 L measuring cup. The sample was allowed to freely flow into the measuring cup and excess material was systematically leveled off. The filled measuring cup was then weighed and the bulk density calculated by dividing the mass of the sample in the measuring cup by the volume of the measuring cup.

Two samples of pelleted feed were taken at the pellet die during manufacture of each dietary treatment. Samples were cooled for 10 min in an experimental counter flow cooler and then allowed to rest for 24 h. All samples were weighed and sifted for separation of fines and pellets using a U.S. No. 6 sieve with 3.35 mm screen openings. Any recovered fines were weighed and fines content calculated by dividing the mass of recovered fines by the initial sample mass multiplied by 100. Sifted pellet samples were retained and used to determine the pellet durability index (PDI) using the tumble box and Holmen NHP100 (TekPro Ltd, Norfolk, UK) methods. According to the standard tumble box method, based on the ASAE Standard S269.4, 500 g of sifted pellets were placed in a revolving chamber for 10 min rotating at 100 rpm. After testing, the pellets were screened using the same U.S. No. 6 sieve as before and their weight recorded. The modified tumble box followed the same procedures as outlined by the standard method, except five 12.7 mm hex nuts were placed in the rotating chamber to increase the abrasive pressure on the pellets during analysis. Based on the Holmen method, 100 g of sifted pellets were placed in the perforated chamber of the NHP100. Pellets were then agitated with forced air at 70 psi for the designated testing time of 30 or 60 sec. After testing, the pellets were screened using a U.S. No. 6 screen and their weight recorded. PDI was then calculated for all methods as the mass of pellets retained on the screen after analysis divided by the initial mass of the pellets multiplied by 100.

To estimate the fat availability of the dietary treatments, 5 g of sample was placed in a cellulose extraction thimble measuring 20 mm in diameter and 50 mm long. The sample was rinsed with 25 ml of petroleum ether for 6 consecutive washes. The sample rested for 30 min between each wash to allow the ether to completely drain from the thimble and the sample temperature to return to ambient. The sample was stirred in the thimble prior to washes 3 and 6. After each wash, the eluate of ether and fat was collected into a beaker of known weight and placed in a forced-air drying oven at 105° C for 1 hr. After evaporating off the ether, the remaining oil residue was quantified by weighing the beaker and subtracting the empty weight. The weight of the recovered fat was then divided by the initial sample weight and multiplied by 100. Results were cumulative, with Wash 6 comprised of the total fat recovery of the sample. The fat recovery was determined on as-fed samples and samples that had been ground through a 0.5 mm screen.

Excreta. Fresh excreta samples were collected once per day for 3 days beginning on day 19 from a pan located beneath each battery cage, with care taken to avoid feed or feather contaminants. Composited samples were frozen and stored at -20° C until processed. Homogenized pen samples were lyophilized (Labconco, Kansas City, MO) and finely ground to pass through a 0.5 mm screen prior to analysis. Excreta and feed samples were then analyzed for gross energy, nitrogen, and fat content. Gross energy content was determined on an adiabatic bomb calorimeter (IKA Works, Inc., Wilmington, NC) with benzoic acid as the calibration standard, while nitrogen content was measured via combustion analysis (LECO Corporation, St. Joseph, MI) with EDTA as the calibration standard. Crude fat content was determined using petroleum ether and Soxhlet extraction. Lastly, all samples were analyzed for recovery of the indigestible marker based on the acid-insoluble ash procedure described in Vogtmann et al. (1975) for the determination of apparent metabolizations.

Pancreas Collection. On day 21, all birds were euthanized via cervical dislocation, weighed and their pancreas excised. Pancreases were carefully removed from the duodenal loop and individually weighed. Resulting weights were then expressed as a percentage of the poult's body weight.

Ileal Digesta Collection. On day 21, all poults were euthanized and their small intestine excised. Ileal contents, beginning 1 cm posterior to the Meckel's diverticulum and ending 1 cm prior to the ileal-cecal junction, were collected and pooled by pen. Composite samples were stored at -20°C prior to lyophilization (Labconco, Kansas City, MO). Samples were finely ground to pass through a 0.5 mm screen and sent to the University of Missouri Agricultural Experiment Station (Columbia, MO) for determination of amino acid concentrations. Additionally, the pooled ileal contents were analyzed for indigestible marker recovery according to Vogtmann et al. (1975), the same method utilized for feed and excreta samples.

Calculations

Retained nitrogen (N_{Ret}), nitrogen-corrected apparent metabolizable energy (AME_n), apparent lipid digestibility (ALD), and apparent ileal digestibility of amino acids (AID) were calculated using the following equations as described by Kong and Adeola (2013):

$$\mathrm{N_{Ret}} = \mathrm{N}_{feed} - \frac{(\mathrm{N}_{excreta} * \mathrm{AiA}_{feed})}{\mathrm{AiA}_{excreta}}$$

$$\mathrm{AME_n} = \mathrm{GE}_{feed} - \left(\frac{\mathrm{GE}_{excreta} * \mathrm{AiA}_{feed}}{\mathrm{AiA}_{excreta}}\right) - \left(8.22 * \mathrm{N_{Ret}}\right)$$

$$\text{ALD}(\%) = 100 - \left[\left(\frac{\text{AIA}_{feed} * \text{C}_{excreta}}{\text{AIA}_{excreta} * \text{C}_{feed}} \right) * 100 \right]$$

$$\text{AID}(\%) = 100 - \left[\left(\frac{\text{AiA}_{feed} * \text{AA}_{sample}}{\text{AiA}_{sample} * \text{AA}_{feed}} \right) * 100 \right]$$

where AiA refers to the concentration of the index compound (Celite®) in the feed, excreta, or ileal digesta sample; N refers to the nitrogen content of the feed or excreta; GE refers to the gross energy of the feed or excreta; C refers to the lipid concentration of the feed or excreta; and AA refers to the concentration of an amino acid in the feed or ileal digesta sample.

Animal Ethics

All handling procedures and euthanasia protocols were carried out in accordance with the guidelines approved by the Institutional Animal Care and Use Committee of North Carolina State University.

Statistical Analysis

All data were analysed using the GLIMMIX procedure in SAS version 9.4 (SAS Institute Inc., Cary, NC). Treatments were analysed as a 3×2 factorial with 3 levels of increasing LTI inclusion: 0, 20, and 40% and 2 feed forms: mash or crumbled pellet. Pre-planned linear and quadratic contrasts were performed to further characterize interactions and main effects as a result of increasing LTI inclusion. Treatment effects were considered significant at $P \leq 0.05$ and marginally significant at $0.10 \leq P \geq 0.05$. The experimental unit was defined as the pen (n=8) for all parameters except relative pancreas weight, where bird (n=324) served as the experimental unit.

RESULTS

Dietary Treatments

Complete diets were analyzed for nutrient composition, particle size, and bulk density with results reported in Table III-2. Diets remained similar in crude protein with slightly more variation in crude fat. Mash diets analyzed at 8.49, 9.27, and 8.27% crude fat as LTI inclusion increased, while crumbled diets had reported crude fat values of 7.87, 9.26 and 9.32%, respectively. An initial petroleum ether wash of the as-fed mash diets showed decreased fat recovery as LTI increased; however, after 6 consecutive washes, more total fat was recovered as LTI increased (Table III-3). Grinding the samples prior to rinsing with petroleum ether yielded numerically greater fat recovery both after the initial Wash 1 and cumulatively after Wash 6, though this may not as accurately predict the accessibility of the fat for the bird when compared to analyzing fat recovery on as-fed samples. Trypsin inhibitor (TI) and urease activity increased in mash diets as LTI increased from 0 to 40%, with 1.04, 2.98, and 4.01 mg TIU per g of diet corresponding to urease activities of 0, 1.10, and 1.96, respectively. There was also a numerical increase in TI and urease activity in the crumbled diets, which analyzed at 1.36, 3.46, 4.05 mg TIU per g of diet with corresponding urease activities of 0, 0.65, and 1.28, respectively. Particle size, or dgw, remained similar among mash diets, ranging from 655 to 730 µm; however, crumbled diets had numerically greater dgw as LTI increased from 0 to 40% in the diet. Crumbled diets had numerically fewer fines and greater PDI as LTI increased. Bulk density remained similar among all dietary treatments, mash and crumbled.

Live Performance

There were no differences in BW at placement (Table III- 4); however, hindered BW gain at 7 and 14 days with increasing inclusion of the LTI bean resulted in linearly reduced BW at 7, 14, and 21 days ($P = 0.004$, $P < 0.001$ and $P = 0.001$, respectively). BW gain (Table III-5) was linearly reduced ($P = 0.005$) with increasing LTI at day 14, while at day 21, gain was hindered more in 40% LTI diets when fed as mash compared to crumble, leading to a linear interaction between LTI inclusion and feed form ($P = 0.003$). LTI inclusion had no effect on BW gain from 14 to 21 days, though the residual effect from the first 2 periods resulted in an overall linear reduction ($P = 0.002$) in gain from 0 to 21 days as LTI was increased. Poults consuming crumbles gained more weight ($P \leq 0.05$) with greater average BW ($P \leq 0.05$) than poults consuming mash at 7, 14, and 21 days. LTI inclusion and feed form did not affect FI ($P > 0.05$), except at day 14 where increasing LTI resulted in a linear decrease ($P = 0.009$) in feed consumption (Table III-6). There was a linear interaction between LTI inclusion and feed form at day 7 and 14 ($P = 0.005$ and $P = 0.007$, respectively), which was driven by the poorer FCR with 0% LTI inclusion in crumbles and 40% LTI in mash.

Digestibility

There was a quadratic interaction ($P < 0.001$) between LTI inclusion and feed form for N_{RET}, which plateaued at 20% LTI and was reduced at 40% LTI in mash at a greater magnitude than in crumbles. AME_n again peaked in the 20% LTI diet and was reduced in the 40% at greater extent in mash compared to crumbles, resulting in a linear interaction between LTI inclusion and feed form ($P = 0.018$). There was a linear decrease in ALD as LTI increased, but pelleting improved digestibility over mash (linear interaction; $P < 0.001$). Greater pancreas hypertrophy (P_{RW}) was observed with increased LTI, however, pelleted diets helped to reduce those weights (linear interaction; $P < 0.001$). Total amino acid digestibility was linearly reduced as LTI inclusion increased ($P < 0.001$). Mash diets had greater total amino digestibility when compared with crumbled pellets ($P = 0.023$).

DISCUSSION

The results of this study indicate that pelleting diets containing 20 or 40% LTI soybeans improve AMEn and ALD of diets compared to mash, allowing for maximum inclusion of the LTI soybean in the diet.

LTI inclusion appeared to have a more significant impact on poult performance early in the trial. BWG was linearly decreased at 7 and 14 days in response to increasing LTI, but poults gained the same at day 21. It appears that the developing digestive systems of the younger poults struggled early on to digest the LTI diets, but were beginning to equilibrate as the trial was concluding. Literature indicates that poult digestive systems continue developing through 21 days of age and have an increased ability to handle TI (Kroghdal and Sell, 1988; Mian and Garlich, 1995). Perhaps if birds were allowed to continue maturing, performance would have continued to improve. The improvements in BWG from 14 to 21 days were not substantial enough to compensate for prior losses and are reflected in lower 21 day BWs in LTI diets.

Feed intake remained unaffected by LTI inclusion or pelleting. This contrasted work by several others who observed reduced intake with inclusion of LTI soybeans fed at various levels lower or within the same range of this trial (Chohan et al., 1993; Herkelman et al., 1993; Perez-Maldonado, 2003). Perez-Maldonado (2003) observed a 10% reduction in 23-day chick consumption with 17% inclusion of an LTI soybean in mash diets, whereas there was only a 2% reduction with 40% LTI inclusion in this trial. The lack of intake difference between mash and crumbled diets was also unexpected and the authors can only speculate that pellet quality was the cause. Increasing LTI inclusion from 0 to 40% resulted in measured PDIs of 59, 84, and 92% and corresponded to fines levels of 30, 15, and 7%, respectively. The increased pellet hardness of the LTI diets led to an increased crumble size during manufacturing. For comparison, mash diets averaged 709 µm, while crumbled diets increased from 934, 1164 and 1637 µm as LTI inclusion increased. Thus, in addition to potential palatability issues with a harder crumble, the crumble size of the LTI diets may have also been too great for the young poult. Conversely, the lack of intake improvement with the 0% crumbles is likely a reflection of poor pellet quality and high fines.

Even with the suspected feed intake issues, LTI inclusion levels up to 40% did not negatively impact FCR in mash and crumble diets at day 21 and were further improved by pelleting. Pelleting the diets increased the AMEn and ALD to a greater degree as LTI inclusion increased. This was in contrast to observed dietary TI, which were shown to increase from mash to pelleted diets. Measured TI levels increased from 1043 to 1363, 2981 to 3463, and 4008 to 4046 g TIU per g of diet in mash and pelleted diets as LTI increased from 0, 20, and 40%, respectively. This increase remains unsupported by the digestibility data or pancreas weights, which were 13.6% smaller in crumbled diets than mash. Furthermore, Perez-Maldonado (2003) observed a decrease in TI and P_{RW} with increased AMEn as diets were steam conditioned at 85°C for approximately 30 sec prior to pelleting. Under the similar pelleting conditions of this trial, the authors would have expected a similar decrease in TI and least of all, an increase.

The improvements in ALD with pelleting are likely the result of increased availability of the oil from the soybeans. Carew and others (1961) reported that mechanical processes like pelleting help to rupture the fat cells in the soybean structure, making them more available for digestion by poultry. Thus, the impact of pelleting is much greater in diets containing increased levels of LTI with more unruptured fat cells. The addition of supplemental oil also likely affected ALD. Supplemental oil was decreased as LTI level increased due to the natural oil content of the soybeans. Poults consuming diets with greater supplemental oil had greater ALD in the mash diets. This would agree with fat availability predictions made using the petroleum ether washes, where greater fat was initially recovered in early washes with diets receiving supplemental oil addition, indicating greater availability for digestion.

Digestible amino acids were linearly decreased with increased LTI inclusion and supported findings by Han and others (1991). This was likely a result of increased endogenous losses of amino acids due to increased hypersecretion by the pancreas at increasing levels of LTI. Pelleting also decreased digestible amino acids, which was likely do to heating during the conditioning process. The reduction in digestible amino acids should be considered when formulating diets with synthetic amino acids to ensure proper protein utilization of the LTI soybean.

CONCLUSIONS AND APPLICATIONS

1. Pelleting turkey starter diets containing low trypsin inhibitor soybeans allows for maximum 40% inclusion with no affect on FCR at 21 days.
2. Improvements in energy and fat digestibility in diets containing low trypsin inhibitor soybeans can be achieved by conditioning at 80°C for approximately 30 sec prior to pelleting.
3. Fat digestibility of unprocessed soybeans can be improved through pelleting, which aids in oil cell rupturing, increasing the availability of fat in the diet.

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Table III.1 Composition of mash dietary treatments with varying levels of unheated soybeans fed to female turkeys from hatch to 21 days of age.

Inclusion Level[1]	0% LTI	20% LTI	40% LTI
Ingredients	--------------	% of Diet	--------------
Corn	30.00	30.00	30.00
SBM[1]	52.70	36.99	21.29
LTI[1]	0.00	20.00	40.00
Soy oil	7.15	3.87	0.60
Limestone	1.70	1.70	1.70
Dicalcium P	3.00	3.00	3.00
Sodium chloride	0.30	0.30	0.30
Mineral Premix[2]	0.10	0.10	0.10
Vitamin Premix[3]	0.10	0.10	0.10
Selenium Premix[4]	0.05	0.05	0.05
L-lysine HCl	0.22	0.22	0.22
DL-methionine	0.35	0.35	0.35
Sodium bicarbonate	0.25	0.25	0.25
Solka Floc®	2.363	1.353	0.323
Santoquin-6®	0.017	0.017	0.017
Celite®	1.700	1.700	1.700
Calculated Analysis			
Crude protein, %	28.1	28.1	28.1
ME, kcal/kg	2890	2890	2890

[1] Diets based on inclusion of LTI (novel, unheated low-trypsin soybean

[2] Each kilogram of mineral premix (0.1% inclusion) supplies the following per kg of complete feed: Iron 20 mg; Copper 2.5 mg; Manganese 30 mg; Zinc 30 mg; Iodine 1.25 mg, Cobalt 1.0 mg per kilogram of complete feed.

[3] Each kilogram of vitamin premix (0.1% inclusion) supplies the following per kg of complete feed: Vit A 13,200 IU; Vit D3 3,960 IU; Vit E 66 IU; Vit B1 4 mg; Vit B2 13 mg; Vit B 6 8 mg; Vit B12 40 µg; Vit K3 4 mg; Nicotinic acid 110 mg; Pantothenic acid 22 mg; Folic acid 2.2 mg; Biotin 254 µg.

[4] $NaSeO_3$ premix provides 0.3 mg Se per kilogram of complete feed.

Table III.2 Nutrient analysis[1] of mash and crumbled pellet diets with varying levels of unheated soybeans fed to female turkeys from hatch to 21 days of age.

Feed Form[2]	Mash			Crumbled Pellet		
Inclusion Level[3], %	0	20	40	0	20	40
Nutrient	---------------------------------- % of Total ----------------------------------					
Crude Protein	30.13	30.15	30.8	30.84	30.62	31.00
Crude Fat	8.49	9.27	8.87	7.87	9.26	9.32
Crude Fibre	4.43	4.71	4.71	3.85	3.99	3.87
Ash	1067	10.99	10.71	9.97	10.28	10.52
Calcium	1.58	1.59	1.49	1.38	1.47	1.53
Phosphorous	1.02	1.05	1.04	0.99	1.05	1.09
Sodium	0.2	0.22	0.21	0.18	0.19	0.19
Trypsin Inhibitor, TIU/g	1043	2981	4008	1363	3463	4046
Urease Activity[4]	0.00	1.10	1.96	0.00	0.65	1.28
Dgw, µm [5]	739	732	655	934	1164	1637
Sgw[6]	2.14	2.04	2.22	2.49	2.39	2.27
Bulk Density, kg/m^3	676	637	611	609	593	629
Fines[4], %	-	-	-	29.56	15.35	6.58
Pellet Durability Index, %						
Standard Tumble[7]	-	-	-	59.37	83.50	91.71
Modified Tumble[7]	-	-	-	7.87	46.23	79.35
NHP100 at 30 sec[8]	-	-	-	20.35	68.72	86.10
NHP100 at 60 sec[8]	-	-	-	3.86	48.09	79.27

[1] Results presented on a dry matter basis.

[2] Dietary treatments fed as mash or crumbled pellet, which was conditioned at 82^0 C for approximately 30 sec and pelleted (PM1112-2, CPM, Crawfordsville, Indiana) using a 4mm × 35 mm die.

[3] Inclusion level of unheated low trypsin inhibitor (LTI) soybean achieved by replacing solvent extracted soybean meal (SBM) in the diet.

[4] Urease Activity expressed as net pH increase.

[5] Geometric mean diameter calculated according ASAE Standard S319.2.

[6] Geometric mean standard deviation calculated according ASAE Standard S319.2.

[7] Standard and modified tumble box methods according to ASAE S269.4 with five 12.7 mm hex nuts used for modification.

[8] Holmen NHP100 (TekPro Ltd, Norfolk, UK) pellet tester set at 70 psi forced air.

Table III-3. Cumulative fat recovery of mash diets with varying levels of unheated soybeans fed to female turkeys from hatch to 21 days of age.

	LTI Inclusion Level[1]		
	0%	20%	40%
	----------- Cumulative % Fat Recovered -----------		
As Fed[2]			
Wash 1	3.70	3.00	2.16
Wash 2	5.16	4.98	4.97
Wash 3	5.44	5.30	5.55
Wash 4	5.54	5.37	6.08
Wash 5	5.62	5.44	6.29
Wash 6[3]	5.72	5.80	6.49
Ground[4]			
Wash 1	4.93	4.03	4.21
Wash 2	6.39	5.56	6.91
Wash 3	6.63	5.79	7.49
Wash 4	6.79	5.70	7.70
Wash 5	6.90	5.97	7.97
Wash 6[3]	6.97	6.01	8.15

[1] Inclusion level of unheated low trypsin inhibitor (LTI) soybean achieved by replacing solvent extracted soybean meal (SBM) in the diet.
[2] Diets analysed as-fed with particle sizes from 655 to 739 µm.
[3] Recovered fat per wash is cumulative, wash 6 represents the total recovered fat after ether washes 1 through 6.
[4] Diets analysed after being ground to pass through a 0.5 mm screen.

Table III-4. Effect of unheated soybean inclusion level on mean body weight (BW) of female turkeys from 0-21 days.

		Age, days		
	0	7	14	21
Effect	------------ g body weight per bird ------------			
Inclusion Level[1]				
0%	57.37	136.55	308.05	641.00
20%	57.48	135.95	285.86	620.25
40%	57.72	124.24	267.91	588.38
SEM[2]	0.49	2.94	5.74	10.51
Feed Form				
Mash	57.41	126.85	278.57	588.45
Crumble	57.64	137.65	295.98	644.64
SEM	0.42	2.37	4.92	8.58
Inclusion Level × Feed Form				
0% Mash	57.18	134.55	307.46	622.46
20% Mash	57.14	128.90	275.97	583.62
40% Mash	57.91	117.09	252.27	559.26
0% Crumble	57.55	138.56	308.63	659.55
20% Crumble	57.82	143.00	295.75	656.87
40% Crumble	57.54	131.39	283.55	617.50
SEM	0.67	4.29	7.69	14.87
Source of Variation	---------------- Probability, *P* < -----------------			
Inclusion Level	0.848	0.006	<0.001	0.004
Linear	0.576	0.004	<0.001	0.001
Quadratic	0.909	0.491	0.738	0.668
Feed Form	0.661	0.002	0.005	<0.001
Inclusion Level × Feed Form	0.695	0.354	0.125	0.480
Linear	0.557	0.207	0.044	0.481
Quadratic	0.539	0.491	0.778	0.326

[1] Inclusion level of unheated low trypsin inhibitor (LTI) soybean achieved by replacing solvent extracted soybean meal (SBM) in the diet.

[2] Standard error of the mean.

Table III-5. Effect of unheated soybean inclusion level on mean body weight gain (BWG) of female turkeys from 0-21 days.

		Age, days			
		7	14	21	0-21
Effect		---------- g body weight gain per bird ---------			
Inclusion Level[1]					
0%		79.19	171.49	332.95	583.64
20%		78.39	155.55	334.39	562.77
40%		67.62	148.23	320.47	535.52
SEM[2]		2.80	3.53	8.03	10.50
Feed Form					
	Mash	71.12	152.90	309.88	534.28
	Crumble	80.01	163.95	348.66	587.00
SEM		2.31	9.39	6.58	8.48
Inclusion Level × Feed Form					
0%	Mash	77.38	172.91	314.99	565.28
20%	Mash	71.59	150.63	307.66	526.48
40%	Mash	61.39	135.18	306.99	511.07
0%	Crumble	81.00	170.08	350.91	601.99
20%	Crumble	85.18	160.48	361.12	599.05
40%	Crumble	73.86	161.29	333.94	559.97
SEM		4.09	5.21	11.32	15.34
Source of Variation		---------------- Probability, *P* < -----------------			
Inclusion Level		0.008	<0.001	0.406	0.008
Linear		0.005	<0.001	0.275	0.002
Quadratic		0.152	0.282	0.437	0.800
Feed Form		0.004	0.005	<0.001	<0.001
Inclusion Level × Feed Form		0.379	0.012	0.496	0.454
Linear		0.263	0.003	0.693	0.679
Quadratic		0.421	0.822	0.266	0.241

[1] Inclusion level of unheated low trypsin inhibitor (LTI) soybean achieved by replacing solvent extracted soybean meal (SBM) in the diet.

[2] Standard error of the mean.

Table III-6. Effect of unheated soybean inclusion level on mean feed intake (FI) of female turkeys from 0-21 days.

		Age, days		
	7	14	21	0-21
Effect	------------- g feed intake per bird -------------			
Inclusion Level[1]				
0%	106.98	333.11	599.04	1055.17
20%	107.00	325.92	621.98	1054.58
40%	103.42	313.01	589.78	1007.40
SEM[2]	5.62	5.88	16.82	22.19
Feed Form				
Mash	104.55	325.20	611.78	1053.02
Crumble	107.05	322.83	595.41	1025.08
SEM	5.11	4.90	14.34	18.88
Inclusion Level × Feed Form				
0% Mash	103.86	335.94	590.58	1062.45
20% Mash	101.34	324.04	631.87	1057.24
40% Mash	108.47	315.62	612.91	1039.37
0% Crumble	110.11	330.29	607.50	1047.89
20% Crumble	112.67	327.81	612.09	1051.92
40% Crumble	98.37	310.39	566.66	975.43
SEM	6.46	8.51	20.49	33.01
Source of Variation	---------------- Probability, *P* < -----------------			
Inclusion Level	0.726	0.028	0.219	0.209
Linear	0.484	0.009	0.638	0.129
Quadratic	0.695	0.646	0.100	0.523
Feed Form	0.557	0.690	0.303	0.270
Inclusion Level × Feed Form	0.110	0.758	0.281	0.586
Linear	0.113	0.977	0.114	0.427
Quadratic	0.153	0.460	0.877	0.523

[1] Inclusion level of unheated low trypsin inhibitor (LTI) soybean achieved by replacing solvent extracted soybean meal (SBM) in the diet.
[2] Standard error of the mean.

Table III-7. Effect of unheated soybean inclusion level on mean feed conversion ratio (FCR) of female turkeys from 0-21 days.

		Age, days		
	7	14	21	0-21
Effect	----- g feed intake: g body weight gain -----			
Inclusion Level[1]				
0%	1.42	1.92	1.83	1.44
20%	1.36	2.11	1.83	1.50
40%	1.56	2.13	1.85	1.50
SEM[2]	0.08	0.06	0.07	0.05
Feed Form				
Mash	1.53	2.13	1.97	1.57
Crumble	1.36	1.97	1.71	1.39
SEM	0.07	0.05	0.06	0.04
Inclusion Level × Feed Form				
0% Mash	1.39	1.90	1.92	1.49
20% Mash	1.41	2.16	1.98	1.60
40% Mash	1.78	2.34	2.00	1.61
0% Crumble	1.45	1.94	1.75	1.39
20% Crumble	1.31	2.07	1.69	1.40
40% Crumble	1.33	1.92	1.70	1.38
SEM	0.09	0.08	0.09	0.06
Source of Variation	---------------- Probability, *P* < ----------------			
Inclusion Level	0.078	0.016	0.971	0.595
Linear	0.105	0.010	0.824	0.391
Quadratic	0.110	0.186	0.938	0.586
Feed Form	0.025	0.017	<0.001	0.001
Inclusion Level × Feed Form	0.015	0.020	0.659	0.531
Linear	0.005	0.007	0.413	0.284
Quadratic	0.567	0.471	0.668	0.748

[1] Inclusion level of unheated low trypsin inhibitor (LTI) soybean achieved by replacing solvent extracted soybean meal (SBM) in the diet.
[2] Standard error of the mean.

Table III-8. Effect of unheated soybean inclusion level on retained nitrogen (N_{Ret}), apparent metabolizable energy (AMEn), apparent lipid digestibility (ALD), and relative pancreas weight (P_{RW}) of female turkeys at 21 days of age.

Effect[1]		N_{Ret}, %	AMEn[2]	ALD, %	P_{RW}[3], %
Inclusion Level[4]					
0%		4.18	3168	92.51	0.33
20%		4.37	3203	88.91	0.41
40%		3.89	3135	84.18	0.49
SEM[5]		0.16	31	0.71	0.01
Feed Form					
	Mash	4.38	3118	85.83	0.44
	Crumble	3.92	3219	91.24	0.38
SEM		0.15	29	0.68	0.01
Inclusion Level × Feed Form					
0%	Mash	4.49	3148	91.48	0.34
20%	Mash	4.79	3159	86.43	0.43
40%	Mash	3.87	3048	79.58	0.54
0%	Crumble	3.88	3189	93.54	0.32
20%	Crumble	3.95	3246	91.39	0.38
40%	Crumble	3.92	3223	88.78	0.44
SEM		0.16	37	0.80	0.01
Source of Variation		---------------- Probability, P < ----------------			
Inclusion Level		<0.001	0.057	<0.001	<0.001
Linear		<0.001	0.228	<0.001	<0.001
Quadratic		<0.001	0.037	0.216	0.610
Feed Form		<0.001	<0.001	<0.001	<0.001
Inclusion Level × Feed Form		<0.001	0.054	<0.001	<0.001
Linear		<0.001	0.018	<0.001	<0.001
Quadratic		<0.001	0.645	0.454	0.838

[1] Experimental unit was pen with n=48 for N_{RET}, AMEn, and ALD; experiment unit was bird with n=324 for P_{RW}.
[2] AMEn expressed as kcal/kg.
[3] Relative pancreas weight (P_{RW}) calculated by dividing pancreas weight by body weight with N= observations.
[4] Inclusion level of unheated low trypsin inhibitor (LTI) soybean achieved by replacing solvent extracted soybean meal (SBM) in the diet.
[5] Standard error of the mean.

Table III-9. Effect of unheated soybean inclusion level on apparent ileal digestibility of indispensable amino acids in female poults at 21 days of age.

	Total[1]	Arg	His	Ile	Leu	Lys	Met	Phe	Thr	Trp	Val
Effect	------	------	------	------	------	%	------	------	------	------	------
Inclusion Level[2]											
0%	73.08	80.48	75.54	74.01	72.72	73.65	82.49	74.54	65.49	77.15	70.23
20%	71.22	79.03	74.40	70.74	69.80	71.22	81.22	71.82	63.69	74.82	66.98
40%	67.08	75.37	69.86	65.94	65.13	66.37	78.32	67.24	58.40	74.21	61.81
SEM[3]	1.59	1.41	1.61	1.60	1.67	2.10	1.38	1.54	2.12	1.33	1.89
Feed Form											
Mash	72.02	80.00	74.51	71.87	70.84	72.47	82.92	72.88	64.10	73.63	67.93
Crumble	68.89	76.58	72.03	68.59	67.60	68.35	78.43	69.52	60.95	77.15	64.75
SEM	1.44	1.29	1.46	1.43	1.51	1.92	1.23	1.38	1.94	1.12	1.72
Inclusion Level × Feed Form											
0% Mash	75.47	82.78	77.71	76.18	74.98	76.58	85.00	76.56	68.22	77.18	72.71
20% Mash	72.34	80.36	74.81	72.15	71.14	72.81	83..49	73.14	64.95	73.43	68.29
40% Mash	68.24	76.85	71.01	67.28	66.39	68.03	80.29	68.94	59.14	70.29	62.78
0% Crumble	70.69	78.18	73.37	71.84	70.46	70.72	79.99	72.52	62.77	77.11	67.74
20% Crumble	70.09	77.70	74.00	69.33	68.46	69.62	78.94	70.50	62.43	76.22	65.67
40% Crumble	65.92	73.88	68.72	64.60	63.87	64.72	76.36	65.53	57.65	78.12	60.84
SEM	1.87	1.82	2.08	2.14	2.20	2.71	1.74	2.05	2.73	1.95	2.46
Source of Variation	------	------	------	------	------	Probability, P <	------	------	------	------	------
Inclusion Level	0.002	0.002	0.002	<0.001	<0.001	0.003	0.021	<0.001	0.003	0.209	<0.001
Linear	<0.001	<0.001	<0.001	<0.001	<0.001	0.001	0.008	<0.001	0.001	0.091	<0.001
Quadratic	0.419	0.352	0.217	0.602	0.553	0.496	0.528	0.505	0.327	0.562	0.557
Feed Form	0.023	0.004	0.061	0.023	0.025	0.018	<0.001	0.015	0.066	0.015	0.046
Inclusion Level × Feed Form	0.684	0.753	0.546	0.867	0.815	0.769	0.935	0.913	0.613	0.072	0.707
Linear	0.452	0.551	0.518	0.627	0.561	0.535	0.717	0.848	0.339	0.025	0.425
Quadratic	0.643	0.633	0.362	0.815	0.777	0.692	0.977	0.699	0.789	0.712	0.798

[1] Average of all amino acids.
[2] Inclusion level of unheated low trypsin inhibitor (LTI) soybean achieved by replacing solvent extracted soybean meal (SBM) in the diet.
[3] Standard error of the mean.

Table III-10. Effect of unheated soybean inclusion level on apparent ileal digestibility of dispensable amino acids in female poults at 21 days of age.

Effect	Ala	Asp	Cys	Glu	Gly	Hyl	Orn	Pro	Ser	Tyr
	%									
Inclusion Level[1]										
0%	71.06	71.94	50.02	78.12	68.22	70.42	56.67	73.68	72.83	74.30
20%	67.93	70.61	50.13	77.15	65.84	70.24	41.27	72.57	71.02	70.45
40%	63.63	67.22	43.66	74.17	61.38	55.79	28.15	68.81	65.25	65.51
SEM[2]	1.85	1.50	2.42	1.23	1.90	6.05	5.03	1.28	1.60	1.63
Feed Form										
Mash	68.92	71.46	49.98	77.83	66.61	71.02	46.41	73.17	70.47	71.85
Crumble	66.15	68.38	45.89	75.12	63.68	59.94	37.66	70.20	68.93	68.32
SEM	1.67	1.36	2.18	1.11	1.74	5.73	4.50	1.14	1.46	1.46
Inclusion Level × Feed Form										
0% Mash	73.54	74.35	54.23	80.12	70.51	81.90	67.56	76.19	74.12	76.89
20% Mash	69.06	71.57	51.56	78.11	67.22	74.89	41.21	73.67	71.04	71.92
40% Mash	64.17	68.47	44.15	75.27	62.11	56.27	30.45	69.65	66.25	66.74
0% Crumble	68.57	69.53	45.81	76.11	65.92	58.94	45.79	71.16	71.54	71.71
20% Crumble	66.80	69.65	48.70	76.18	64.47	65.59	41.32	71.47	71.00	68.98
40% Crumble	63.09	65.97	43.17	73.07	60.66	55.30	25.86	67.96	64.24	64.27
SEM	2.42	1.96	3.18	1.62	2.21	7.32	6.81	1.71	2.03	2.17
Source of Variation	Probability, $P <$									
Inclusion Level	0.001	0.009	0.015	0.008	0.002	0.002	<0.001	0.002	<0.001	<0.001
Linear	<0.001	0.003	0.014	0.003	<0.001	0.002	<0.001	<0.001	<0.001	<0.001
Quadratic	0.718	0.431	0.130	0.360	0.513	0.085	0.811	0.261	0.135	0.710
Feed Form	0.078	0.017	0.050	0.012	0.058	0.005	0.063	0.011	0.218	0.015
Inclusion Level × Feed Form	0.577	0.611	0.309	0.678	0.695	0.054	0.137	0.430	0.678	0.704
Linear	0.305	0.447	0.139	0.475	0.397	0.017	0.141	0.224	0.849	0.430
Quadratic	0.815	0.506	0.667	0.589	0.932	0.744	0.174	0.621	0.388	0.763

[1] Inclusion level of unheated low trypsin inhibitor (LTI) soybean achieved by replacing solvent extracted soybean meal (SBM) in the diet.
[2] Standard error of the mean.