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Evaluating the Inclusion of Phytase Super-dose in Various Broiler Diet Formulations and Novel Pellet Binders

John W. Boney

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Evaluating the Inclusion of Phytase Super-dose in Various Broiler Diet Formulations and Novel Pellet Binders

John W. Boney

Thesis submitted
to the Davis College of Agriculture, Natural Resources, and Design
at West Virginia University

in partial fulfillment of the requirements for the degree of

Master of Science
In
Nutrition and Food Science

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Keywords: phytase, Spirulina algae, super-dose, pellet binder, broiler performance

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ABSTRACT

Evaluating the Inclusion of Phytase Super-dose in Various Broiler Diet Formulations and Novel Pellet Binders

John W. Boney

Grains commonly used in poultry diets contain phytic acid, a known anti-nutrient that binds dietary phosphorus rendering it unavailable for absorption also decreasing the digestibility of other nutrients. Phytic acid also increases mucin production and endogenous amino acid secretions causing a gut irritant effect which may decrease performance. An abundance of phytase literatures suggests that phytase enzymes can liberate bound phosphorus enhancing phytate phosphorus utilization and bone mineralization. Recently, the use of super-doses of phytase has been suggested to, not only liberate bound phosphorus, but to alleviate the gut irritant effect associated with phytate phosphorus. Therefore, an experiment was conducted at West Virginia University utilizing varying inclusions of a commercially available phytase product in broiler diets that varied in corn Distillers dried grains with solubles (DDGS) inclusion to assess feed manufacture and performance variables. 1,740 straight-run Hubbard x Cobb 500 broilers were placed on study for 38 days to evaluate live performance. Broilers (6/pen for starter, 2/pen for grower and finisher) were randomly selected following the conclusion of each growth phase and were euthanized for tibia excision to examine bone mineralization efficiency among broilers consuming differing experimental diets. The results revealed a phytase x formulation interaction demonstrating a 5 point feed conversion ratio (FCR) benefit when broilers consumed a diet devoid of DDGS and containing a super-dose of phytase when compared to broilers fed diets with no phytase or DDGS. Differences in bone mineralization were only apparent in the starter phase, which may have been marginal in non-phytate phosphorus, suggesting that a super-dose of phytase may alleviate the gut irritant effect of phytate phosphorus.

Additionally, improved pellet quality has been shown to improve live performance in broilers. Pellet binders may be utilized to improve pellet quality although there is a lack of nutritive pellet binders available. *Spirulina* algae has been suggested to contain pellet binding qualities, along with its remarkable amino acid profile and protein content. Therefore, a study was conducted at the West Virginia University pilot feed mill to explain the effects of varying inclusions of Spirulina algae when manufactured at varying conditioning temperatures. Two broiler diets were formulated to be similar in nutrient content. A diet containing 0% algae and a diet containing 10% algae were batched and blended to create 5 experimental diets with varying algae inclusions. Each diet was pelleted at 3 conditioning temperatures as a randomized complete block design. The results revealed algae x temperature interactions demonstrating that as algae inclusion and conditioning temperature increased pellet durability increased, but algae inclusion was more beneficial to pellet quality at lower conditioning temperatures. Algae inclusions in diets conditioned at low temperatures may maximize pellet durability and nutrient digestibility.
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KEY

Chapter 1

1. Mixer-added fat- MAF
2. Calcium Lignosulfonate- CaLS
Chapter 2

1. Meat and bone meal- mbm
2. Distillers dried grains and solubles- corn DDGS
3. Non-phytate phosphorus- nPP
4. Feed conversion ratio- FCR
5. Phosphorus- P
6. Analysis of Variance- ANOVA
7. Pellet durability index- PDI
8. New Holmen Pellet Tester- NHPT
9. Live weight gain- LWG
10. Feed Intake- FI

Chapter 3

1. Pellet durability index- PDI
2. Modified pellet durability index- MPDI
3. New Holmen Pellet Tester- NHPT
4. Electrical energy usage- EEU
CHAPTER 1: LITERATURE REVIEW

I. Pelleting

A. Diet Formulation and Feed Manufacture

Feed ingredients and manufacturing account for 60-70% of total production costs associated in a vertically integrated poultry operation. Due to recent increases in corn usage for ethanol production, the price of corn has risen dramatically. Elevated ingredient cost has led to increased inclusions of by-products in diet formulations. By-product inclusion in diet formulations creates challenges in optimizing diet digestibility. Exogenous enzymes have been utilized to improve diet digestibility. However, enzyme retention and ultimate efficacy in digesting by-product substrates has been shown to be dependent on feed manufacture, more specifically the pelleting process. Recently, researchers have suggested that the practice of increasing conditioning temperature and decreasing mixer added fat may decrease bird performance via decreased enzyme efficacy or amino acid digestibility [1]. High mixer-added fat inclusion has been shown to improve enzyme efficacy by coating the feed particles [2] and providing lubrication within the die to decrease frictional heat [3].

Generally, commercial broiler feed is pelleted as an abundance of past research supports increased growth performance when birds are provided with pelleted feed compared to mash feed [4-9]. However, the maintenance of structural integrity or pellet quality dictates the extent of broiler performance benefit [10]. Pellets are created after mash feed is steam conditioned at a desired temperature and time, and extruded through a pellet die. The physical form is thought to elicit beneficial effects in several ways. Scheideler reports that pelleted feed has a higher nutrient density which allows broilers to consume more feed.
efficiently. Compressing feed into pellets reduces ingredient segregation and consequently improves feed efficiency as well as uniformity of broiler growth [11]. When chickens are fed pelleted feed they utilize energy more efficiently by spending less time on the activity of prehension, thus more net energy could be available for body gain [11]. In order to obtain the greatest benefit, the pellet must maintain integrity throughout processing, transport, and conveyance in the grow-out house to the feed pan [12]. Diet formulation and pelleting technique directly affect pellet quality; several techniques can be employed to increase pellet quality.

B. Steam Conditioning

One technique that can be employed is manipulation of conditioning temperature prior to pelleting. High temperatures are needed for proper agglomeration of nutrients and are essential for achieving high pellet quality [13]. Increasing conditioning temperature increases moisture and heat within the feed, thus improving pellet quality [14]. Loar et al. reported that pellet quality was significantly improved with increased conditioning temperature [1]. Cutlip and cohorts reported increased pellet durability and increased modified pellet durability when diets were conditioned at high temperatures compared to low temperatures [3]. Although pellet durability results are improved when high conditioning temperatures are obtained, there may be a heat threshold. If pelleting temperatures are too high, then nutrient availability may be compromised, particularly the availability of proteins [3].

High conditioning temperatures are sometimes coupled with low mixer-added fat. Increases in conditioning temperature have been shown to decrease relative electrical energy
usage at the pellet mill [1]. This is most likely due to the increased steam volume necessary to increase temperature producing a lubricating action at the die, thus reducing energy needed to extrude mash [15].

C. Fat Addition in Pelleting

Another technique that may be employed to increase pellet quality is the manipulation of supplemental fat addition. A practice becoming more popular in feed manufacture is the application of supplemental fat via post-pellet application [1]. Previous research has demonstrated that increased mixer-added fat (MAF) results in decreased pellet quality [16-18]. Corey et al. reported significant increases in pellet quality when utilizing low MAF compared to high MAF [19]. Wamsley and Moritz reported that the use of increased MAF reduced the detrimental pelleting effects on nutrient digestibility, specifically amino acids [17]. Corey and cohort’s findings agreed with Wamsley and Moritz, where a significant MAF x CaLS x Feed Form interaction showed decreased amino acid digestibility for treatments with low MAF, while amino acid digestibility improved in pelleted diets manufactured with high MAF. Fat added at the mixer may contribute to improved nutrient utilization and enzyme efficacy by coating feed particles, decreasing frictional heat, and reducing opportunities for thermal denaturation [2, 20]. Since no universally accepted recommendations for pellet manufacture exists, integrators have freedom to add supplemental fat at the mixer, post-pellet, or a combination of each. Maximum bird performance is often dictated by feed manufacturing techniques. Either pellet quality or nutrient digestibility is often hindered as a result of an integrator’s manufacturing methods.

D. Pellet Binders
Feed volume requirements to satisfy production needs supersedes the time investment necessary to create high-quality pellets. Consequently, feed manufacturers strive to economically maintain pellet quality without sacrificing high throughput [17]. Pellet binders offer a wide range of benefits to feed producers by improving and providing a consistent feed pellet quality and pellet durability, reducing fines, lubricating the pellet die, increasing production rates and energy efficiency [21].

Calcium Lignosulfonate (CaLS) is a commercially available product commonly used as a pellet binder. One of the most pronounced properties of CaLS is the ability to disperse particles in aqueous solutions; therefore, it is commonly used to disperse the cement and fine particles in a solution to improve the workability of dense suspensions, such as mortar or concrete [22]. Studies conducted using CaLS as a pellet binder show significant increases in pellet quality. Increases were likely due to CaLS becoming liquid with steam conditioning, filling interstitial space of feed particles, and hardening upon drying, thus improving pellet quality [23]. Corey and cohorts speculated that perhaps CaLS demonstrated dispersing agent qualities that allowed rheology of particles to be maintained during pellet extrusion, decreasing friction throughout the period of manufacture [19].

E. Algae

A recent study using a *Spirulina* algae source reported descriptive feed manufacture data that suggests algae may be a pellet binder [24]. Evans and cohorts observed increases in pellet durability when diets contained up to 21% algae in diet formulation. Diets containing no algae had a reported pellet durability of 45.5% while diets containing 21% algae had a reported pellet durability of 97.4% [24], using a New Holmen Pellet Tester, a more aggressive durability measure relative to standard tumbling methods. The specific algae product used by Evans et al.
had a powdery consistency, therefore we can speculate that the algae product worked in a similar manner as the aforementioned CaLS pellet binder and turned to a liquid when exposed to the high heat and steam conditions in the conditioning chamber. This speculation would conceivethe thought that the algae product filled interstitial space of feed particles, and hardened upon drying, thus improving pellet quality. Due to the remarkably high protein content of the *Spirulina* algae product (76%), protein gelation likely occurred at an elevated level. Proteins affect pellet formation by binding feed particles [25]. Feed particles are primarily agglomerated by starch gelatinization and protein denaturation-aggregation [16]. These findings indicate the need for more research on the pellet binding qualities of algae products.

II. Enzymes and Substrates in Feed

A. Phosphorus

Phosphorus (P) is an essential element for all life forms [26]. Phosphorus is a critical and expensive nutrient provided to poultry that supports growth and development of a strong skeleton to withstand the rigors of growth, transport, and processing and must be supplied in adequate, but not excessive, amounts [27]. Bone is the main storage organ for P containing 85% of the body’s total P. Phosphorus is essential for animals to obtain their optimum genetic potential in growth, feed efficiency and skeletal development. The requirements for this mineral are highest during the time the animal is growing [28]. Phosphorus is an essential structural component of cell membranes and nucleic acids, as well as involvement in several biological processes, including bone mineralization, energy production, cell signaling through phosphorylation reactions, and regulation of acid-base homeostasis [29].

The challenge in P nutrition is how to best make P available to the animal [30]. Complications exist because of the bioavailability of P, its numerous dietary sources, and the fact
that it is not a static number. Biological availability of P can vary depending on dietary factors such as the level of other nutrients in the diet, the relationship between the level of other nutrients and P in the diet and the type and level of P in the diet [30]. Numerous other factors can affect P availability, such as environment, management, age, sex, strain, and health status of the animal [30].

Overfeeding of P is a common commercial practice due to the lack of centralized, up-to-date publication on poultry P requirements [30]. In addition, animal availability trials are time consuming and costly. On concern surrounding the overfeeding of P is increased P excretion in the manure. With an increase in size and number of large agricultural operations, manure application to the land is also increased. This application of manure has resulted in more P being added than crops require, an accumulation of P, and consequent increased potential in P surface runoff [31]. Increased runoff and accumulation of P in water likely led to the algal blooms in the Chesapeake Bay watershed district. Algal blooms are the result of eutrophication or the increase of the rate of supply of organic matter [32]. When poultry are fed P levels closer to their requirements and strategies are implemented to improve phytate-P digestibility, P excretion can be reduced [30].

**B. Phytate and Phytase**

A large portion of the P of seed based ingredients used in a poultry diet is in the phytic acid molecule, making P poorly available. Phytic acid, or IP6, is a highly reactive acidic compound that readily binds mineral cations, and in this form, is called phytin [33]. Due to the binding of minerals, especially P, the addition of exogenous P is necessary in diet formulation. The partial availability of the P component of phytate to simple-stomached species assumes importance as
the world’s rock phosphate reserves are not renewable, which could lead to a P supply crisis in the future [34, 35]. Selle and Ravindran estimate that poultry consume in the order of one million tonnes of phytate-P annually [36]. Scientists have developed ways to reduce the amount of supplemental P addition required in commercial broiler diets. Phytase enzymes have been created to alleviate this bound phosphorus, making it available for uptake by the broiler chicken.

Enzymes are proteins or protein-based substances that speed up or catalyze chemical reactions [37]. It was not until 1991 that the first phytase feed enzyme became commercially available; it was derived from \textit{A. niger}. Phytase activity may be defined as fytase units (FTU), where one FTU is the amount of enzyme that liberates 1 micromol inorganic orthophosphate/min from 0.0051 mol L$^{-1}$ sodium phytate at pH 5.5 and a temperature of 37 °C [38].

There are two categories of phytase enzymes; 3-phytases and 6-phytases. The 3-phytase preferentially liberates the P moiety at position C$_3$, whereas 6-phytase commences at position C$_6$ of the \textit{myo-}inositol phosphate esters, via a progression of step-wise dephosphorylation reactions, to yield inositol and six inorganic P moieties [36]. Phytate degradation occurs throughout the gastrointestinal tract. Commercially available phytase enzymes are often differentiated by their original point of hydrolysis, pH profile, stability within the digestive tract, and resistance to pelleting temperatures [39].

\textbf{C. Superdoses of Phytase}

The effect of unconventionally high doses of phytase has attracted attention more recently. High inclusions of phytase are being included in an attempt to “de-phytinise” the diet [40]. The effects of such super-doses of phytase can be considerable, and often beyond that which may reasonably be expected based on improvements in P digestibility [40]. The definition of a super-dose of phytase differs among scientists. Cowieson defines a phytase super-dose as a
phytase dosage greater than 2,500 FTU/kg from *A. niger* or *E. coli* while York and Wyatt of AB Vista Feed Ingredients define a phytase super-dose as a dosage greater than 1,500 FTU/kg of an *E. coli* phytase.

Initially a higher dose of phytase would have been considered to give an equivalent decrease in the need for supplemental inorganic P in the diet, looking for a further reduction in the cost of the diet while maintaining animal performance [41]. However, substrate is limited and scientists now want to consider other possibilities when using superdoses of phytase. One alternative is to look to eliminate the anti-nutritional effect of the phytate through higher enzyme doses, thereby increasing nutrient absorption and animal performance [41]. Ravindran reported that the aim of super-dosing is to dephosphorylate the phytic acid as quickly as possible to less-reactive inositol phosphate esters during the early gastric phase of digestion and to reduce its antinutritional effects [42]. Ravindran’s strategy was tested by Shirley and Edwards (2003) and it was reported that increasing phytase inclusions were associated with substantial increases in total tract phytate degradation ranging from 40.3 to 94.8%. Shirley and Edwards also noted that this phytate degradation correlated to marked improvements in bird performance, nutrient retention, tibia ash, and apparent metabolizable energy (AME), and these increases were most pronounced at the highest phytase inclusion rate, 12,000 FTU/kg [43].

Adeola and Cowieson hypothesized three principle mechanisms for which using high doses of phytase may elicit beneficial effects. The first is by releasing more phosphorus. The second is increased degradation of insoluble antinutritional phytate esters; they are then converted to more soluble myo-inositol esters. Third, by generating more soluble myo-inositol esters which may have vitamin-like or lipogenic effects [44]. These are only suggested mechanisms as the exact mechanism is still being investigated.
D. Wheat and Non-Starch Polysaccharides

Wheat has long been considered a superior poultry feed ingredient, but its feeding value can be highly variable and dependent to a large extent on its content of high molecular weight, water soluble non-starch polysaccharides (NSP) [45]. The level of pentosans, which are the principal viscous NSP in wheat, vary considerably from sample to sample, a factor that may partially explain the variability in reported results [46]. Wheat is the preferred cereal grain in many broiler diets made in most wheat-producing countries [47]. Although wheat contains high levels of NSP, numerous studies have proven that the addition of enzymes can reduce or even eliminate these negative effects. Silversides and Bedford reported that the addition of a commercial enzyme with xylanase and protease activities to wheat-based diets was effective in reducing intestinal viscosity and improving broiler performance. They also reported that over 80% of wheat-based broiler feeds now contain enzyme [48].

The use of whole grains, such as wheat, is becoming more common. This practice is known to decrease feed costs, as well as meet consumer demands for more “natural” feeding systems that provide gizzard stimulation and improved animal welfare [49]. Forbes and cohorts speculated that whole grain diets could be successfully fed to poultry because the birds select whole grain and pellets from the feed trough in the proportions that best meet their individual nutritional needs [50]. Feeding whole grains is known to affect the digestive tracts of the bird by increasing the gizzard weight. This increased weight is due to increased contractions to cope with the extra grinding needed to process the large particle size for further digestion in the distal parts of the intestine [49].

Increased fermentation occurs in the small intestine when a large amount of viscous NSP’s are present in the diet and this is detrimental to the performance and well-being of poultry [51].
The addition of soluble NSP significantly (P<0.01) increased gut viscosity, reduced the AME of the diet and depressed the growth and feed conversion ratio (FCR) of the birds. Enzyme supplementation of the NSP-enriched diet has been shown to reverse the adverse effects, increasing (P< 0.01) weight gain, FCR and AME [51].

References


21. LignoTech USA Inc., Rothschild, WI.


   rapid determination of phytase activity. J. AOAC Int. 77, 760–764.


41. dos Santos, T.T., and R. ten Doeschate. 2011. Phytase use in poultry diets: Going beyond 
   phosphorus release. World Poultry. 
   http://www.worldpoultry.net/Broilers/Nutrition/2011/8/Phytase-use-in-poultry-diets- 
   Going-beyond-phosphorus-release-WP009252W/

42. Ravindran, V. 2013. Feed enzymes: The science, practice, and metabolic realities. J. 

   Standards Improves Broiler Performance. Poult. Sci. 82:671-680


   43 (1):35-41


47. Soleimani Roudi, P., A. Golian, and M. Sedghi. Metabolizable energy and digestible 
   amino acid prediction of wheat using mathematical models. 2012 Poultry Science 
   91:2055-2062

   Recovery and Efficacy of a Xylanase Enzyme in Wheat-Based Diets. Poultry Science 
   78:1184-1190


CHAPTER 2: HiPhos dose effects in practically formulated diets that vary in ingredient composition on feed manufacture and broiler performance

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SUMMARY Ingredient variation and use of enzymes can affect diet cost, milling efficiency and broiler performance. The objectives of this study were to describe the manufacture and feeding effects of a corn, soybean meal, wheat, and meat and bone meal (mbm) based diet with varying levels of corn distillers dried grains and solubles (DDGS) and Ronozyme HiPhos phytase. Treatments comprised a 3 × 2 factorial arrangement that varied in phytase (0, 900, and a super-dose of 6,000 FTU/kg) and DDGS inclusion (0 or 5%). Phytase inclusion decreased dietary non-phytate phosphorus (nPP) and total Ca in the formulation by 0.12 and 0.1% respectively. All diets were steam conditioned at 82°C for 10s, extruded through a 4.7 × 38 mm pellet die, and fed as crumbles in the starter and grower phases and as pellets in the finisher phase. Phytase activity was evaluated using AOAC procedures and demonstrated an average HiPhos thermal stability of 87%. Ten replicate pens of straight-run Hubbard × Cobb 500 chicks were fed one of six dietary treatments for 38 d. Phytase improved feed conversion ratio (FCR) in the starter phase (P=0.05) but benefits were not carried over into the grower phase (P=0.29). Phytase and DDGS inclusion main effects interacted to affect overall FCR (P=0.05), demonstrating a 5 point improvement in FCR when birds were fed a diet without DDGS and a super-dose of HiPhos phytase. Based on tibia ash measures, performance improvement associated with the super-dose of HiPhos was likely associated with reducing phytate phosphorus irritation rather than meeting bird phosphorus requirement. Incorporation of high levels of phytase in diet formulations may be a cost effective method to improve broiler performance.

Keywords: HiPhos, FCR, broiler performance, super-dose
DESCRIPTION OF PROBLEM

Phosphorus (P) is a critical and expensive nutrient provided to poultry that supports growth and development of a strong skeleton to withstand the rigors of growth, transport, and processing and must be supplied in adequate but not excessive amounts [1]. A major portion of the P in poultry diets is in the form of phytate P, an organically bound form of the mineral that is poorly available to monogastric animals [2]. Exogenous phytase has been used in diets to improve performance and reduce production costs and environmental impact [3]. However, phytate P has additionally been found to irritate the gastrointestinal tract [4] and generate an energy expensive immune response [5-8]. Even though the mechanism is not clear, dietary phytases may be capable of reducing the immune response and subsequent production of free radicals in broiler chickens [9].

The use of super-doses of phytase has been reported to alleviate anti-nutritional effects of phytate P [10] as well as improve P availability [11]. Enzyme efficacy has been shown to be dependent on substrate concentration, enzyme dose, ingredient type, and ingredient by ingredient interactions [12-15]. Today’s commercial broiler diets contain a variety of cereals and by-products that can manipulate substrate type and enzyme accessibility. The objectives of this study were to describe the manufacture and feeding effects of a corn, soybean meal, wheat, and meat and bone meal (mbm) based diet with varying levels of DDGS and Ronozyme HiPhos phytase.
MATERIALS AND METHODS

Experimental diets were formulated to be corn, soybean meal, wheat, and meat and bone meal based (Table 1). The fat source was an animal and vegetable blend [16] and the rock phosphorus source was dicalcium phosphate. Wheat inclusion was similar across all diets at 25%; however, xylanase was not included in any of the formulations. Diets varied in corn DDGS and RONOZYME HiPhos phytase inclusion. The phytase product used in this experiment was commercially available [17], RONOZYME HiPhos phytase, with a suggested P and Ca sparing effect of 0.12 and 0.1%, respectively, that were incorporated into diet formulations. Phytase additions (0, 0.005, and a super-dose of 0.03%) were applied to diets with or without DDGS.

Dietary treatments were manufactured at the West Virginia University Pilot Feed Mill in Morgantown, West Virginia, using a 40 HP California Pellet Mill [18]. A premix of micro-ingredients, less salt and phytase, was made for each diet according to formulation. On the day of feed manufacture 4.5kg of ground corn and appropriate inclusions of salt, and phytase were placed in a Hobart mixer and allowed to mix for 10 minutes. This combination of ingredients was then added to the mixer, along with the aforementioned premix, prior to dry mixing to allow for mixer coefficient of variation to be determined through chloride analysis [19] as a predictor of uniformity of mixed phytase.

All diets were individually batched, mixed for 10 minutes dry, mixed for an additional 10 minutes post animal vegetable blend fat addition, and then conveyed to the pellet mill. All six diets per growth phase (starter, grower, and finisher) were batched and pelleted on the same day. Diets were conditioned at 82°C for 10 seconds and extruded through a 4.8 x 38.1 mm pellet die. Hot pellet samples were collected immediately following pellet extrusion through the pellet die.
and used to measure hot pellet temperature. Starter diets (D1-10) were finely crumbled so that chicks could easily consume the feed on D1. Particle size of finished feed was manipulated by changing the gap distance between rolls on the roller mill. Grower diets (D11-22) were coarsely crumbled to create a large crumble to prepare the broilers for a pelleted diet that would be fed in the finisher period (D23-38). Feed samples were collected throughout each stage of feed manufacture for analysis of pellet durability, phytase activity and mineral content.

Particle size analysis was conducted on starter and grower diets, while percent pellet analysis was conducted on finisher diets. Pellet durability analyses were conducted on all dietary treatments for all three growth phases. Pelleted samples were collected post-cooling and prior to the roller mill. Pellet durability was analyzed using three separate methods that varied in agitation. Descriptive feed manufacture data can be found in Table 2.

Standard AOAC 2000.12 phytase activity analysis was performed on all dietary treatments. Duplicate feed samples were sent to a commercial laboratory [20] and the phytase activity values represent an average. Phytase analysis has been described as erroneous and this should be considered when viewing these data (Table 3). Native phytase activity was found in all 0% phytase diets across the three growth phases, likely due to wheat inclusion.

Duplicate feed samples were sent to another commercial laboratory [21] for mineral analyses to determine the total phosphorus, phytic acid, and calcium content of finished feed. Total phosphorus and phytic acid were used to calculate nPP [22]. These results demonstrated the formulation reduction in nPP and Ca with either standard or super-doses of phytase compared to formulations with no added phytase (Table 3).
A total of 1,740 Hubbard x Cobb 500 straight run day old broilers chicks were obtained from a commercial hatchery [23] and randomly placed at a count of 29 broilers per pen in 60 pens. The six dietary treatments were randomly allotted to adjacent pens blocked by location in the barn located at the West Virginia University Animal Sciences Farm. Each dietary treatment was applied to 10 replicate pens of broilers. Lighting was continuous from D1-3, reduced 1 hour per day from D4-7, reduced 4 hours per day from D8-24 and reduced 6 hours per day from D25-28. Feed and water were provided *ad libitum* throughout the study and temperature was manipulated daily based on the Cobb Performance Guide [24]. Feed intake was measured throughout the study. On D10 chicks were weighed as a pen and six chicks per pen were randomly selected, and euthanized for tibia excision and subsequent ash analysis. Two birds per pen were randomly selected, and euthanized on days 22 and 38 for tibia excision and ash analysis [25].

All animals were reared according to protocols established by the West Virginia University Animal Care and Use Committee [ACUC 11-0703].

**Statistical Analysis**

Two separate analyses were performed on the randomized complete block design. Analysis of variance (ANOVA) was considered using the GLM procedure of SAS [26]. The pen served as the experimental unit for analysis. A phytase x formulation factorial analysis was performed to explore the main effects and interactions. Additionally, an analysis including all treatments was performed and significant differences from ANOVA (P=0.05) justified multiple comparison testing. Means were further compared using Fisher’s least significant difference test and letter superscripts were used to denote differences among treatment means.
RESULTS AND DISCUSSION

Feed Manufacture

Feed manufacture and pellet quality data were not replicated and should be considered descriptive (Table 2). Pellet durability index (PDI) values ranged from 80.2 to 85.2%, 76.2 to 81.5%, and 77.4 to 82.1% in the starter, grower, and finisher periods, respectively. New Holmen Pellet Tester (NHPT) values ranged from 62.0 to 74.8%, 54.0 to 62.4%, and 54.6 to 69.9% in starter, grower, and finisher periods, respectively. Hot pellet temperatures ranged from 80.0 to 81.8˚C across all dietary treatments and growth phases. Starter phase particle size ranged from 1150 to 1352 microns. Grower diet particle size ranged from 1921 to 2244 microns. Finisher treatment percent pellet ranged from 76 to 85%.

Phytase Activity

Mixer samples indicated chloride coefficients of variation below 10 that suggested phytase was evenly distributed throughout the batch of feed prior to steam conditioning. (data not shown). The target activity for diets containing standard and super-dose inclusions of phytase was 1,000 and 6,000 FTU/kg, respectively. The aforementioned errors associated with phytase analysis are notable in phytase activity results. Diets without added exogenous phytase had a native phytase activity ranging between 200 and 800 FTU/kg. Diets with phytase, standard and super-dose, had an analyzed activity ranging from 900 to 1,200 and 3,900 to 6,400 FTU/kg, respectively (Table 3).

Live Performance
Starter performance data demonstrated a decrease in FCR (P=0.05) as phytase inclusion increased across formulation (Table 4). A similar effect was not apparent in the grower or finisher period. Overall FCR demonstrated a Phytase by Formulation interaction (P=0.05) indicating that phytase inclusion decreased FCR for broilers fed diets without DDGS (Table 7). In fact, a 5 point decrease in FCR was demonstrated when a super-dose of phytase was included relative to diets without phytase. When DDGS were included in the formulation the benefit was lost. Perhaps, the DDGS utilized in this study contained less digestible amino acids and/or had a fiber content that interfered with phytase access to diet substrate. Past research has shown that during the thermal processing of DDGS that certain amino acids necessary for chick growth can be negatively affected [27]. These effects have been attributed to differences in processing and drying temperatures [28]. Moreover, non-starch polysachharides of DDGS may reduce the capacity for absorption by reducing enzyme accessibility to substrate [29].

Numerical increases in starter live weight gain (LWG) (P=0.08) were observed as phytase inclusions increased across formulations. A significant phytase effect in the grower period demonstrated that birds consuming diets with a super-dose of phytase had higher LWG relative to other phytase inclusions (P= 0.02, Table 4). Shirley and Edwards (2003) found similar D16 results when birds were supplemented with super-doses of phytase up to 12,000 FTU/kg [30]. Live weight gain in the finisher period was effected by the interaction of phytase and formulation (P=0.02) indicating improved LWG for broilers provided increased phytase across formulations devoid of DDGS (Table 4). Overall LWG trended towards an interaction between phytase and formulation (P=0.06), demonstrating an 80 gram LWG increase for broilers provided the super-dose of phytase relative to no phytase in diet formulations devoid of DDGS (Table 5). The authors speculate that with a 25% wheat inclusion, a 5 % inclusion of DDGS, and
no inclusion of a xylanase enzyme that the intestinal viscosity of the bird moved past a critical point and uptake of nutrients was negatively affected. In addition, it is likely that enzyme accessibility to substrate was affected due to increased digesta viscosity from NSP compounds that encapsulated nutrients [31]. The observed increased LWG for broilers provided the diet containing DDGS and no phytase may support this speculation due to the 0.12% increase in nPP and 0.1% increase in Ca relative to diets containing phytase. If the phytase was unable to interact with diet substrates due to increased intestinal viscosity, the resulting decreased LWG may be expected. Increased intestinal viscosity is generally associated with reduced growth performance [32].

Differences in feed intake (FI) were only observed in the grower phase (P=0.02) demonstrating that birds fed diets devoid of DDGS consumed more feed than birds fed diets containing DDGS. Overall FI results demonstrated no differences (P=0.25) among experimental treatments. Loar and cohorts found similar FI results when feeding broiler diets containing 0 or 8% DDGS to 28 days of age [33].

**Bone Mineralization**

The starter phase was the only phase that significant differences were observed in bone mineralization (Table 6). Starter mg tibia ash per bird showed a significant formulation effect (P=0.04) indicating that birds provided the diet without DDGS had improved mineralization. The authors found it remarkable that a 5% inclusion of DDGS would depress mineralization and speculate that perhaps the starter diets were at a marginal level of nPP and that the phytase was being used to meet the nutrient requirements for skeletal growth. Recent work by Phillips and cohorts suggests nPP requirements for 0-10 day old broilers may be as high as .58-.62% in order
to maximize tibia ash [34]. Perhaps, as previously mentioned, intestinal viscosity surpassed a critical point when relatively small inclusions of DDGS were added to the basal diet that contained 25% wheat. It has been reported that DDGS contain higher levels of fiber and non-starch polysachharides than the parent grain which can inhibit efficient digestion [35]. Past research demonstrates that DDGS inclusion in corn-soy based diet formulations can be increased across broiler growth phases. Wamsley and cohorts reported that inclusions of 8% DDGS in broiler starter diets and inclusions of 15% DDGS in grower diets resulted in no detriment to bird performance [36]. Similar research that utilized DDGS inclusions in corn-soy based broiler diets has demonstrated similar conclusions [37-39]. The combination of marginal nPP levels, 25% wheat, and 5% DDGS inclusion may have led to significant differences in starter tibia ash results. The percent tibia ash per bird in the starter phase showed a phytase effect (P=0.02), demonstrating improved mineralization with increasing phytase. Phytase likely increased phosphorus availability, especially in super-dose formulations to meet nPP requirements for tibia ash.

Bone mineralization in selected birds from the grower and finisher phase showed no treatment differences or trends (P>0.05, Table 6). Perhaps, sufficient nPP levels negated bone mineralization sensitivity to experimental treatments. Therefore, these data support that performance benefits associated with phytase were associated with reducing phytate phosphorus gut irritation than meeting nPP requirements.

CONCLUSIONS AND APPLICATIONS
1. Diets containing a super-dose of phytase and devoid of DDGS demonstrated a 5 point FCR benefit compared to similar diets without phytase possibly due to reducing phytate phosphorus gut irritation.

2. The inclusion of DDGS in this study either provided reduced nutrient availability relative to calculated formulation and/or contributed to a level of NSP’s that decreased LWG and phytase efficacy.

REFERENCES AND NOTES


16. Valley Proteins Inc., Linville, VA.

17. DSM Nutritional Products Inc., Parsipanny, NJ.

18. Master Model Pellet Mill, California Pellet Mill Company (CPM), Crawfordsville, IN.

19. Chloride QuanTab Test Strips. Hach Company. Loveland, CO.
20. Eurofins USA, Des Moines, IA

21. NP Analytical Laboratories, St. Louis, MO.

22. nPP= Total Phosphorus – (Phytic acid * 0.282)

23. Pilgrim’s Pride Corporation, Moorefield, WV.

24. Cobb-Vantress, Inc.

25. The left tibia was excised from randomly selected birds and placed in a freezer until tibia ash analysis began. Tibiae were placed in a drying oven for 72 hours. Once dried, tibiae were wrapped in filter paper and placed in a Soxhlet apparatus for 18 hours. Following fat extraction the tibiae were allowed to dry. Tibiae were removed from filter paper, weighed and placed in an ashing oven at 600°C for 16 hours. The inorganic matter remaining was weighed and ash content was determined. Three measurements were analyzed from the tibiae ash collection; mg of tibia ash per bird, mg of tibia ash per kg of bird, and tibia ash percentage per bird.


Table 1. Ingredients and Calculated Nutrient Composition of Diets Provided to Broilers Across All Growth Phases

<table>
<thead>
<tr>
<th>Diet Formulation</th>
<th>Growth Period</th>
<th>Ingredients</th>
<th>ME (kcal/kg)</th>
<th>CP (%)</th>
<th>Dig. Met + Cys</th>
<th>Dig. Threonine</th>
<th>Dig. Lysine</th>
<th>Sodium</th>
<th>Calcium</th>
<th>Phosphorus</th>
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</thead>
<tbody>
<tr>
<td>No DDGS and No Phytase$^1$</td>
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<td>36.41</td>
<td>25.00</td>
<td>24.59</td>
<td>8.50</td>
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<td>0.73</td>
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<td>5.06</td>
<td>0.31</td>
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<td>4.93</td>
<td>0.30</td>
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<tr>
<td>No DDGS + Standard Phytase$^2$</td>
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<td>No DDGS + Super-dose Phytase$^2$</td>
<td>Starter</td>
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<td>24.39</td>
<td>8.50</td>
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<td>0.32</td>
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<td>38.80</td>
<td>25.00</td>
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<td>4.64</td>
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<td>8.00</td>
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<td>4.52</td>
<td>0.29</td>
<td>0.03</td>
<td>0.24</td>
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<td>8.50</td>
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<td>25.00</td>
<td>23.03</td>
<td>8.50</td>
<td>5.00</td>
<td>2.78</td>
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<td>5.00</td>
<td>4.80</td>
<td>0.31</td>
<td>0</td>
<td>0.24</td>
</tr>
</tbody>
</table>

$^1$ Adequate nPP and Ca
$^2$ Formulation was reduced in calculated nPP by 0.12% and calculated Ca by 0.1%.
Monensin Sodium 60 gpb (90 g/ton inclusion), Elanco Animal Health, Indianapolis, IN. As an aid in the prevention of coccidiosis caused by *Eimeriaacervulina, Eimeriatenella, Eimeria brunette, Eimeriamivati,* and *Eimeria maxima.*

Bacitracin Methylene Disalicylate 50 g/lb (50 g/ton inclusion), Alpharma, Fort Lee, NJ. For increased rate of weight gain and improved feed efficiency.

Table 2. Descriptive Feed Manufacture and Quality Data for Starter, Grower, and Finisher Phases

<table>
<thead>
<tr>
<th>Diet Formulation</th>
<th>Manufacture Data</th>
<th>Particle Analysis</th>
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<tr>
<td></td>
<td>Mill Amperage (A)</td>
<td>Production Rate (tonne/hr)</td>
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</tr>
<tr>
<td>Finisher</td>
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<td>Finisher</td>
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1 Adequate nPP and Ca
2 Formulation was reduced in calculated nPP by 0.12% and calculated Ca by 0.1%.
3 New Holmen Pellet Tester
4 Pellet durability index using Pfost tumbling method.
5 Modified pellet durability index using Pfost tumbling method with five 13-mm hex nuts.
6 WS Tyler Ro-Tap Sieve Shaker
7 Complete feed is passed through a No. 6 Tyler Sieve. Pellets remaining on sieve are weighed back and calculated as a percentage.
<table>
<thead>
<tr>
<th>Diet Formulation</th>
<th>Growth Period</th>
<th>Pelleted Phytase Analysis</th>
<th>Mineral Analyses</th>
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<tr>
<td></td>
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<td>Phytase Activity (FTU/kg)</td>
<td>Total Phosphorus</td>
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<tr>
<td>No DDGS and No Phytase $^3$</td>
<td>Starter</td>
<td>690</td>
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<td>No DDGS + Standard Phytase $^4$</td>
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<td>Finisher</td>
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<td></td>
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<td>5700</td>
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$^1$ Eurofins USA, Des Moines, IA  
$^2$ NP Analytical Laboratories, St. Louis, MO  
$^3$ Adequate nPP and Ca  
$^4$ Formulation was reduced in calculated nPP by 0.12% and calculated Ca by 0.1%.  
$^5$ nPP = Total Phosphorus – (Phytic acid * 0.282)
Table 4. Effects on Performance Variables for Starter, Grower, and Finisher Phases

<table>
<thead>
<tr>
<th>Phytase Inclusion</th>
<th>Formulation</th>
<th>Live Weight Gain per Bird d10 (kg)</th>
<th>Feed Intake d1-10 (kg)</th>
<th>Feed Conversion Ratio d1-10</th>
<th>Live Weight Gain per Bird d22 (kg)</th>
<th>Feed Intake d11-22 (kg)</th>
<th>Feed Conversion Ratio d11-22</th>
<th>Live Weight Gain per Bird d38 (kg)</th>
<th>Feed Intake d23-38 (kg)</th>
<th>Feed Conversion Ratio d23-38</th>
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<tbody>
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<td>No Phytase¹</td>
<td>No DDGS</td>
<td>0.213</td>
<td>7.485</td>
<td>1.22</td>
<td>0.707¹</td>
<td>27.125⁵</td>
<td>1.69</td>
<td>1.389⁴</td>
<td>53.708</td>
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<td>Super-dose Phytase²</td>
<td>No DDGS</td>
<td>0.222</td>
<td>7.591</td>
<td>1.19</td>
<td>0.733¹²</td>
<td>26.902⁷</td>
<td>1.63</td>
<td>1.435¹³</td>
<td>52.587</td>
<td>1.81</td>
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<tr>
<td></td>
<td>DDGS</td>
<td>0.216</td>
<td>7.368</td>
<td>1.21</td>
<td>0.719¹⁴</td>
<td>25.927⁷</td>
<td>1.66</td>
<td>1.386¹⁵</td>
<td>51.263</td>
<td>1.89</td>
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<td>Fisher’s LSD¹⁷</td>
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<td>0.008</td>
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<td>0.00008</td>
<td>0.130</td>
<td>0.007</td>
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<td>0.961</td>
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<td>10.261</td>
<td>0.005</td>
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Marginal Means

<table>
<thead>
<tr>
<th>Phytase Inclusion</th>
<th>Formulation</th>
<th>Live Weight Gain per Bird d10 (kg)</th>
<th>Feed Intake d10-10 (kg)</th>
<th>Feed Conversion Ratio d10-10</th>
<th>Live Weight Gain per Bird d22 (kg)</th>
<th>Feed Intake d11-22 (kg)</th>
<th>Feed Conversion Ratio d11-22</th>
<th>Live Weight Gain per Bird d38 (kg)</th>
<th>Feed Intake d23-38 (kg)</th>
<th>Feed Conversion Ratio d23-38</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Phytase¹</td>
<td>No DDGS</td>
<td>0.211</td>
<td>7.366</td>
<td>1.226*</td>
<td>0.709⁶</td>
<td>26.530</td>
<td>1.681</td>
<td>1.418⁸</td>
<td>53.243</td>
<td>1.860</td>
</tr>
<tr>
<td></td>
<td>DDGS</td>
<td>0.212</td>
<td>7.410</td>
<td>1.212*</td>
<td>0.706⁸</td>
<td>26.554</td>
<td>1.666</td>
<td>1.392⁹</td>
<td>53.431</td>
<td>1.871</td>
</tr>
<tr>
<td>Standard Phytase²</td>
<td>No DDGS</td>
<td>0.219</td>
<td>7.479</td>
<td>1.201*</td>
<td>0.726¹⁰</td>
<td>26.415</td>
<td>1.651</td>
<td>1.405¹¹</td>
<td>51.925</td>
<td>1.856</td>
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<tr>
<td></td>
<td>DDGS</td>
<td>0.216</td>
<td>7.514</td>
<td>1.211</td>
<td>0.713¹²</td>
<td>26.834¹</td>
<td>1.667</td>
<td>1.394¹³</td>
<td>53.075</td>
<td>1.856</td>
</tr>
<tr>
<td>No DDGS</td>
<td>No DDGS</td>
<td>0.216</td>
<td>7.323</td>
<td>1.215</td>
<td>0.714¹⁴</td>
<td>26.165¹</td>
<td>1.665</td>
<td>1.416¹⁵</td>
<td>52.657</td>
<td>1.869</td>
</tr>
</tbody>
</table>

Main Effect and Interaction Probabilities

| Phytase          | 0.08 | 0.75 | 0.05 | 0.02 | 0.92 | 0.29 | 0.36 | 0.28 | 0.80 |
| Formulation      | 0.19 | 0.12 | 0.58 | 0.88 | 0.02 | 0.86 | 0.14 | 0.62 | 0.52 |
| Phytase X Formulation | 0.75 | 0.90 | 0.44 | 0.19 | 0.13 | 0.44 | 0.02 | 0.48 | 0.08 |

¹ Adequate nPP and Ca
² Formulation was reduced in calculated nPP by 0.12% and calculated Ca by 0.1%.
Table 5. Phytase and Formulation effects on Overall (D1-38) Period Performance Variables

<table>
<thead>
<tr>
<th>Phytase Inclusion</th>
<th>Formulation</th>
<th>Live Weight Gain per Bird d38 (kg)</th>
<th>Feed Intake d1-38 (kg)</th>
<th>Feed Conversion Ratio (^1) d1-38</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Phytase (^1)</td>
<td>No DDGS</td>
<td>2.46(^{b})</td>
<td>88.31</td>
<td>1.73</td>
</tr>
<tr>
<td></td>
<td>DDGS</td>
<td>2.53(^{a})</td>
<td>85.96</td>
<td>1.72</td>
</tr>
<tr>
<td>Standard Phytase (^2)</td>
<td>No DDGS</td>
<td>2.45(^{b})</td>
<td>86.87</td>
<td>1.73</td>
</tr>
<tr>
<td></td>
<td>DDGS</td>
<td>2.49(^{ab})</td>
<td>87.91</td>
<td>1.72</td>
</tr>
<tr>
<td>Super-dose Phytase (^2)</td>
<td>No DDGS</td>
<td>2.54(^{a})</td>
<td>87.08</td>
<td>1.68</td>
</tr>
<tr>
<td></td>
<td>DDGS</td>
<td>2.50(^{ab})</td>
<td>84.55</td>
<td>1.73</td>
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</table>

ANOVA P-Value

<p>| | | | |</p>
<table>
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</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0.03</td>
<td>0.36</td>
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Fisher’s LSD\(^4\)

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<tbody>
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<td></td>
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<td>0.061</td>
<td>3.667</td>
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SEM\(^4\)

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<td>0.004</td>
<td>16.573</td>
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Marginal Means

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<tr>
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<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>No Phytase (^1)</td>
<td></td>
<td>2.498</td>
<td>87.14</td>
</tr>
<tr>
<td>Standard Phytase (^2)</td>
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<td>2.475</td>
<td>87.39</td>
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<tr>
<td>Super-dose Phytase (^2)</td>
<td></td>
<td>2.525</td>
<td>85.82</td>
</tr>
</tbody>
</table>

Main Effect and Interaction Probabilities

|                |            |                                  |                        |
| Phytase        |            | 0.09                             | 0.45                   | 0.35                             |
| Formulation    |            | 0.16                             | 0.25                   | 0.52                             |
| Phytase X Formulation |      | 0.06                             | 0.33                   | 0.05                             |

\(^1\) Adequate nPP and Ca
\(^2\) Formulation was reduced in calculated nPP by 0.12% and calculated Ca by 0.1%.
Table 6. The Effects of Varying Levels of Phytase and DDGS on Bone Mineralization for Starter, Grower, and Finisher Phases

<table>
<thead>
<tr>
<th>Phytase Inclusion</th>
<th>Formulation</th>
<th>Starter (D1-10)(^3)</th>
<th>Grower (D11-22)(^4)</th>
<th>Finisher (D23-38)(^5)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Mg ash/bird</td>
<td>Mg ash/kg of BW</td>
<td>% Tibia ash/bird</td>
</tr>
<tr>
<td>No Phytase(^1)</td>
<td>No DDGS</td>
<td>523.3(^{ab})</td>
<td>2088.8</td>
<td>15.0</td>
</tr>
<tr>
<td></td>
<td>DDGS</td>
<td>493.7(^7)</td>
<td>1994.0</td>
<td>14.8</td>
</tr>
<tr>
<td></td>
<td></td>
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<td></td>
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<td></td>
<td></td>
<td>508.5</td>
<td>2041.4</td>
<td>14.9(^5)</td>
</tr>
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<td></td>
<td></td>
<td>506.5</td>
<td>2004.1</td>
<td>15.1(^{ab})</td>
</tr>
<tr>
<td></td>
<td></td>
<td>529.0</td>
<td>2025.4</td>
<td>15.6(^a)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>524.4(^7)</td>
<td>2045.6</td>
<td>15.3</td>
</tr>
<tr>
<td></td>
<td>DDGS</td>
<td>505.0(^7)</td>
<td>2001.7</td>
<td>15.1</td>
</tr>
<tr>
<td></td>
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<td></td>
</tr>
</tbody>
</table>

1 Adequate nPP and Ca
2 Formulation was reduced in calculated nPP by 0.12% and calculated Ca by 0.1%.
3 Left tibiae was excised from 6 randomly selected birds per pen
Left tibiae was excised from 2 randomly selected birds per pen.
CHAPTER 3: The Effects of *Spirulina* Algae Inclusion and Conditioning Temperature on Feed Manufacture and Pellet Quality

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Division of Animal and Nutritional Sciences, West Virginia University, Morgantown, West Virginia, 26506

Phone: 304-293-1911

Fax: 304-293-2232
SUMMARY Pellet quality has been shown to influence poultry performance; however, these variables may also influence nutrient availability of diets. Diet formulation and manufacturing techniques dictate pellet quality. Steam conditioning temperature and *Spirulina* algae inclusion has been suggested to be highly influential on pellet quality. The objective of this study was to describe the manufacturing effects of practical broiler diets with varying inclusions of *Spirulina* algae and steam conditioning temperature. Treatments were arranged as a 5 x 3 factorial that varied in algae inclusion (0, 0.5, 1, 5, and 10%) and steam conditioning temperature (74 (165°F), 82 (180°F), and 91°C (195°F)). Each treatment combination was replicated three times across three days of feed manufacture. Treatments were steam conditioned for 10 seconds and extruded through a 4.7 x 38 mm pellet die. Hot pellet temperature was affected by both algae inclusion and conditioning temperature, separately, demonstrating increased hot pellet temperature with either increased algae inclusions or steam conditioning temperature. Pellet mill motor amperage was affected similarly to hot pellet temperature, while electrical energy usage was only different when conditioning temperature was elevated from 74 or 82°C to 91°C. However, production rate was not affected by treatment. Pellet durability analyses revealed interactions between algae inclusion and conditioning temperature (P<0.0001) that demonstrated as algae inclusion and conditioning temperature increased pellet durability increased, but algae inclusion was more beneficial to pellet durability at low conditioning temperature. The combination of algae inclusion and low conditioning temperature may improve pellet quality without subsequent detriment to nutrient availability.

Keywords: feed manufacture, algae, conditioning temperature
DESCRIPTION OF PROBLEM

The benefits associated with pelleting include improved feed handling, decreased ingredient segregation and feed spillage, increased productive energy of poultry, and subsequent improved performance [1]. Currently there are no industry standards for manufacturing pellets. High throughput demands have forced integrators to manufacture feed with lower percentages of intact pellets because of less conditioning exposure, poor quality steam, thin pellet mill die extrusions, or various other measures that increase throughput [2-3].

Various techniques have been utilized to resolve poor pellet quality which may include manipulation of steam conditioning temperature, supplemental fat addition technique, and the use of pellet binders. Past research has demonstrated that high temperature steam conditioning significantly increase pellet durability [4-6]. Although pellet durability improves with high conditioning temperature, there may be a threshold. If pelleting temperatures are too high, then nutrient availability may be compromised. [6].

A practice becoming more popular in feed manufacture is the application of low mixer-added fat with the remaining supplemental fat being applied post-pelleting [5]. Low mixer-added fat may increase frictional heat in the pellet die and potentially denature exogenous feed enzymes and reduce nutrient availability [7]. Previous research has demonstrated that increased mixer-added fat results in decreased pellet quality [7-9], although Wamsley and Moritz reported that this practice can reduce detriment to amino acid digestibility [8]. Fat added at the mixer may contribute to improved nutrient utilization and enzyme efficacy by coating feed particles, decreasing frictional heat, and reducing opportunities for thermal denaturation [10, 11].
Another technique has been the inclusion of pellet binder, both inherent in ingredients and commercially available additives [12]. Although various commercial pellet binders have demonstrated efficacy, increased pellet quality may be somewhat offset by nutrient dilution. Currently, very few nutritive pellet binders are available to the poultry industry [13]. It is plausible that relatively small changes in diet formulations have the potential to alter pellet quality significantly [14]. Evans and cohorts reported that *Spirulina* algae inclusion in diets at a high percentage (21%) decreased pellet production rate and increased hot pellet temperature and pellet durability, suggesting that algae has potential as a pellet binder [15].

The objective of this study was to examine the feed manufacture and pellet binding capabilities of modest inclusions of *Spirulina* algae, a proposed novel pellet binder, along with the known pellet quality improvement strategy of increasing steam conditioning temperature.
MATERIALS AND METHODS

A pelleting study was conducted at the West Virginia University pilot feed mill to determine if *Spirulina* algae possessed pellet binding qualities. Two corn and soybean meal based broiler diets, differing in algae inclusion (0 or 10%), were formulated to be similar in calculated nutrient composition (Table 2), as well as meeting NRC recommendations for broiler chickens [16]. Mineral content, metabolizable energy and digestible amino acids were determined and utilized for diet formulation from a previous research study that utilized the same batch of Spirulina algae (Table 1) [15]. Diets were blended to create three additional diets containing 0.5, 1 and 5% algae, respectively.

A 40 HP California Pellet Mill [17] was used to pellet the five blended diets at three conditioning temperatures (74, 82, and 91°C) creating 15 treatments arranged in a 5 by 3 factorial randomized complete block design. Each treatment was replicated three times across three consecutive days.

A mash sample was collected from the feed screw auger prior to steam conditioning. Multiple samples were collected from each treatment replicate during feed manufacture to determine feed moisture content. Samples were collected from the feed screw auger prior to steam conditioning, from the stream of steam conditioned feed prior to pellet extrusion, and post-cooling as pelleted feed exited the horizontal cooling deck. Cooled, pelleted samples were used to determine pellet durability index (PDI) and modified pellet durability index (mPDI) [18] using a Pfost Tumbler Box, as well as pellet survivability using a New Holmen Pellet Tester (NHPT) [19]. Each durability analysis was run in duplicate.
Measured feed manufacture variables included: hot pellet temperature, total fines produced, percent pellets produced, production rate [20], motor amperage [21], and electrical energy usage [22]. Two representative pellet/fine samples were sifted using a No. 6 Tyler sieve [23] to determine pellet and fine percentages in an approximately 4.5 kg sample. Resulting data was used to calculate percent pellet of the run (Table 4).

**Statistical Analysis**

Variables were analyzed in a 5 (Algae Inclusion) x 3 (Conditioning Temperature) factorial arranged in a randomized complete block design. The experimental unit consisted of one 147.4 kg (325 lbs) batch of experimental diet. All data were statistically analyzed using the GLM procedure of SAS [24]. Alpha was designated as 0.05. Block criterion was day of manufacture. Algae inclusions and conditioning temperature main effects, as well as Algae x Temperature interactions were also estimated. Post ANOVA, treatment means were further compared using Fisher’s least significant difference test.

**RESULTS AND DISCUSSION**

**Pellet Durability**

Pellet durability analyses revealed interactions between algae inclusion and conditioning temperature (P<0.0001) that demonstrated as algae inclusion and conditioning temperature increased pellet durability increased, but algae inclusion was more beneficial to pellet durability at low conditioning temperature (Table 3). The 10% algae diet showed a 30.4 percentage point increase in PDI relative to the 0% algae diet when both diets were conditioned at 74°C.
Differences in PDI were apparent at the highest conditioning temperature, 91˚C, but were less pronounced, only revealing 5 percentage points difference in PDI when considering the aforementioned diets.

Similar interactions were apparent for MPDI (P<0.0001) and NHPT (P<0.0001). When using more aggressive durability assays the differences between dietary treatments were even more pronounced. The NHPT demonstrated a 51 percentage point increase in durability when comparing the 10% algae diet to the 0% algae diet at a conditioning temperature of 74˚C. The authors believe that these aggressive durability values are more indicative to feed attrition associated with conveyance and transport from the feed mill to the feed pan. Moreover, the authors believe that these durability benefits have the potential to assist integrators in obtaining desired pellet quality without the constraints and difficulties of increasing conditioning temperature. Constraints that limit increased conditioning temperature may include ingredient moisture content or geographical climate restrictions that can limit increased steam addition and efficient feed manufacture.

There is ample evidence that high conditioning temperature improves pellet quality [4-6]. Past research conclusions on conditioning temperature and pellet quality support the current study observations. However, high conditioning temperature has also been shown to decrease nutrient digestibility [6], especially amino acid digestibility [5]. The current study demonstrates that the use of algae may improve pellet quality at low conditioning temperatures that may be less detrimental to nutrient digestibility.

*Feed Manufacture*
Electrical Energy Usage (EEU) of the pellet mill was affected by conditioning temperature (P<0.0001) demonstrating that diets conditioned at 91°C required the most EEU while diets conditioned at 74 and 82°C required less amounts of electrical energy for manufacture. Past research has shown decreased EEU at the pellet mill when utilizing formulations with low inclusions of mixer-added fat and high conditioning temperatures [5]. This is most likely due to the increased steam volume necessary to increase temperature producing a lubricating action at the die, thus reducing energy needed to extrude mash [25]. All supplemental fat addition in the current study was added at the mixer, prior to pelleting. The authors speculate that with all supplemental fat being added prior to pelleting coupled with increased conditioning temperatures that the feed moved passed a critical point of lubrication, decreasing frictional force necessary for extrusion, causing feed to slip between the pellet mill rolls and die interface, ultimately reducing pelleting efficiency with increased EEU.

Both algae inclusion and conditioning temperature significantly affected pellet mill motor amperage. Conditioning temperature demonstrated higher motor amperage when diets were conditioned at 91°C compared to 74 and 82°C (P<0.0001). Although apparent through EEU and amperage, the decrease in frictional force at the roll/die interface did not occur at a level that stopped production. Algae inclusion also affected the pellet mill motor amperage (P=0.013) demonstrating the highest pellet mill motor amperage in diet formulations with 10% algae. Diets containing 0% algae required the least amount of motor amperage and all other diet formulations were intermediate. The fine particle composition of the algae likely filled interstitial space in the mash feed and increased frictional force necessary for pelleting. This speculation was supported by the incremental increases in pellet mill motor amperage due to increased algae inclusion.
Algae inclusion and conditioning temperature individually affected hot pellet temperature (Table 4). Algae inclusion affected hot pellet temperature (P<0.0001) demonstrating increased hot pellet temperature for formulations including 10% algae. Formulations including 0, 0.5, and 1% algae revealed decreased hot pellet temperature while the formulation including 5% algae was intermediate. These data support the speculation of algae inclusion increasing friction within the die and energy necessary for pelleting. This increased frictional force and energy required for pelleting diets with increased inclusions of algae can explain higher hot pellet temperatures. Hot pellet temperature was also affected by conditioning temperature (P<0.0001) demonstrating increased hot pellet temperature as conditioning temperature increased.

**Feed Moisture**

Mash moisture was affected by algae inclusion (P<0.0001, Table 4) demonstrating decreasing mash diet moisture as algae inclusion increased. The diet formulation containing 5% algae revealed an elevated mash moisture content similar to the diet formulations containing 0 and 0.5% algae. The authors speculate that the low moisture content of the algae product decreased the overall moisture content of the mash diet as algae inclusion increased (Table 1). Conditioning temperature significantly affected hot mash moisture and feed moisture as would be expected (P<0.0001, 0.0001, respectively).

**CONCLUSIONS AND APPLICATIONS**

1. Spirulina algae inclusion generated the most pronounced pellet durability differences at 74°C steam conditioning temperature.
2. The combined strategy of algae incorporation in diets and low temperature steam conditioning/pelleting may maximize pellet durability and minimize nutrient digestibility detriment.

REFERENCES AND NOTES


17. Master Model Pellet Mill, California Pellet Mill Company (CPM), Crawfordsville, IN.

tumbled in the container, dimensions 5 × 12 × 12 in., with a 2 × 9 in. plate fixed diagonally along the 12 × 12 in. side, for approximately 10 min at 50 rpm. The sample was then sifted again through the No. 6 (ASTM) mm screen, weighed, and the percentage of pellets was calculated by dividing the weight of pellets after tumbling by the weight of pellets before tumbling and then multiplying that value by 100. Modified pellet durability index was similarly measured, with the exception of the addition of five, 13-mm hexagonal bolts to the 500 g of sample in the tumbler. Both analyses are meant to simulate the deleterious effects of transferring and handling the pellets.

19. Pellet quality was assessed one day following production using the New Holmen NHP Portable Pellet Durability Tester, Lignotech USA, INC., Rothschild, WI. 100 g of pellets were placed in the chamber, blown about for 60 seconds by a jet of air, then weighed, giving a direct read of pellet durability. Fines are removed during the blowing process.

20. Instantaneous production rate was calculated by collecting all pellets post extrusion for 1 minute. Feed was weighed and used to calculate production rate in tonnes/hour.

21. Pellet mill motor amperage data was collected by the pellet mill operator from an Amp meter. Reported amperage represents average motor amperage calculated from the respective motor amperage range.

22. Electrical energy usage was collected from a kWh meter. The meter was re-set to read zero prior to pelleting each run. Total electrical energy usage was determined immediately following pellet extrusion.

23. W. S. Tyler Industrial Group, Mentor, OH.


Table 1. Algae\textsuperscript{1} nutrient composition used for diet formulation

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>True Metabolizable Energy (kcal/kg)</td>
<td>2,839</td>
</tr>
<tr>
<td>Moisture\textsuperscript{2} (%)</td>
<td>5.10</td>
</tr>
<tr>
<td>Crude Protein\textsuperscript{2} (%)</td>
<td>76.0</td>
</tr>
<tr>
<td>Crude Fat\textsuperscript{2} (%)</td>
<td>4.95</td>
</tr>
<tr>
<td>Digestible Lysine (%)</td>
<td>2.10</td>
</tr>
<tr>
<td>Digestible Methionine (%)</td>
<td>1.07</td>
</tr>
<tr>
<td>Digestible Cysteine (%)</td>
<td>0.42</td>
</tr>
<tr>
<td>Digestible Threonine (%)</td>
<td>1.97</td>
</tr>
<tr>
<td>Calcium\textsuperscript{2} (%)</td>
<td>1.20</td>
</tr>
<tr>
<td>Sodium\textsuperscript{2} (%)</td>
<td>0.28</td>
</tr>
<tr>
<td>Total Phosphorus\textsuperscript{2} (%)</td>
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</tr>
<tr>
<td>Phytic Acid\textsuperscript{2} (%)</td>
<td>1.01</td>
</tr>
<tr>
<td>Non-Phytate Phosphorus\textsuperscript{2,3} (%)</td>
<td>1.08</td>
</tr>
</tbody>
</table>

\textsuperscript{1} Algae was obtained from Earthrise Nutritionals (Calipatria, CA). This is a full fat product grown in outdoor ponds.
\textsuperscript{2} Analysis was performed by Eurofins Scientific, Des Moines, IA.
\textsuperscript{3} Non-phytate phosphorus = total phosphorus – (0.282 X phytic acid)
Table 2. Broiler diet formulations used to blend together to create 5 experimental diets

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>0% Algae Diet</th>
<th>10% Algae Diet</th>
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<tbody>
<tr>
<td><strong>Corn</strong></td>
<td>54.78</td>
<td>54.63</td>
</tr>
<tr>
<td><strong>Soybean meal</strong></td>
<td>31.66</td>
<td>23.37</td>
</tr>
<tr>
<td><em>Spirulina algae</em></td>
<td>0.00</td>
<td>10.00</td>
</tr>
<tr>
<td><strong>Wheat Middlings</strong></td>
<td>4.93</td>
<td>4.93</td>
</tr>
<tr>
<td><strong>Animal/Veg Fat</strong></td>
<td>4.06</td>
<td>3.07</td>
</tr>
<tr>
<td><strong>Dicalcium phosphate</strong></td>
<td>1.71</td>
<td>1.58</td>
</tr>
<tr>
<td><strong>Limestone</strong></td>
<td>1.38</td>
<td>1.19</td>
</tr>
<tr>
<td><strong>DL-Methionine</strong></td>
<td>0.34</td>
<td>0.28</td>
</tr>
<tr>
<td><strong>Salt</strong></td>
<td>0.31</td>
<td>0.39</td>
</tr>
<tr>
<td><strong>Lysine</strong></td>
<td>0.25</td>
<td>0.27</td>
</tr>
<tr>
<td><strong>Poultry Premix</strong></td>
<td>0.25</td>
<td>0.25</td>
</tr>
<tr>
<td><strong>Sodium Bicarbonate</strong></td>
<td>0.21</td>
<td>0.00</td>
</tr>
<tr>
<td><strong>L-Threonine</strong></td>
<td>0.12</td>
<td>0.05</td>
</tr>
</tbody>
</table>

**Calculated Nutrients**

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>0% Algae Diet</th>
<th>10% Algae Diet</th>
</tr>
</thead>
<tbody>
<tr>
<td>ME (kcal/kg)</td>
<td>3,030.5</td>
<td>3,030.5</td>
</tr>
<tr>
<td>Crude Protein</td>
<td>20.71</td>
<td>22.52</td>
</tr>
<tr>
<td>Dig. Met + Cys</td>
<td>0.89</td>
<td>0.89</td>
</tr>
<tr>
<td>Dig. Threonine</td>
<td>0.77</td>
<td>0.77</td>
</tr>
<tr>
<td>Dig. Lysine</td>
<td>1.18</td>
<td>1.18</td>
</tr>
<tr>
<td>Dig. Sodium</td>
<td>0.20</td>
<td>0.20</td>
</tr>
<tr>
<td>Calcium</td>
<td>0.96</td>
<td>0.96</td>
</tr>
<tr>
<td>Av. Phosphorus</td>
<td>0.45</td>
<td>0.45</td>
</tr>
</tbody>
</table>

1 Diets formulations containing 0 and 10% *Spirulina algae* were blended together based on calculations to create 5 diets containing 0, 0.5, 1, 5, and 10% algae.

2 Supplied per kilogram of diet: 0.02% manganese; 0.02% zinc; 0.01% iron; 0.0025% copper; 0.0003% iodine; 0.00003% selenium; 0.69 mg of folic acid; 386 mg of choline; 6.61 mg of riboflavin; 0.03 mg of biotin; 1.38 mg of vitamin B6; 27.56 mg of niacin; 6.61 mg of pantothenic acid; 2.20 mg of thiamine; 0.83 mg of menadione; 0.01 mg of vitamin B12; 16.53 IU of vitamin E; 2,133 ICU of vitamin D3; and 7,716 IU of vitamin A.
Table 3. Feed Moisture Data\(^1\)

<table>
<thead>
<tr>
<th>Algae Inclusion (%)</th>
<th>Conditioning Temp (˚C)</th>
<th>Mash Moisture (%)</th>
<th>Hot Mash Moisture (%)</th>
<th>Complete Feed Moisture (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0%</td>
<td>74</td>
<td>12.9(^a)</td>
<td>16.0(^{def})</td>
<td>13.5(^g)</td>
</tr>
<tr>
<td></td>
<td>82</td>
<td>12.9(^a)</td>
<td>16.1(^{bcd})</td>
<td>14.4(^{bc})</td>
</tr>
<tr>
<td></td>
<td>91</td>
<td>12.9(^a)</td>
<td>17.0(^{ab})</td>
<td>15.1(^{ab})</td>
</tr>
<tr>
<td>0.5%</td>
<td>74</td>
<td>12.7(^{ab})</td>
<td>15.8(^{def})</td>
<td>13.5(^g)</td>
</tr>
<tr>
<td></td>
<td>82</td>
<td>12.7(^{ab})</td>
<td>16.0(^{bcd})</td>
<td>14.4(^{bcd})</td>
</tr>
<tr>
<td></td>
<td>91</td>
<td>12.7(^{ab})</td>
<td>17.0(^{ab})</td>
<td>15.0(^{bc})</td>
</tr>
<tr>
<td>1%</td>
<td>74</td>
<td>12.5(^{bc})</td>
<td>15.8(^{e})</td>
<td>14.4(^{e})</td>
</tr>
<tr>
<td></td>
<td>82</td>
<td>12.5(^{bc})</td>
<td>15.9(^{def})</td>
<td>14.7(^{abcd})</td>
</tr>
<tr>
<td></td>
<td>91</td>
<td>12.5(^{bc})</td>
<td>17.0(^{ab})</td>
<td>14.9(^{abc})</td>
</tr>
<tr>
<td>5%</td>
<td>74</td>
<td>12.8(^{ab})</td>
<td>15.5(^{e})</td>
<td>14.2(^{de})</td>
</tr>
<tr>
<td></td>
<td>82</td>
<td>12.8(^{ab})</td>
<td>16.5(^{acea})</td>
<td>14.4(^{a})</td>
</tr>
<tr>
<td></td>
<td>91</td>
<td>12.8(^{ab})</td>
<td>16.8(^{abc})</td>
<td>15.0(^{ab})</td>
</tr>
<tr>
<td>10%</td>
<td>74</td>
<td>12.3(^{c})</td>
<td>15.2(^{e})</td>
<td>14.1(^{bcde})</td>
</tr>
<tr>
<td></td>
<td>82</td>
<td>12.3(^{c})</td>
<td>16.3(^{cde})</td>
<td>14.4(^{cde})</td>
</tr>
<tr>
<td></td>
<td>91</td>
<td>12.3(^{c})</td>
<td>17.4(^{a})</td>
<td>15.4(^{a})</td>
</tr>
</tbody>
</table>

ANOVA P-value: 0.0026 0.0014 0.0010
Fisher’s LSD: 0.357 0.9729 0.7907
SEM: 0.123 0.335 0.272

Marginal Means

<table>
<thead>
<tr>
<th>Algae Inclusion (%)</th>
<th>Conditioning Temp (˚C)</th>
<th>Mash Moisture (%)</th>
<th>Hot Mash Moisture (%)</th>
<th>Complete Feed Moisture (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0%</td>
<td>--</td>
<td>12.9(^a)</td>
<td>16.4</td>
<td>14.3</td>
</tr>
<tr>
<td>0.5%</td>
<td>--</td>
<td>12.7(^{ab})</td>
<td>16.2</td>
<td>14.3</td>
</tr>
<tr>
<td>1%</td>
<td>--</td>
<td>12.5(^{bc})</td>
<td>16.3</td>
<td>14.6</td>
</tr>
<tr>
<td>5%</td>
<td>--</td>
<td>12.8(^{a})</td>
<td>16.3</td>
<td>14.5</td>
</tr>
<tr>
<td>10%</td>
<td>--</td>
<td>12.3(^{c})</td>
<td>16.3</td>
<td>14.6</td>
</tr>
</tbody>
</table>

Main Effects and Interaction Probabilities

<table>
<thead>
<tr>
<th>Factor</th>
<th>Probability Value</th>
<th>Fishers LSD</th>
<th>SEM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Algae Inclusion (%)</td>
<td>&lt;0.0001</td>
<td>0.9928</td>
<td>0.3332</td>
</tr>
<tr>
<td>Conditioning Temp</td>
<td>--</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Algae x Temp</td>
<td>--</td>
<td>0.5254</td>
<td>0.4808</td>
</tr>
</tbody>
</table>

\(^1\) A 5 x 3 factorial arrangement of treatments
\(^{a}\) Values within comparisons with different subscripts differ (P<0.05)
Table 4. Pellet Durability and Feed Manufacture Data

<table>
<thead>
<tr>
<th>Algae Inclusion (%)</th>
<th>Conditionin g Temp (°C)</th>
<th>PDI (%)</th>
<th>mPDI (%)</th>
<th>NHPT (%)</th>
<th>Hot Pellet Temp (°C)</th>
<th>Electrical Energy Usage (kWh)</th>
<th>Motor Amperage (A)</th>
<th>Production Rate (tonnes/hr)</th>
<th>Percent Pellet (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0%</td>
<td>74</td>
<td>61&lt;sup&gt;a&lt;/sup&gt;</td>
<td>48&lt;sup&gt;b&lt;/sup&gt;</td>
<td>38&lt;sup&gt;c&lt;/sup&gt;</td>
<td>77&lt;sup&gt;d&lt;/sup&gt;</td>
<td>1.3&lt;sup&gt;e&lt;/sup&gt;</td>
<td>18.5&lt;sup&gt;d&lt;sup&gt;e&lt;/sup&gt;&lt;sup&gt;de&lt;/sup&gt;</td>
<td>0.84</td>
<td>92&lt;sup&gt;f&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>82</td>
<td>74&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>66&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>56&lt;sup&gt;cd&lt;/sup&gt;</td>
<td>82&lt;sup&gt;e&lt;/sup&gt;</td>
<td>1.2&lt;sup&gt;c&lt;/sup&gt;</td>
<td>18.1&lt;sup&gt;e&lt;/sup&gt;</td>
<td>0.83</td>
<td>93&lt;sup&gt;ef&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>91</td>
<td>91&lt;sup&gt;b&lt;/sup&gt;</td>
<td>89&lt;sup&gt;b&lt;/sup&gt;</td>
<td>88&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>90&lt;sup&gt;de&lt;/sup&gt;</td>
<td>1.6&lt;sup&gt;b&lt;/sup&gt;</td>
<td>19.6&lt;sup&gt;de&lt;/sup&gt;</td>
<td>0.80</td>
<td>96&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>0.5%</td>
<td>74</td>
<td>66&lt;sup&gt;d&lt;/sup&gt;</td>
<td>54&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>40&lt;sup&gt;cd&lt;/sup&gt;</td>
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<td>1.3&lt;sup&gt;c&lt;/sup&gt;</td>
<td>18.6&lt;sup&gt;d&lt;/sup&gt;</td>
<td>0.83</td>
<td>92&lt;sup&gt;ef&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
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<td>68&lt;sup&gt;cd&lt;/sup&gt;</td>
<td>58&lt;sup&gt;de&lt;/sup&gt;</td>
<td>82&lt;sup&gt;e&lt;/sup&gt;</td>
<td>1.3&lt;sup&gt;c&lt;/sup&gt;</td>
<td>19.9&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>0.79</td>
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</tr>
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<td>1.8&lt;sup&gt;c&lt;/sup&gt;</td>
<td>18.2&lt;sup&gt;e&lt;/sup&gt;</td>
<td>0.83</td>
<td>93&lt;sup&gt;de&lt;/sup&gt;</td>
</tr>
<tr>
<td>1%</td>
<td>74</td>
<td>70&lt;sup&gt;d&lt;/sup&gt;</td>
<td>60&lt;sup&gt;d&lt;/sup&gt;</td>
<td>47&lt;sup&gt;e&lt;/sup&gt;</td>
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<td>1.4&lt;sup&gt;b&lt;/sup&gt;</td>
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<td>93&lt;sup&gt;de&lt;/sup&gt;</td>
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<td>73&lt;sup&gt;e&lt;/sup&gt;</td>
<td>64&lt;sup&gt;c&lt;/sup&gt;</td>
<td>83&lt;sup&gt;d&lt;/sup&gt;</td>
<td>1.3&lt;sup&gt;c&lt;/sup&gt;</td>
<td>18.8&lt;sup&gt;d&lt;/sup&gt;</td>
<td>0.82</td>
<td>94&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>91</td>
<td>91&lt;sup&gt;b&lt;/sup&gt;</td>
<td>90&lt;sup&gt;d&lt;/sup&gt;</td>
<td>90&lt;sup&gt;c&lt;/sup&gt;</td>
<td>91&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>1.5&lt;sup&gt;c&lt;/sup&gt;</td>
<td>20.0&lt;sup&gt;e&lt;/sup&gt;</td>
<td>0.83</td>
<td>96&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>5%</td>
<td>74</td>
<td>85&lt;sup&gt;d&lt;/sup&gt;</td>
<td>81&lt;sup&gt;d&lt;/sup&gt;</td>
<td>76&lt;sup&gt;c&lt;/sup&gt;</td>
<td>78&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.4&lt;sup&gt;b&lt;/sup&gt;</td>
<td>19.2&lt;sup&gt;e&lt;/sup&gt;</td>
<td>0.83</td>
<td>96&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
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<td>88&lt;sup&gt;c&lt;/sup&gt;</td>
<td>86&lt;sup&gt;bc&lt;/sup&gt;</td>
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</tr>
<tr>
<td></td>
<td>91</td>
<td>94&lt;sup&gt;bc&lt;/sup&gt;</td>
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<td>94&lt;sup&gt;d&lt;/sup&gt;</td>
<td>92&lt;sup&gt;e&lt;/sup&gt;</td>
<td>1.5&lt;sup&gt;c&lt;/sup&gt;</td>
<td>19.9&lt;sup&gt;d&lt;/sup&gt;</td>
<td>0.83</td>
<td>97&lt;sup&gt;ab&lt;/sup&gt;</td>
</tr>
<tr>
<td>10%</td>
<td>74</td>
<td>92&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>90&lt;sup&gt;c&lt;/sup&gt;</td>
<td>89&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>78&lt;sup&gt;de&lt;/sup&gt;</td>
<td>1.5&lt;sup&gt;c&lt;/sup&gt;</td>
<td>19.8&lt;sup&gt;e&lt;/sup&gt;</td>
<td>0.83</td>
<td>98&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>82</td>
<td>93&lt;sup&gt;bc&lt;/sup&gt;</td>
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<td>92&lt;sup&gt;c&lt;/sup&gt;</td>
<td>84&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.4&lt;sup&gt;b&lt;/sup&gt;</td>
<td>18.8&lt;sup&gt;d&lt;/sup&gt;</td>
<td>0.83</td>
<td>98&lt;sup&gt;a&lt;/sup&gt;</td>
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<tr>
<td></td>
<td>91</td>
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<td>95&lt;sup&gt;c&lt;/sup&gt;</td>
<td>93&lt;sup&gt;e&lt;/sup&gt;</td>
<td>1.6&lt;sup&gt;b&lt;/sup&gt;</td>
<td>20.1&lt;sup&gt;e&lt;/sup&gt;</td>
<td>0.82</td>
<td>98&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

ANOVA P-value: <.0001 <.0001 <.0001 <.0001 <.0001 <.0001 <.0001 .4993 <.0001

Fisher’s LSD: 4.4253 5.486 8.2438 1.8168 0.2375 0.815 0.0504 1.249

SEM: 1.527 1.894 2.846 0.627 0.082 0.281 0.017 0.431

Marginal Means

| 0% | 74 | 66 | 60 | 83 | 1.4 | 18.7 | 0.82 | 93<sup>d</sup> |
| 0.5% | 74 | 71 | 63 | 83 | 1.5 | 19.2 | 0.81 | 94<sup>a</sup> |
| 1% | 80 | 74 | 67 | 83 | 1.4 | 19.0 | 0.83 | 94<sup>c</sup> |
| 5% | 90 | 87 | 84 | 84 | 1.4 | 19.3 | 0.84 | 96<sup>b</sup> |
| 10% | 94 | 93 | 92 | 85 | 1.5 | 19.6 | 0.83 | 98<sup>a</sup> |

Main Effects and Interaction Probabilities

Algae <.0001 <.0001 <.0001 <.0001 0.5550 0.0134 0.3423 <.0001
Conditioning Temp <.0001 <.0001 <.0001 <.0001 <.0001 <.0001 0.1706 <.0001
Algae x Temp <.0001 <.0001 <.0001 0.1312 0.3372 0.1478 0.7298 0.0011

<sup>a</sup>Values within comparisons with different subscripts differ (P<0.05).
CIRRICULUM VITAE

John W. Boney

327 Dawson Road, Morgantown, WV 26501
jboney@mix.wvu.edu
(740) 581-0493

Enjoys the challenges associated with working under pressure, within a team or as an individual. Confident with abilities to lead a group and make critical decisions, as necessary.

Goals: To always continue to learn. I hope to obtain research experience, predominantly in non-ruminant nutrition, production and feed manufacture. I plan to continue gaining knowledge of the poultry industry and one day use this knowledge to help integrators supply protein to consumers.

EDUCATION

Degree obtained: Bachelors of Science, Animal and Nutritional Science; May 2013

Undergraduate GPA: 3.38

Expected Degree: Masters of Science, Animal and Food Science

Expected Graduation Date: December 2014

Current GPA: 3.86

EDUCATION HONORS/AWARDS

Graduated from Caldwell High School in 2009

• National Honors Society Member, Caldwell High School Chapter (2008-2009)

Scholarships/Grants Received

• Academic Competitiveness Grant: 2009-2011
• Scholarship Office Award: 2009-2010
• PELL Grant: 2009-2013
• Louretta and Earle Elmore Scholarship: 2012-2013

Awards and Honors
• American FFA Degree Recipient
• Caldwell High School FFA Chapter 2013 Annual Awards Banquet- Keynote Speaker
• National Society of Collegiate Scholars member
• Davis College of Agriculture, Natural Resources, and Design Dean’s List
  ➢ Fall 2009, Spring 2010, Spring 2013, and Fall 2013
• Davis College of Agriculture, Natural resources, and Design President’s List
  ➢ Spring 2014
• WVU Mentorship Program- Mentored by Joel Newman, President of the American Feed Industry Association (AFIA) (October 2014-present)

First Author Publications

Abstracts

J.W. Boney and J.S. Moritz. 2014. HiPhos dose effects in practically formulated diets that vary in ingredient composition on formulation cost and broiler performance. Poult. Sci. (Accepted Abstract (26)).

J.W. Boney, A.E. Lamp, and J.S. Moritz. 2013. The effects of wheat supplementation to corn and soybean meal based diets on manufacture of pellets and subsequent turkey performance. Poult. Sci. (Accepted Abstract (43)).

Co-Author Publications:

Abstracts

R.B. Sellers¹, J.W. Boney², C.McDaniels¹, J.S. Moritz², and K.G.S. Wamsley¹. 2013. Feed form and liquid application method effects on feed augering segregation. Poult. Sci. (Accepted Abstract (67)).

R.B. Sellers¹, J.W. Boney², C. McDaniels¹, J.S. Moritz², and K.G.S. Wamsley¹. 2015. Liquid application method (LAM), feed form (FF), and feed pen location effects on D28-56 Ross x Ross 708 male broiler performance and processing characteristics. (Abstract Pending).

RESEARCH EXPERIENCE

National Meeting Paper Presentations

• 2013 Poultry Science Association (San Diego, CA) (Undergraduate Student)
  ➢ “The effects of wheat supplementation to corn and soybean meal based diets on manufacture of pellets and subsequent turkey performance”
• 2014 Poultry Science Association (Corpus Christi, TX) (Graduate Student)
  ➢ “HiPhos dose effects in practically formulated diets that vary in ingredient composition on formulation cost and broiler performance”

Graduate Teaching Assistant
Teaching Assistant for Poultry Judging Class
  ➢ Roles included instructing classes and selecting top individuals to compete in the national competition. I assisted in taking the poultry judging team to Baton rouge, LA for the national competition, where I was involved in setting up the competition and grading participants score cards.

Teaching Assistant for Poultry Production Laboratory
  ➢ Roles included assisting students as they gained knowledge of poultry operations. I also organized a class field trip to Pilgrim’s Pride, Inc. where students could obtain first-hand knowledge of the operations of a vertically integrated operation.

Graduate Research Assistant

• WVU Pilot Feed Mill Manager (May 2012- present)
• Led a contract study with DSM Nutritional Products, Inc. utilizing a phytase enzyme product that was included in varying inclusions in diets that varied in ingredient composition. (November 2013- January 2014)
• Led a pelleting trail utilizing Spirulina algae to determine its pellet binding qualities. (August 2014)
• Attended the National Poultry Science Association annual meeting
  o 2012- Athens, GA
  o 2013- San Diego, CA
  o 2014- Corpus Christi, TX
• Attended the Biomass Utilization for Green Materials and Energy Conference (September 2014)
• Pendleton County Poultry Judging Team Training (2014)
  o Students were to represent West Virginia at the national 4-H poultry judging competition
• Assisted with a traveling poultry processing demonstration in various locations throughout the state of West Virginia (2013-present)
• Led numerous WVU Animal Science Farm tours
  o Preston County Elementary
  o Minnesota FFA chapter
  o Cub Scout Pack 44
  o WVU Residence Hall (Fieldcrest Hall)
  o Values and Ethics Class
  o Charleston, WV 5th grade class
  o Dodridge County FFA chapter
  o Clay County High School
• Urban Agriculture Conference (2014)
• Mason-Dixon Elementary School
  o Career day participant
• International Poultry Exposition, Atlanta, GA
  o 2012, 2014
• Hands-on Ag Day- 2012, 2014
  o North Marion High School
• Ag in the Classroom (2012)
• Poultry Festival (Moorefield, WV) (2012-2014)
  o Assisted with conducting an annual poultry judging competition
  o Attended annual business meetings
  o Attended summer educational meetings
• Family Farm Day poultry display (2011-2014)
• State and County Fair Poultry Displays
  o WV State Fair
  o Monongalia County Fair
• Monongalia County Fair- Poultry Exhibit Judge
  o 2013-2014
• Cub Scout Pack 44 Activities- Volunteer
  o Fire, Axe, and Wildlife Conservation Skills Demonstration
  o Merit Badge University
  o Annual Parents Meeting/ Cookout
• West Virginia State FFA Poultry CDE Competition (2012-2014)
  o Created classes and coordinated the competitions
• A-STEM of Mind Cooking with Math Camp
• Organic Field Days (2012-2013)
  o Organic Poultry Demonstration
  o Prepare/Serve Food
• Davis College Welcome Back BBQ
  o Prepare/Deliver Food

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**EXPERIENCE**

**WVU Poultry Judging Team Member** 2012-2013

• University of Arkansas
  ▪ 19th overall
• Louisiana State University
  ▪ 12th overall

**Federal Work Study** 2011-2012

• Introduction into feed manufacture, poultry rearing, and applied poultry research at West Virginia University

**Aviagen Turkey, Inc. Summer Internship Program** 2011

• Overview of commercial turkey breeding operations
• 8 week program

**Shoney’s of Morgantown** 2011

• Temporary/College Employment
• Bar quality coordinator

**On and Offshore Drilling, Inc.** 2010

• Temporary/Summer Employment
• Lawn care and maintenance
Singer’s Auto Sales  

2008-2011

- Temporary/Summer Employment
- Detailed vehicles
- Facilities maintenance
- Farm hand

SKILLS

- Feed mill management
- Ingredient sourcing and ordering
- Proficient in Window’s Microsoft Programs
- Poultry judging, handling, and husbandry
- Feed manufacture
- Diet formulation
- Cecectomy surgery
- Precision-feeding
- Tibia extraction
- Ileum extraction
- Experience using SAS software
- Pellet durability analysis
- Ether extraction using Soxhlet apparatus
- Poultry vaccination experience
- Commercial turkey hatchery experience
- Commercial turkey rearing experience
- Commercial turkey artificial insemination experience
- Commercial pedigree turkey selection experience
- Agricultural/Animal Specific Undergraduate courses taken:
  - Intro to Animal Science 150
  - Principles of Animal Science 251
  - Poultry Production/Lab 367/369
  - Equine Hoof and Limb 343
  - Animal Nutrition 260
  - Intro to Animal Physiology 301
  - Livestock Evaluation 338
  - Applied Nutrition 1 & 2/Lab 361&362/393
  - Poultry Evaluation 338

Graduate Courses:

- Intro to Biochemistry 410
- Statistics 511 & 512
- Grants and Grantsmanship 593
- Nutritional Biochemistry 512
- Nutrition and Disease Prevention 614
- Leadership Essentials 594