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Final Report

Field Study to Validate Research Results on Phosphorus (P) Requirements and use of Feed Additives in Broilers

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Executive Summary

Three projects were conducted to fulfill the objectives of this grant, after the initial protocol calling for demonstration trial type research was postponed indefinitely by industry partners.

In the first project, implementation of lower phosphorus diet levels in the presence of phytase was done on farms in Delmarva and changes in litter phosphorus and water-soluble phosphorus were determined. The amount of phosphorus fed per pound of live bird produced was decreased by 14%, when looking across all poultry meat production in Delmarva (including broilers, roasters and small birds), when phytase was used. This decrease in fed phosphorus in the presence of diet phytase was associated with a decrease in total phosphorus in litter of 26.9%. Broilers were fed 19.6% less phosphorus per pound of live weight produced. This reduction in phosphorus paired with the use of phytase resulted in a decrease in litter phosphorus of 31.5%. Further decreases are possible and are needed to minimize increases in water-soluble phosphorus and further decrease fed and excreted phosphorus. Work will continue in the fall of 2005 and spring of 2006 with three industry partners.

The second project focused on answering the question industry always poses when phosphorus-feeding levels are proposed to them. Are there losses at processing due to bone breakage when these phosphorus levels are implemented? A large-scale study was done, with full implementation of battery generated phosphorus levels and phytase and 25-hydroxycholecalciferol efficacies, to quantify under industry type conditions the impact on performance and processing yields and losses. Once corrected for any impact of diet on body weight (which was not significant), phosphorus consumed, versus industry phosphorus consumption, was decreased by 13% when birds were fed at requirements and by 23% when birds were fed the phytase containing diet formulated at phosphorus requirements minus the phosphorus phytase makes available in the diet (spares). Percent bone ash was not affected by diet and there were no effects of diet on incidence of broken bones or on bruised parts at processing or on carcass yields. Given the high replication and the fact that this work was done at a commercial processing plant using commercial standard operating procedures, the question asked by industry can be answered. There were no measurable losses at processing when the research generated phosphorus requirement levels were implemented or when phytase was correctly added to the diet.

The third project sought to provide factual information on nitrogen excretion by broilers on an industry diet, based on a balance trial. A prediction equation was developed for nitrogen excretion and 43 days old male broilers (2072.99 grams) were found to have an average nitrogen content in the carcass of 3.1% and consumed an average of 113.7 grams of nitrogen. These birds excreted an average of 893.57 g of dry matter, 39.92 g of nitrogen and 11.94 g of phosphorus.

Demonstration type research is scheduled to continue. A project is ongoing on diet, performance and litter work in commercial houses and a demonstration trial is planned for the spring of 2006 where lower phosphorus levels will be tested.

The three projects conducted for this grant have demonstrated that:

1. Reductions in broiler-litter phosphorus content have occurred since the start of the use of phytase in diets but further decreases are possible and needed to minimize increases in water-soluble phosphorus. This decrease has been associated with a decrease in fed phosphorus in the presence of phytase of 10%.
2. A decrease of 23% in fed phosphorus vs. industry average can be implemented without having negative effects on performance or on processing yields. A safety margin needs to be implemented under commercial conditions but decreases of at least 18% are warranted when phytase is used.
3. Previously calculated values for phosphorus and nitrogen excretion in broilers, as published by ASAE (American Society of Agricultural Engineers) and as used to establish baselines for regulations by EPA, grossly overestimate excretion of nitrogen and phosphorus by today's broilers and thus no longer apply. New values have been published by ASAE in 2005 for all species and broiler values are based on this work.

Summary of the Proposed Project As Submitted:

This research project entailed working with poultry companies on the Eastern Shore to validate, under commercial conditions, research results obtained under controlled conditions at the University of Maryland. These research results were obtained under battery conditions in Dr. Angel's lab on work funded by the Maryland Department of the Environment (1999-2000).

Six paired broiler houses per company were proposed to be used. One house within each farm was be fed the diet that the company was currently feeding broilers on Delmarva. The second house within each paired house system was to be fed diets based on the results from Dr. Angel's research. Each house within a farm was to contain the same number of birds. Mortality, number of birds placed, feed consumption, number of birds sent to the processing plant, and final weight of birds were to be determined for each house. A total of 75 birds (taken at random) from each house were to be sampled for analytical work. Feed samples were to be taken of all feeds fed and the amount of each feed fed recorded.

Feed was to have been analyzed for dry matter, nitrogen (N) and phorphorus (P). Animal Care and Use Committee approval was obtained from the University of Maryland before the work started. Birds would be taken to the USDA-BARC facility for Dual X-Ray absorptiometry for total mineral and N content analysis. Birds would then go to Dr. Angel's lab for sample preparation. Individual birds were to be ground, freeze dried, re-ground and analyzed for N, moisture, and P.

By knowing the amount of N and P that was consumed by the flock and the amount of N and P in the bird at processing, the amount of N and P that was excreted could be determined. This will allow a determination to be made of the impact of the experimental feed management system on decreasing P excretion. Also of importance would be that this work would allow the investigator to prove to commercial integrators how the proposed feed management system would work when they implemented it under commercial conditions. The N numbers provide a very important starting point in looking ahead to ammonia and N emission work. Today there are no good numbers on N excretion from broilers under commercial systems (that include litter N and ammonia N). These numbers could provide a total excreted N that can be used later on to back calculate ammonia N when litter N is known.

From the above, the potential decrease in litter P could be high when switching from commercial average use levels to University of Maryland recommended levels. This change is important to document as compared to actual specific broiler integrator fed levels to show the broiler companies that these changes have an impact and that these changes do not have a negative impact on performance. This is key in getting integrators on the Delmarva Peninsula to adapt new technologies that can substantially decrease litter P.

Table 1. Proposed feed management system

Growth phase	Starter	Grower	Finisher	Withdrawal
Age, days	0-18	18-32	32-42	42-50
Avg Feed Consumed, lb/bird	1.8	3.0	2.7	2.8
Commercial*				
Non-phytate P, %	0.45	0.35	0.30	0.30
Total P, %	*0.72*	*0.61*	*0.55*	*0.55*
UMD Recommended**				
Non-phytate P, %	0.35	0.23	0.15	0.12
Total P, %	*0.61*	*0.49*	*0.41*	*0.37*

Average consumed per bird is based on published average consumption numbers for 2000 (Agri Stats, Ft. Wayne, IN).
*Commercial levels are without phytase and as reported by Agri Stats 2000 (Ft. Wayne, IN). The actual P fed for commercial feed will be determined during the trial, by the company, when diet analyses are done.
**Recommended levels are based on research results from R. Angel's lab (Angel and coworkers published in Poultry Science Journal Supplement 2000, and 2001). These numbers are based on diets with 70ug of 25-hydroxycholecalciferol/kg of diet (from HI-D, Monsanto) and 500 FTU of phytase/kg of diet.

Changes in Project Methodology and Causes for Change

The objective of the work that was to be done for this grant was to demonstrate to industry that reductions in dietary phosphorus could be achieved under commercial conditions without any losses in productivity.

Initially the focus was on demonstration trial type work to get industry to adapt some of the technology that had been developed under controlled battery research conditions. At the time the project was to start, that is within a week of the starting date, industry put a hold on the project due to what appeared to be an outbreak of coccidiosis that they had been unable to control through their normal coccidiostat programs. Given this, their focus turned towards solving that problem and the phosphorus project was delayed. Given this delay and no certainty that the project as it had been conceived could be started within the time frame of the grant, three projects were developed to meet the objectives of the grant.

Project 1: Limited implementation work with phosphorus and phytase that allowed for documentation of actual litter changes associated with diet changes under field conditions.

Project 2: A processing study to determine the impact of implementing battery trial generated phosphorus requirements and feed additive efficacies on processing yields and losses.

Project 3: A balance trial to document actual nitrogen excretions in broilers fed industry diets.

Project 1: Determination of performance and litter phosphorus changes associated with inclusion of phytase and lower phosphorus diet concentrations

Methods

The target was to sample 50 production houses prior (Pre) to the implementation of dietary phytase and lower phosphorus concentrations and post (Post) implementation of dietary phytase and lower phosphorus concentrations. Houses were selected such that all bird types were represented, but primarily one bird type was grown within the house within a phase. Bird types represented were roasters, straight run (broilers), and small birds. Selected houses had a minimum of 10 flocks grown in them without litter removal. All performance information, diet phosphorus and phytase levels were documented.

At the time of sampling, the houses were empty but the "cake" from the last flock was still in place. Samples were taken from selected areas within the house in a specific manner. The house was divided into 2 areas – the brooding area and the rest of the house. A total of 30 sub-samples were taken throughout the house (12 sub-samples from the brooding area and 18 sub-samples from the rest of the house). Sub-samples within the brooding area were taken as follows: five samples at random close (within 12 inches) (but not under) feeders and waterers and seven random samples in other areas. The samples within the rest of the house (non-brooding areas) were taken as follows: eight samples at random close (within 12 inches) (but not under) feeders and waterers and 10 random samples in other areas. Sub-samples were taken with a trowel and care was taken to collect litter to the depth where litter removal occurs and such that no soil was taken. Sub-samples within a house were placed into a five gallon bucket. Once all the litter in the house had been collected, the litter was mixed well within the bucket then poured onto a large tarp and again mixed well. Litter was spread out thinly on the tarp and squared off into 32 quadrilles. Sub-samples were taken from each quadrille and mixed well, and this represented the final sample to be analyzed. Samples were stored frozen until analyzed.

Litter samples were dried in a force draft oven at 50^{o}C for dry matter determination (AOAC, 1990). After drying, samples were ground to pass through a 1 mm screen and stored frozen until analysis. Samples were analyzed for phosphorus (Heinonen and Lahti, 1981), water-soluble phosphorus (Self-Davis and Moore, 2000) and nitrogen (AOAC, 1990).

The data were analyzed as a factorial experiment with two phases (pre-phytase and low phosphorus and post-phytase and low phosphorus), and with three bird types (roasters, small birds and straight run (broilers)). Pairwise means comparisons were performed using Tukey's method (Tukey, 1991) to control experiment-wise error rate. All analyses were done using SAS Version 8.2 (SAS Institute, 1999).

Results

It was important in this study that houses with different types of production be included since even though broiler production remains the highest on Delmarva, roaster and small birds together represent at least 40% of the poultry meat production on Delmarva. In the Post phase, phytase was added at 500 U phytase/kg diet and diets analyzed between 225 and 1018 U phytase/kg with an average analyzed value of 535 (standard deviation 235) U phytase/ kg diet. Results are presented in Table 2. In both phases, regardless of type of bird produced, all houses had at least 10 flocks grown in them before samples were taken. There were no fundamental differences in animal productivity between the phases. The amount of phosphorus fed per pound of live bird produced was decreased by 14% across all types of birds, in the presence of phytase, resulting in a decrease in total phosphorus in litter of 26.9%. Broilers were fed 19.6% less phosphorus per pound of live weight produced. This reduction in phosphorus paired with the use of phytase resulted in a decrease in litter of 31.6%.

Table 2. Effect of dietary inclusion of phytase and lower phosphorus (P) concentrations on performance of roasters, broilers and small birds and on litter dry matter (DM), nitrogen (N), P and water soluble P (WSP) concentrations expressed on a dry matter basis

		Performance measures[1]				Litter measures[2]				
Phase[3]	Bird Type[4]	Days[5]	Bird wt, lb[6]	Feed to gain	P (g)/ lb bird[7]	P	WSP	WSP/ P	DM	N
						------------------Percent------------------				
Pre	Broiler	458[ab]	5.37[b]	2.00[b]	5.96[b]	2.35[a]	0.22[c]	9.23[b]	75.0[ab]	4.58[a]
	Roaster	503[b]	6.18[a]	2.13[a]	6.46[a]	2.37[a]	0.23[c]	9.82[b]	73.3[b]	4.38[ab]
	SB	551[ab]	4.54[d]	2.01[b]	6.08[b]	2.34[a]	0.24[c]	10.34[b]	72.7[b]	3.84[c]
Post	Broiler	479[b]	5.00[c]	1.92[c]	4.79[c]	1.61[c]	0.40[b]	24.36[b]	75.0[ab]	4.21[bc]
	Roaster	617[a]	6.17[a]	2.09[a]	6.03[b]	1.87[b]	0.51[a]	27.61[b]	76.2[a]	4.64[a]
	SB	458[b]	4.44[d]	1.97[bc]	5.09[c]	1.69[bc]	0.46[ab]	27.22[b]	73.4[b]	3.92[c]
SEM[8]		24.66	0.08	0.018	0.085	0.05	0.026	1.020	0.74	0.162
P Value		P<0.01	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01
Main effect means										
Phase	Pre	537	5.37	2.05	6.17	2.35	0.23	9.79	73.6	4.27
	Post	518	5.21	1.99	5.30	1.72	0.46	26.40	75.1	4.26
Type	Broiler	504[b]	5.18[b]	1.99[b]	5.38[b]	1.97[b]	0.31[b]	16.80[b]	75.0[a]	4.40[a]
	Roaster	560[a]	6.18[a]	2.11[a]	6.25[a]	2.12[a]	0.37[a]	18.72[a]	75.0[a]	4.51[a]
	SB	519[b]	4.50[c]	1.96[b]	5.59[b]	2.01[ab]	0.35[ab]	18.78[a]	73.1[b]	3.88[b]
Main effect P values										
Phase		0.323	0.017	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01	0.014	0.904
Type		0.045	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01	0.050	0.019	P<0.01
Phase x type		P<0.01	0.046	0.272	0.047	0.042	0.026	0.284	0.013	0.065

[1] Performance measures for the whole house or complex. [2] Litter measures by house. [3] Phase is either pre or post implementation of dietary phytase and lower diet phosphorus concentrations. [4] Bird type is either broiler (straight run), roaster, or small bird (SB). There were 23 broiler houses sampled in the PRE phase and 18 in the POST phase. There were 18 roaster houses sampled in the PRE phase and 26 in the POST Phase. There were eight small bird houses sampled in the PRE phase and 10 in the post phase. [5] Days is the number of days the house had birds in it during the phase. [6] Average weight of birds produced in the house within the phase. [7] Grams of phosphorus fed (disappeared) per lb of live weight produced in the phase within the house. [8] Weighted average of the standard error of the means.
[a-d] Means within the same column with different superscript letter differ (P<0.05).

Unfortunately the amount of water-soluble phosphorus doubled, when phytase was used across all types of birds produced. Litter from houses that housed roasters contained the highest water-soluble phosphorus both in proportional increase and in absolute terms. This was probably due to an increase in phosphorus fed (Table 3) to roasters over time after the addition of phytase due to an increased incidence of leg problems.

Thus, even though at the start of the POST phase all diets contained an average of 0.1% less phosphorus than in the PRE phase, the roaster program returned to PRE- phosphorus levels while phytase was left in the diet. It is unclear why the increase in leg problems occurred as there was no clear indication that this was due to a phosphorus deficiency.

Table 3. Dietary phosphorus levels for the Pre and Post phases

Type of bird	Phase[1]	Starter	Grower	Finisher	Withdrawal
		-----------Formulated total phosphorus, %-------			
Broilers	Pre	0.73	0.67	0.60	
	Post	0.63	0.57	0.48	
Roasters	Pre	0.73	0.68	0.66	0.61
	Start of Post	0.63	0.57	0.56	0.48
	End of Post	0.72	0.67	0.64	0.56
Small Birds	Pre	0.73	0.66	0.60	
	Post	0.63	0.55	0.50	

[1]Phase is either pre or post implementation of dietary phytase and lower diet phosphorus concentrations.

Discussion

Based on worst-case scenario assumptions, where diets contained no meat and bone meal, the broiler Pre non-phytin phosphorus concentrations would have been 0.46, 0.41, and 0.35% in the starter, grower, and withdrawal diets. These concentrations are very close to National Research Council (NRC, 1994) recommended levels for broilers. Implementation of phytase was done per manufacturer's recommendations that suggest a reduction of 0.1% phosphorus when phytase is added, resulting in Post non-phytin phosphorus concentrations of 0.36, 0.31, and 0.25% in the starter, grower, and withdrawal diets. These levels, at least in the finisher/withdrawal phase are still above research requirements of 0.23 and 0.17 % non-phytin phosphorus (Angel et al 2000a and b, 2001; and Dhandu and Angel, 2003). Concentrations of phosphorus used were still, at least in some phases, above the bird's needs for phosphorus. The use of phytase in diets formulated above the bird's needs results in release of phosphate groups from phytin above what the animal needs. This leads to increased available phosphorus being excreted by the bird and increases in water-soluble phosphorus.

Under commercial conditions, some margin of safety must exist, given that not all factors are known. That is, the exact phosphorus and non-phytin phosphorus content of every batch of grain or of minerals is not known at the time the feed is formulated. Given this, formulation systems rely on average nutrient concentrations of ingredients. This leads to the need for implementation of safety margins in accordance with the accuracy of each system. Accuracy of mixing is also not 100%, another factor requiring safety margins. Because safety margins are needed, commercial diet phosphorus concentrations will always be above requirements but not to the degree seen in this study. Further decreases in diet and thus litter phosphorus are possible and will continue to be pursued in similar work ongoing with industry.

Project 2: A processing study to determine the impact of implementing battery trial generated phosphorus requirements and feed additive efficacies on processing yields and losses.

A study was conducted with industry, where broilers were fed per industry standards as well as with low phosphorus, phytase (PHY) and 25-hydroxycholecalciferol (25D) and taken to a commercial processing plant to determine impact, under commercial conditions and with industry (using a commercial processing plant and its employees), of full implementation of feed PHY, phosphorus and 25D on processing yields and losses. This has been one of the consistent concerns industry has had in the implementation of lower diet phosphorus concentrations.

Methods

Performance

All procedures used in the broiler chicken experiments were approved by the institutional Animal Care and Use Committee. A floor pen experiment was conducted using male Ross 308 broilers obtained from a commercial hatchery on day of hatch. Fifty-six broilers were randomly allocated to each of 55 pens ($0.074m^2$/bird). A four-phase feeding program was employed: starter (St: hatch to 18 d), grower (Gr; 18 to 32 d), finisher (Fn; 32 to 42 d) and withdrawal (Wd; 42 to 49 d). Birds were fed corn-soybean based diets formulated to meet or exceed the National Research Councils (NRC, 1994) nutrient recommendations, except for nPP (non-phytin phosphorus) and calcium. Basal diet formulated nPP is given in Table 4. Briefly, six dietary treatments were tested: Industry, AgriStast (2000) nPP recommendations; research recommendations, nPP recommendations (Angel et al., 2000a, b, 2001; Dhandu and Angel, 2003); RR+PHY, UMD nPP concentrations reduced by 0.064% nPP plus 600 U units (U) PHY/kg diet; UMD+PHY+25D, UMD nPP concentrations reduced by 0.090% nPP plus 600 U PHY and 70 ug 25D/kg diet; C+PHY, mimicked industry practice of reducing NRC (1994) concentrations by 0.1% when PHY is added; and Negative Control (NC) that had 90% UMD nPP concentrations (Table 3). One phytase U is defined as the amount of enzyme required to liberate 1μmol of inorganic P from 1.5 mM Na phytate at pH 5.5 and 37º C (Engelen et al., 1994). The PHY used was Ronozyme P (CT) and the 25D was HI-D (DSM Nutritional Products, Basel, Switzerland).

In previous work, when sparing effects of PHY and 25D were determined (Angel et al., 2001), diets were fed in mash form. A preliminary study to determine losses of PHY activity due to pelleting was conducted at the feed mill and pellet mill (California Pellet Mill California Pellet Mill Co., Merrimack, NH, Model # 3016-4, 8 ton per hour feed production capacity) where the experimental diets were to be made. This preliminary study was run at the conditions normally used at this plant (2 ton run, 82.2^{o}C and 20 second conditioner temperature and time) with a pelleting rate of 22 minutes per ton, average pellet temperature taken at pellet die exit was 82.1^{o}C (standard deviation 3). The results of this preliminary study indicated that pelleting at 82^{o}C resulted in, on average, 15% (standard deviation 2) loss of activity of the PHY used in these experiments. Therefore, the concentration of PHY added to the diets was increased by 15% to 690 U PHY/kg of diet, so that the desired concentrations of 600 U PHY/kg diet could be met in the pelleted feed. Formulated diet calcium concentrations were 0.91, 0.81, 0.71 and 0.61% for the St, Gr, Fn and WD phases based on previous research (Angel et al., 2000a and b; Ling et al., 2000: Dhandu and Angel, 2003).

Fluorescent light provided light intensity of 10 to 20 lux for the first two weks and 5 to 10 lux, thereafter, for the following daily photoperiods: hatch to 4 d, 24 h light (L); 5 to 12 d, 14 h L; 13 to 20 d, 16 h L; 21 to 30 d, 20 h L; 31 to 42 d, 22 h L; 43 to 49 d, 23 h L. A coccidiostat (Deccox®, Alpharma Inc., One Executive Drive, Fort Lee, NJ 07024) was included at 0.05% in all diets except those of the withdrawal phase. Birds were provided ad libitum access to feed and water. Mortality was checked twice daily and the body weight of dead chickens was used to correct body weight gain, feed consumption and feed to gain ratio.

High and low phosphorus basal diets, that met or exceeded NRC (1994) recommendations for all nutrients except nPP and calcium, were formulated and mixed. Basal diets were analyzed for calcium by atomic absorption spectroscopy (Perkin Elmer, 1992), phosphorus (Heinonen and Lahti, 1981), phytin phosphorus (Rounds and Nielsen, 1993 as modified by Newkirk and Classen, 1998), and nPP was determined. Experimental diets were prepared by mixing high and low phosphorus basal diets in appropriate ratios, based on analyzed values, to obtain desired concentrations of nPP. A PHY premix was made for each phase with a small amount of low phosphorus basal. For RR+PHY+25D treatment diets, 25D was added to the PHY premix. Basal diets were analyzed for crude protein, moisture and ether extract (AOAC, 1990). Dietary PHY in the commercial product, as well as in the diets, was analyzed according to the procedures of Engelen et al. (1994) and 25D in diets was determined according to the procedure described by Hollis et al. (1993).

Processing

At the end of the experiment, 22 birds per pen were randomly selected, weighed and wing-banded for processing plant work. These 22 birds per pen were considered as an experimental unit for the processing work. After birds were wing-banded and weighed, they were placed in one common area within the broiler house, with only water available (feed withdrawal period, 5 to 7 hours) to better simulate whole house catching and to maximize mixing of birds from different pens and treatments, thus minimizing catching pen effects. A commercial catching crew caught the wing-banded birds and transported them to a commercial processing plant 40 miles away. Birds were processed at the end of a regular shift in the processing plant (Allen's Family Foods, Inc., processing plant at Hurlock, MD) to avoid mixing of experimental with commercial broilers. The line speed was 93 birds per minute. Carcasses were removed from the line just before entry into the chill tank. Broiler chickens were subjected to standard processing procedures (stunning, bleeding, three stage scalding, picking, mechanical evisceration and normal inspection). Plant as well as federal quality control personnel inspected all birds for defects. Plant personnel condemned whole carcasses and removed parts for discarding, employing normal commercial standards. The only exception was that the wing-banded wing, if defective, was not removed so that bird identification would not be lost. If the wing was defective (broken or heavily bruised), it was noted upon visual inspection and broken wings were considered to have been removed during normal processing and inspection for the purpose of calculating yield.

Hot carcass weight was recorded and used to determine dressing percentage. The hot carcasses were examined for bruises by area [breast, legs (thigh and drumstick) and wings] and for broken legs (thigh and drumstick) and wings. Plant personnel then de-boned carcasses to obtain breast meat (both pectoralis major and minor) with skin, legs (thigh and drumstick), wings and barrel (rib cage and back bone). All parts were saved and weighed to determine percent parts yield. Right tibia and femur from all processed birds were then removed and dry de-fatted ash wt and ash percent were determined (AOAC, 1990).

The experimental design was a randomized complete block with unequal replication. Pen was the experimental unit. The visual observation data (bruised or broken back, breast, wings and legs) were transformed (arc sine square root) to meet the assumptions of ANOVA (Sokal and Rohlf, 1996). Data presented in tables are the arc sine mean values that were back transformed. If the overall effect of treatment was significant ($P \leq 0.05$) in the model, then differences between individual Trt means were separated by LSD (least square difference) test (Sokal and Rohlf, 1996) using SAS (1999).

Table 4. Formulated non-phytin phosphorus concentrations in the starter (hatch to 18 days), grower (18 to 32 days), finisher (32 to 42 days), and withdrawal (42 to 49 days) phases

Treatments[1]	Starter	Grower	Finisher	Withdrawal
	----------% non-phytin phosphorus--------			
Industry	0.45	0.35	0.35	0.30
Research Requirements (RR)	0.45	0.31	0.23	0.18
RR+PHY	0.39	0.25	0.17	0.12
RR+PHY+25D	0.36	0.23	0.14	0.13
Industry+PHY	0.35	0.25	0.25	0.20
Negative Control	0.41	0.28	0.16	0.10

[1] Treatments were Industry (Agri Stats 2000), nPP research requirements (RR) based on work by Angel et al (2000 a and b, 2001) and Dhandu and Angel (2003); RR+PHY RR nPP concentrations minus 0.064% and 600 U phytase/kg diet; RR+PHY+25hydroxycholecalciferol (25D), UMD nPP concentrations minus 0.09% plus 600 U phytase/kg diet and 70 ug 25-hydroxycholecalciferol/kg diet; Industry+PHY, as industry nPP concentrations minus 0.1% plus 600 u phytase/kg diet; and negative control 90% or lower nPP versus RR concentrations.

Results

Performance results are presented in Table 5. All experimental treatments except the RR+PHY+25D and the negative control performed similarly. The negative control treatment had the lowest performance while the RR+PHY+25D treatment had smaller birds than industry but better feed to gain ratios. Once corrected for any impact of diet on body weight, phosphorus consumed, versus industry phosphorus consumption, was decreased by 13% when birds were fed at requirements, and by 23% when birds were fed the RR+PHY diet.

Percent ash was not affected by diet either for the femur or tibia except in the negative control (Table 6). There were no effects of diet on incidence of broken bones or bruised parts at processing (Table 7) or on carcass yields (Table 8).

Table 5. Cumulative (hatch to 49 d) performance and phosphorus consumed

Treatment[1]	49 d BW[2]	Feed intake	Feed to gain	Phosphorus Consumed[3]
	---------- kg/bird ----------		kg/kg	g/ lb live bird
Industry	2.962[a4]	5.597[a]	1.93[bc]	4.98[a]
Research Requirements (RR)	2.935[ab]	5.605[a]	1.94[ab]	4.33[b]
RR+PHY	2.984[a]	5.550[ab]	1.90[c]	3.85[c]
RR+PHY+25D	2.883[b]	5.454[b]	1.92[ab]	3.57[e]
Industry+PHY	2.932[ab]	5.504[ab]	1.91[bc]	4.27[b]
Negative Control	2.861[c]	5.470[b]	1.95[a]	3.91[d]
SEM[5]	0.0528	0.0569	0.023	0.126
P Value	0.0103	0.0084	0.0021	0.0001

[1] See Table 3 for treatment details. [2] Body weight (BW). [3] Based on feed consumed analyzed diet phosphorus concentrations .[4] Values are means of 9 replicate pens of 56 broilers/pen, except for the negative control treatment that had 10 replicate pens. [5] Weighted average of the standard error of the means.
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[a-d] Values in a column and an experiment with different superscript letters differ ($P \leq 0.05$).

Table 6. Bone measurements of 49–d-old broilers processed in a commercial processing plant

Treatment[1]	----------- Femur[2] ---------			----------- Tibia[2] ----------		
	wt, g	ash, g	ash, %	wt, g	ash, g	ash, %
Industry	4.97[a3]	2.31[a]	46.51[a]	6.68[ab]	3.29[a]	49.25[a]
Research Requirements (RR)	4.83[ab]	2.22[b]	46.16[a]	6.54[ab]	3.19[ab]	48.69[a]
RR+PHY	4.94[a]	2.26[ab]	45.89[a]	6.75[a]	3.27[a]	48.53[a]
RR+PHY+25D	4.84[ab]	2.19[b]	46.15[a]	6.42[b]	3.12[b]	48.74[a]
Industry+PHY	4.80[bc]	2.23[ab]	46.53[a]	6.45[b]	3.16[b]	49.13[a]
Negative Control	4.44[c]	1.93[c]	43.35[b]	6.07[c]	2.80[c]	46.11[b]
SEM[4]	0.101	0.033	0.421	0.092	0.045	0.257
P Value	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01

[1] See Table 3 for treatment details. [2] Dry de-fatted femur or tibia weight and respective ash weight and ash percentage. [3] Values are means of nine replicate pens except for the negative control treatment that had 10 replicate pens; 22 birds per pen were randomly selected for processing. [4] Weighted average of the standard error of the means. .

[a-c] Values in a column with different superscript letters differ ($P \leq 0.05$).

Table 7. Incidence (%) of broken wings, broken legs and bruised back, breast, wings and legs following commercial catching and processing of 49-d-old broilers

Treatment[1]	---Broken---		------------- Bruised -------------			
	Wings	Legs[2]	Back	Breast	Wings	Legs
Industry	5.00[3]	1.70	0.50	1.00	22.80	4.00
Research Requirements (RR)	6.99	2.20	2.00	0.50	13.95	2.20
RR+PHY	5.40	1.60	0.60	0.50	15.93	2.20
RR+PHY+25D	7.29	4.30	1.00	0.00	14.94	2.10
Industry+PHY	7.49	3.20	1.00	1.00	15.93	6.99
Negative Control	5.00	1.00	1.00	1.00	9.98	2.40
SEM[4]	2.00	1.40	0.80	0.60	3.30	1.40
P Value	0.88	0.58	0.72	0.74	0.20	0.09

[1] See Table 3 for treatment details.[2] Leg defined as both thigh and drumstick. [3] Values are means of 9 replicate pens except for negative control treatment that had 10 replicate pens; 22 birds per pen were randomly selected for processing. Percentage data were transformed using the arcsine function to satisfy the assumptions of normality and homogeneity of variances for statistical analysis; the values shown are inverse conversion of arcsine means to percent. [4] Weighted average of the standard error of the means..

Table 8. Live wt, hot carcass wt and wt and yield of carcass components of processed 49–d-old broilers

Treatment[1]	Hot carcass		Carcass parts weight, g			Carcass component yield, %[2]		
	Weight[3], g	Yield[4], %	Breast[5]	Wings[6]	Legs[7]	Breast	Wings	Legs
Industry	2,091[a8]	71.71	525	269	665[a]	25.64	12.56	32.71
Research Req (RR)	2,072[ab]	71.68	523	269	666[a]	25.78	12.59	31.36
RR+PHY	2,098[a]	71.87	536	272	671[a]	26.09	12.65	31.70
RR+PHY+25D	2,045[bc]	71.77	511	266	657[a]	25.58	12.68	31.36
Industry+PHY	2,070[ab]	72.20	530	266	666[a]	26.17	12.55	31.33
Negative Control	2,018[c]	71.67	509	267	636[b]	25.85	12.82	30.68
SEM[9]	19.7	0.246	1.2	2.5	7.8	0.380	0.157	0.68
P value	<0.01	0.408	0.097	0.492	<0.01	0.763	0.515	0.434

[1] See Table 3 for treatment details. [2] Percent yield, hot carcass weight basis. [3] Hot carcass weight taken just prior to chill tank. [4] Hot carcass as a percent of full farm BW. [5] Wt of both pectoralis major and minor (butterfly cut) with skin, removed by processing plant de-boning personnel.
[6] Wing includes humerus, radio-ulna and metacarpals. [7] Legs defined as thigh and drumstick.
[8] Values are means of 9 replicate pens except for negative control treatment that had 10 replicate pens; 22 birds per pen were randomly selected for processing. [9] Weighted average of the standard error of the means..
[a-c] Values in a column with different superscript letters differ ($P \leq 0.05$).

Discussion

The results that had been obtained under controlled battery research conditions were confirmed and were applicable under floor pen conditions. Application of PHY with its sparing effect to diets formulated at nPP requirements resulted in substantial fed phosphorus reductions. There were no indications from this study that reduction in diet phosphorus to account for the sparing effect of PHY and 25D on dietary phosphorus, do not lead to losses at the processing plant. Progressive implementation of these diet concentrations of phosphorus and PHY should be pursued with industry. Current costs of 25D curtail its use commercially if the purpose of its inclusion is only to replace a portion of the diet phosphorus.

Project 3: A balance trial to document actual nitrogen excretions in broilers fed industry diets.

To fulfill the nitrogen excretion baseline determination part of the grant, a balance study was done to document actual nitrogen excretions using industry type diets and industry nutrient concentrations.

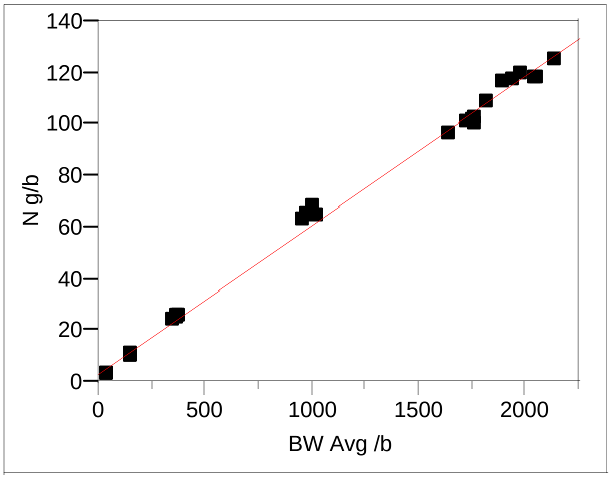
Methods

All procedures used in the broiler chicken experiments were approved by the institutional Animal Care and Use Committee. A broiler study was done with Ross 308 male broilers raised in battery pens. Fifteen pens of birds (eight birds per pen) were fed an industry diet that reflected industry nutrient levels (Agri Stats, 2002). Levels formulated were 21, 19, 17.5 and 16.5% protein in the starter, grower, finisher and withdrawal phases, respectively. The levels of non-phytate P (nPP) formulated were 0.45, 0.35, 0.35 and 0.30% in the starter, grower, finisher and withdrawal phases, respectively. Feed consumption was measured (feed offered minus wastage) by phase and excreta collected in its totality every day. At the end of each phase, all the birds in three pens were fasted for 16 hours to minimize intestinal digesta content, killed and frozen. The carcasses (including feathers and viscera) were ground, freeze-dried and then ground again using a freeze grinder. Excreta were weighed as collected by phase, freeze-dried and ground. Feed, carcasses and excreta were analyzed for dry matter, nitrogen and phosphorus as described for section B. For the purpose of write up only the excretion numbers will be used. Pen values for excreta dry matter, nitrogen and phosphorus were regressed against body weight. Best fit for all measurements was linear regression.

Results

Average weight of the birds at the end of the experiment (43 days of age) was 2072.99 grams. Average nitrogen content in the carcass was 3.1% and average nitrogen consumed was 113.7 grams. These birds excreted an average of 893.57 g of dry matter, 39.92 g of nitrogen and 11.94 g of phosphorus. The prediction formula for nitrogen excretion based on body weight is given in Figure 1.

<u>Figure 1.</u> Nitrogen excretion regressed on Average body weight per bird (BW Avg/b)



Linear Fit

Nitrogen excretion (grams per bird) = 3.087345 + 0.0578082 BW (r square = 0.9959), where BW is body weight.

Discussion

In 2003, the National Academy of Sciences published nitrogen excretion estimates based on model predictions (NRC, 2003). As can be seen from Table 9, the NRC (2003) estimates grossly overestimate excretion. The results of the mass balance trial done for this grant were instrumental in developing the ASAE (2005) broiler excretion model that more closely reflects actual excretions and allows for variability and margin of safety.

The model proposed by NRC (2003) for nitrogen is based on NRC (1994) growth and efficiency values and on typical carcass nitrogen content values (NRC, 2003) of 2.6, 2.5, and 2.3% nitrogen in starter, grower, and finisher birds, respectively. Based on this model the estimated nitrogen excretion for a 7 week-old broiler weighing 2250 grams is 96.5 g of nitrogen. It is important to note what information was used in the model. The information is based on a carcass balance or true balance; on consumptions and N content in feed according to NRC (1994) (overestimates as compared to today's commercial broiler) and on an average carcass nitrogen of 2.4% derived primarily from a reported carcass nitrogen determined on 56 day old female broilers (Santoso et al., 1995). This body nitrogen content is not applicable to males of the same weight.

If the assumptions of the NRC (2003) model are revised to reflect more current nitrogen consumption numbers and the nitrogen content of the carcass is corrected to reflect nitrogen content of a carcass with feathers and viscera (3.1% vs 2.4% nitrogen carcass content the model uses currently) then the prediction would be for 56.35 grams of nitrogen excreted per 2360 gram bird. This number would include any nitrogen lost to the environment through volatilizations. Other reports on nitrogen content in carcasses agree with the values obtained in the current broiler balance trial. Values of 3.9% in 16 day-old unsexed broiler chickens (Van Der Hel et al., 1992) and of 3.2% in male broiler chicks weighing 1083 grams (Brady et al., 1978) support the 3.1% nitrogen content found in this study. . In another study, Havenstein et al. (1994) reported a 2.9% nitrogen content in the featherless, eviscerated carcasses.

Table 9. Model estimates of nutrients in excreta versus mass balance trial results

		Excreted Nutrients, grams per bird		
	Body weight, g	Dry matter	Nitrogen	Phosphorus
ASAE 2005 Model	2360	1269.3	53.18	15.82
NRC (2003) Model	2250		96.5	
Mass Balance Trial	2073	893.57	39.92	11.94
Estimate to equal body weight				
ASAE 2005 Model	2360	1269.3	53.18	15.82
NRC (2003) Model	2360		103.58	
Mass Balance Trial	2360	1010.77	45.76	13.73

Conclusions

The three projects conducted for this grant have demonstrated that:

Reductions in broiler-litter phosphorus content have occurred since the start of the use of phytase in diets but further decreases are possible and needed to minimize increases in water-soluble phosphorus. The current apparent decrease in phosphorus excretion in broilers, based on this study, of 30% is associated with a decrease in fed phosphorus in the presence of phytase of 10%.

A decrease of 23% in fed phosphorus vs. industry average can be implemented without having negative effects on performance or on processing yields. A safety margin needs to be implemented under commercial conditions but decreases of at least 18% are warranted when phytase is used. Compared to the current maximum decrease of 10% in fed phosphorus, the 18% decrease that is feasible based on this study, would further reduce excreted phosphorus.

Previously calculated values for phosphorus and nitrogen excretion in broilers, as published by ASAE (American Society of Agricultural Engineers) and as used to establish baselines for regulations by EPA, grossly overestimate excretion of nitrogen and phosphorus by today's broilers and thus no longer apply. If values used to establish baselines for regulating poultry farms are based on excretion values that are grossly high then the number of birds in a farm that will establish that farm as AFO (animal feeding operations) and CAFO (confined animal feeding operations) for regulatory purposes will be much lower. This would place a heavier burden on the producer than is warranted. Fortunately new values have been published by ASAE in 2005 for all species and the broiler values in the ASAE (2005) publication are based on the work from this grant.

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