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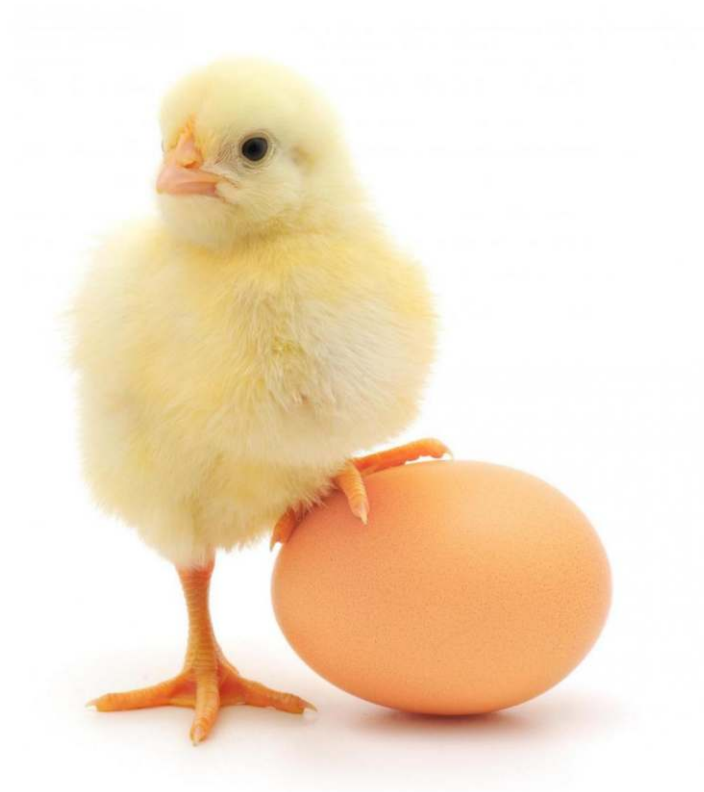

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Short Communication
Cholesterol, Yield, Tibia and Clavicle Ash of Broilers fed High Available Phosphorus Corn and/or Phytase with/without Alum Litter Treatment

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Abstract
Objective: Three pen trials were conducted to assess the effects of the source of dietary phosphorus (P) on cholesterol, tibia and clavicle ash and yield in broilers raised on litter with and without alum addition. These are additional data to supplement a previous report on the main effect of alum addition to litter on the form of P in litter. **Materials and Methods:** A 2×2 factorial structure was used for the diets (subunit treatments) that included high available phosphorus/low phytate corn (HAPC) and phytase (PHYT). **Results:** Overall, there did not appear to be differences in breast cholesterol, thigh cholesterol, or yield when comparing alum vs. no alum litter treatment. In addition, breast tissue cholesterol was not affected by diet. For thigh cholesterol, HAPC was least when a difference was detected. The only differences in meat yield were in Trial 1, where the greatest were PHYT and yellow dent corn (YDC) at 71.55 and 71.26%, respectively. HAPC had the lowest yield at 70.43%. The meat yield for the combination diet of HAPC and PHYT (H and P) did not differ from any of the other diets (71.02%). Tibia ash only exhibited some differences in trial 2. For the alum treated pens at 6 weeks, HAPC had the greatest tibia ash with lower ash apparent for the other diets. Clavicle ash was only assessed in trial 2 and did not appear different among the treatments. In the previous study, alum addition reduced water soluble P for all diets but only H and P did so where alum was not added. In this report, marginally, HAPC had least cholesterol and greatest tibia ash but the lowest yield. **Conclusion:** It is concluded that the combination of diet, H and P, shows promise for sustainable broiler production by maintaining comparable cholesterol and meat yield as well as reducing excretion of water soluble P.

Key words: Alum, broiler, cholesterol, high available phosphorus corn, phytase, tibia ash

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Population growth, greater income and urbanization are driving the demand for animal derived food and poultry is the fastest growing agricultural sub-sector, especially in developing coutries[1]. Challenges of production include food security as well as social, health and environmental impacts[1]. Research around poultry production strives to meet these challenges to formulate better strategies for sustainability, requiring a combination of approaches.

Specifically, for discussion is use of alum litter treatment and broiler diets containing either or both high available phosphorus/low phytate corn (HAPC) and phytase (PHYT). Alum has been added to poultry litter for a number of years to reduce NH_3 volatilization inside broiler houses and to reduce phosphorus runoff where litter is applied as fertilizer. Moore Junior *et al*.[2] reported these benefits in addition to heavier birds, lower mortality, improved feed efficiency and lower electricity and propane costs in alum-treated houses. High available phosphorus corn was developed by USDA scientists in the 1990s and produced by a major seed company (DuPont Pioneer, formerly Pioneer Hi-Bred, Johnston, IA) in the U.S. It has similar total P content to that of YDC but greater available P so that birds excrete less P. Phytase enzyme added to the diet of poultry increases the digestibility of phytate bound P, the major source of P in YDC and reduces the need to supplement diets with inorganic P, which reduces P excretion[3]. Huff *et al*.[3] and Miles *et al*.[4] discuss HAPC and PHYT in more detail.

Miles *et al*.[4] previously incorporated alum litter treatment and included HAPC and PHYT in broiler diets to determine the concentrations of total P and water soluble P in broiler litter during three growouts. The aim of this publication was to report the remainder of the data captured during those studies: breast and thigh tissue cholesterol, meat yield, tibia and clavicle ash. These parameters are published herein to aid in formulation of comprehensive sustainability strategies for broiler production.

MATERIALS AND METHODS

Experimental design: A split-plot experimental design was used where the main unit treatment (randomized complete block with four replications) was alum addition to litter and the sub-unit treatments were inclusion of HAPC and PHYT in a 2×2 factorial structure. The detailed materials and methods were previously published in Miles *et al*.[4], including dietary formulations and litter analysis. As in the previous report[4] of litter nutrient values (total P and water-soluble P), parameters were analyzed on a trial by trial basis, using the general linear model of SAS[5]. There were no interactions of HAPC and PHYT. The SAS mixed model assessed overall differences between alum vs no alum pens. Breast tissue cholesterol, thigh tissue cholesterol, meat yield, tibia ash (at 3 and 6 weeks of age) and clavicle ash were reported. Significant differences were based on $p \leq 0.05$.

Broiler rearing: For each trial, fifty-five male, Ross×Ross broilers were raised in each of 32, 1.52×2.44 m pens. Thus, each trial raised 1,760 male broilers to 42 days where available P was 0.45% in starter diets and 0.35% in grower diets. Each pen had nipple waterers and a hanging feeder. For the first 3d, lighting was 26 lx and was then reduced to 3.6 lx for the remainder of each flock. Chicks were brooded at 32°C for one week with weekly reductions of 2.8°C until 35 days. The rest of the growout maintained an aerial temperature of 21°C. Kiln-dried pine shavings were weighed into each pen before the first trial and spread to a depth of 10 cm. No alum was added to the pens before trial 1, which eliminated the main unit treatment and doubled the dietary replications for this trial. Alum was spread by hand and raked into the litter before trials 2 and 3 at a rate of 0.091 kg alum/bird.

Tissue and bone analyses: Table 1 reports breast and thigh tissue cholesterol and broiler meat yield for trials 1 and 2. A humidity and temperature spike at 41 days during the 3rd trial caused significant mortality (~47%); the decision was made to evaluate bone ash for this trial but not cholesterol and yield. At 42 days, five birds/rep were processed for yield, tissue sampled for cholesterol and tibia bone ash. Also, five birds/rep had been randomly selected at 21 days to determine tibia ash.

Tissue samples were included the breast muscle (p. major and p. minor) and the complete thigh muscle from either the left or right side of the bird. All samples were maintained on a per bird basis and were frozen prior to shipment for analysis. Two sub-samples of each type of tissue were analyzed for % cholesterol by Optimum Quality Grains, LLC (Des Moines, IA).

Table 2 reports tibia bone ash at 3 and 6 weeks of age, as well as clavicle bone ash at 6 weeks for the trial 2 flock. A colleague had suggested looking at clavicle ash, because clavicle strength could impact the deboning process and the trend for consumers wanting a greater percentage of deboned meat was growing. A common boiling/extracting method was used to determine bone ash[6]. Briefly, thawed bones were placed in boiling water for 2 min, meat was removed with cheese cloth, cartilage was removed and the bone was retagged if necessary. Bones were dried at 90°C for 24 h and placed in a desiccator until fat extraction with ethanol (24 h) followed by extraction with anhydrous ether (24 h). Bones were then air dried for 24 h, weighed, ashed for 24 h at 600°C and reweighed.

Table 1: Effect of alum or no alum treatments combined with yellow dent corn (YDC), high available phosphorus corn (HAPC), phytase (PHYT) and HAPC and PHYT combination (H and P) diets on broiler breast and thigh cholesterol and broiler meat yield

	Trial 1		Trial 2	
Dietary Treatments	No ALUM	ALUM	No ALUM	ALUM
Breast tissue cholesterol (%)				
YDC	0.373	-	0.437	0.436
HAPC	0.368	-	0.426	0.435
PHYT	0.371	-	0.431	0.437
H and P	0.376	-	0.429	0.436
LSD	0.011	-	0.022	0.025
Mean	0.372	-	0.431	0.436
Thigh tissue cholesterol (%)				
YDC	0.350[a]	-	0.369	0.385[a]
HAPC	0.335[b]	-	0.382	0.366[b]
PHYT	0.353[a]	-	0.372	0.378[ab]
H and P	0.351[a]	-	0.372	0.393[a]
LSD	0.012	-	0.021	0.019
Mean	0.347	-	0.374	0.381
Yield				
YDC	71.260[a]	-	72.710	73.270
HAPC	70.430[b]	-	72.890	72.820
PHYT	71.550[a]	-	72.740	72.730
H and P	71.020[ab]	-	72.890	72.880
LSD	0.750	-	0.750	0.720
Mean	71.030	-	72.810	72.930

[a, b]Within trial and within no alum or alum treatments, means with different letter differ significantly ($p\leq 0.05$)

Table 2: Effect of alum or no alum treatments combined with yellow dent corn (YDC), high available phosphorus corn (HAPC), phytase (PHYT) and HAPC and PHYT combination (H and P) diets on broiler tibia ash at 3 and 6 weeks of age and broiler clavicle ash at the end of flock 2

	Trial 1		Trial 2		Trial 3	
Dietary Treatments	No ALUM	ALUM	No ALUM	ALUM	No ALUM	ALUM
tibia ash (%) at 3 weeks of age						
YDC	51.33	-	50.87[b]	47.64[b]	48.68[b]	48.07
HAPC	50.66	-	52.55[a]	48.87[ab]	49.66[ab]	49.91
PHYT	50.44	-	51.00[b]	50.51[a]	50.78[a]	49.68
H and P	50.95	-	51.66[ab]	50.74[a]	49.84[ab]	49.62
LSD	1.02	-	1.02	1.02	1.02	1.02
Mean	50.84	-	51.52	49.43	49.73	49.31
tibia ash (%) at 6 weeks of age						
YDC	52.27	-	53.31	52.66[b]	52.60	53.35
HAPC	52.45	-	53.55	53.61[a]	53.11	52.03
PHYT	52.27	-	53.14	52.56[b]	52.30	52.86
H and P	53.15	-	53.31	52.71[b]	52.89	52.65
LSD	1.14	-	0.91	0.80	1.10	2.00
Mean	52.54	-	53.33	52.91	52.72	52.73
clavicle ash (%) at 6 weeks of age						
YDC			50.71	50.63		
HAPC			56.78	52.52		
PHYT			49.35	49.49		
H and P			53.45	52.40		
LSD			13.11	12.36		
Mean			52.50	51.20		

[a, b]Within trial and within no alum or alum treatments, means with different letter differ significantly ($p\leq 0.05$)

RESULTS AND DISCUSSION

Breast tissue cholesterol was approximately 0.37% for all diets in trial 1, none of which appeared to differ at $p\leq 0.05$ (Table 1). Further, there were no apparent differences in trial 2 among the no alum or alum treatments. Mean breast cholesterol was approximately 0.43% in trial 2.

For thigh tissue cholesterol in trial 1 (Table 1), HAPC had the least cholesterol of 0.335%. The other diets, YDC, PHYT and H and P, appeared similar to each other having ~0.35%

cholesterol (LSD = 0.012). In trial 2, the no alum pens did not differ among the diets and thigh cholesterol was approximately 0.37%. For the pens where alum was added, HAPC again had the least cholesterol (0.366%); YDC and H and P had the most at 0.385 and 0.393%, respectively. The PHYT diet with 0.378% thigh cholesterol did not appear different from the other diets.

Meat yield (Table 1) was greatest for PHYT and YDC (71.55 and 71.26%) in trial 1. The yield of the H and P diet (71.02%) did not appear different from any of the diets. However, the HAPC was lowest having 70.43% meat yield. No dietary differences or effect of alum treatment was evident in trial 2 yield. Mean yield in trial 2 was greater than trial 1 at approximately 72.9%. In fact, among the non-alum pens HAPC and H and P had numerically greater yield, whereas, YDC was numerically superior among the alum treated pens. These results are similar to Huff *et al.*[3] They investigated Cobb×Cobb broilers in two trials and found decreased serum cholesterol for the HAPC diet but no significant or consistent treatment effects on yield.

Tibia ash at 3 weeks (Table 2) indicated no significant differences in trial 1 or in the alum pens during trial 3. In trial 2 and the non-alum pens of trial 3, diets that included HAPC and/or PHYT generally had greater percentage bone ash. By 6 weeks, the only significant differences detected were in the alum treated pens in trial 2. At that time, HAPC had the greatest bone ash (56.61%). The other diets had lower bone ash (52.56-52.71%) and did not show differences from each other. No differences for tibia bone ash were observed at 6 weeks in trial 1, non-alum pens in trial 2, or in trial 3. Again, similarly, Huff *et al.*[3] also reported no significant or consistent treatment effects on bone diameter, strength, or ash.

Determining clavicle ash was a novel idea but the data was extremely inconsistent. The overall average was 52.23±18.02%. The qualitative comments showed that 46 of 158 clavicles had some part missing; these were excluded from the statistical analysis. The new overall clavicle average was 51.86±13.82%, still very inconsistent. It is not surprising that no differences were observed among the diets or alum treatments. A recent study on the effect of bone choice on quantification of mineralization in broilers[7] showed that bird age is a significant factor, suggesting femur ash was more appropriate for 6 weeks and older and that foot ash was comparable to tibia ash for broilers 2-5 weeks of age. Determinations for clavicle ash were not found in the literature.

Relative to phytase inclusion in diets, the literature[8] demonstrates improved performance of broilers, especially at higher rates (>500 FTU kg^{-1}) and that inclusion is common. Fewer recent manuscripts on HAPC were found in the literature but one of the more recent[9] discussed possible adoption by farmers, noting that an optimal variety would have low technology fees, low yield drag and a high harvest premium.

CONCLUSION

This study, combined with others from the same flocks of broilers, demonstrates that combining HAPC and PHYT in diets is one way to reduce farmgate imports of P and thus improve the overall sustainability of poultry production. The lack of significant differences across flocks suggests that the performance of broilers was not consistently negatively impacted by dietary modification or use of alum litter treatments. Use of clavicle ash was a novel method to determine the impact of these treatments to possible impacts on boneless meat products; however, due to issues with the fragility of the clavicle, other bones are likely to provide more complete information.

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