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Research Article Use of Phytase to Enhance Nutritional Value of Broiler Breeder Diets

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Abstract

Objective: This study was designed to determine the effects of dietary phytase on broiler breeder grower and layer diets. **Materials and Methods:** Chicks received a common starter diet up to 42 days of age. Treatments were 0 (control), 300, 600 and 1200 FTU kg⁻¹ phytase feed applied from 43 days to 4 replicate pens per treatment. Dietary calcium (Ca) was 0.7 and 2.7% while available phosphorus (AvP) was 0.35 and 0.12%, in the control grower and layer diets, respectively. Egg production, hatchability, fertility, egg quality, bone breaking strength, body weight (BW) and mortality to 64 weeks of age were measured. Data were analyzed using one-way ANOVA in 10 weeks quartile periods from 25-64 weeks of age as well as on an overall basis. **Results:** During the first quartile, phytase increased (p<0.05) egg production with 300 FTU when compared to the control with the 600 and 1200 FTU diets intermediate. All phytase treatments increased (p<0.05) egg production compared to the control on overall basis and during the final quartile (56-64 weeks), hatchability of fertile eggs was increased (p<0.05) by 600 and 1200 FTU phytase relative to the control with 300 FTU intermediate during the fourth quartile. Fertility, egg weight, shell thickness and bone breaking strength were not affected. **Conclusion:** As evidenced by increased egg production without effect on fertility, egg shell quality or bone strength, it was concluded that 300 FTU of phytase was sufficient to liberate adequate Ca and AvP as well as additional metabolizable energy (ME) but not crude protein (CP) in broiler breeder grower and layer diets.

Key words: Broiler breeders, phytase, egg production, hatchability, fertility

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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.

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INTRODUCTION

The concept of "minimum cumulative nutrition" during rearing of broiler breeders has been established¹⁻⁵. There is a minimum amount of crude protein (CP) required during rearing to support persistency of female fertility and a minimum amount of metabolizable energy (ME) required to support persistency of egg production. The minimum cumulative intakes to 140 days of age have been estimated to be 1,200 g CP and 23,000 kcal ME³⁻⁵. Poor feathering and poor hatchability have also been associated with a high inclusion of byproducts, such as wheat bran that have high levels of poorly digested phytate⁶. Over the past 15 years, phytase has been introduced to the poultry feed industry⁷ as an exogenous feed enzyme to liberate phosphorus from phytate. More recently, phytase has also been employed to liberate additional nutrient value in the forms of CP and ME from feedstuffs. Broiler breeders are, by definition, always nutrient deficient. The best evidence of an extra-phosphoric effect of phytase in growing broiler breeders should be expected to be an improved persistency of lay and/or fertility to the end of the broiler breeder production period. A fertility response should be interpreted due to an increment in accumulated CP during rearing¹⁻³. An egg production response should be interpreted due to an increment in accumulated ME during rearing^{1,4}. Further, the amount of daily ME intake at any time during the laying period would be expected to affect egg mass either by altering egg number (% egg production) or egg weight. Therefore, a marginal nutrient intake during the 7-25 weeks grower feed period and a greater than normal feed restriction during the laying period was programmed as a base for demonstration of any extra-phosphoric effects of phytase.

MATERIALS AND METHODS

Rearing phase management: Care of the birds in this study conformed to the Guide for Care and Use of Agricultural Animals in Research and Teaching⁸. A total of 1280 female Ross 708SF and male Ross 344 (Aviagen, Huntsville, AL) broiler breeder chicks were placed in an enclosed fan-ventilated house with 16 pens each for females and males. All birds were permanently identified with a neck tag. The ambient temperature was approximately 32°C during the first 7 days of brooding and the litter temperature was 35°C for the first 2 days. In order to maintain feeder space proportional to bird

size, female pens had 4 tube feeders to 14 days of age, followed by 3 tube feeders to 10 weeks of age, 4 tube feeders from 11-15 weeks and 5 tube feeders from 16 weeks until birds were moved to the laying house. Males had one tube feeder per pen at all times. Two bell drinkers per female pen were used, while males had one bell drinker per pen. There were 6 additional feeder lids per female pen and 3 feeder lids per male pen used during the first week. There were 2 additional font drinkers per female pen and one font drinker per male pen used during the first week. The pen floors were covered with fresh wood shavings. At 43 days of age, four grower diets initiated the dietary treatments. The lighting program consisted of 23 h of light per day to 7 days followed by 8 h to 21 weeks of age when the flock was moved to the laying house and photostimulated.

Laying phase management: At 21 weeks of age, 64 females and 8 males that had received the same dietary treatment from 43 days of age were moved to and co-mingled, in a 16-pen breeder house. Each of the 16 pens covered 15.9 m² with two-third slats and one-third pine shavings litter. There were four tube female feeders with restriction grills to exclude males (45×58 mm holes) and one male feeder in each breeding pen. Each pen had two bell drinkers and a conventional nest box comprised of two double and four single nests. Eggs were collected and recorded twice daily before being stored in an egg cooler prior to incubation. The lighting program was 14 h at photostimulation, 15 h 10 days later, 15.5 h at 5% production and finally 16 h from 50% production to end of study.

Pullet and breeder diets: Day-old chicks (females and males) were fed a common crumbled starter feed to 6 weeks of age (Table 1) followed by grower and layer diets from 7-25 and 26-67 weeks of age, respectively. A total of 4 dietary treatments for grower and layer breeder diets (Table 2 and 3) were employed. A *Buttiauxella* spp. derived phytase enzyme (DuPont Animal Nutrition, Marlborough, UK) was added at appropriate quantities at the expense of filler to achieve the dietary treatments, which were 0 (basal control), 300, 600 and 1200 FTU kg⁻¹. Feeding programs for males and females during rearing are outlined in Table 4. The female feeding to peak from 5% lay program is outlined in Table 5 and male and feeding programs during lay are outlined in Table 6. Following 21 days at the maximum feed intake of 155 g/hen/day, there was a weekly reduction in feed allocation (Table 6).

Table 1: Starter diet fed to 6 weeks of age

Table 1: Starter diet red to 6 weeks or age	
Ingredients	(%)
Corn	60.66
Soybean meal	23.23
Wheat bran	7.00
Corn gluten meal	1.40
Poultry fat	3.45
Limestone	0.98
Dicalcium phosphate	1.97
Salt	0.50
DL-methionine DL-methionine	0.14
L-threonine	0.02
Choline chloride	0.20
Vitamin premix ¹	0.10
Mineral premix ²	0.20
Coccidiostat ³	0.05
Selenium premix ⁴	0.10
	100.00
Calculated nutrient content ⁵	
Metabolizable energy (kcal g ⁻¹)	2.90
Crude protein	17.00
Calcium	0.90
Available phosphorus	0.45
Total lysine	0.85
Total threonine	0.56
Total methionine+cysteine	0.65

 1 Vitamin premix supplied the following per kg of diet: 7,000 IU vitamin A, 2,000 IU vitamin D3, 35 IU vitamin E, 0.02 mg vitamin B12, 0.13 mg biotin, 2 mg menadione (K_{3}), 2 mg thiamine, 7 mg riboflavin, 11 mg d-pantothenic acid, 4 mg vitamin B6, 55 mg niacin and 1 mg folic acid. 2 Mineral premix supplied the following per kg of diet: 0.30 mg manganese, 120 mg zinc, 120 mg iron, 80 mg copper, 10 mg iodine and 2.5 mg cobalt. 3 Coccidiostat supplied amprolium at 125 mg kg $^{-1}$ feed. 4 Selenium premix provided 0.2 mg Se (as Na $_{2}$ SeO $_{3}$). 3 Confirmed by proximate analysis

Table 2: Grower diets fed from 7-25 weeks of age

	Dietary treatments (FTU kg ⁻¹) (%)				
Ingredients	0	300	600	1200	
Corn	64.63	64.63	64.63	64.63	
Soybean meal	13.35	13.35	13.35	13.35	
Wheat bran	16.60	16.60	16.60	16.60	
Corn gluten meal	1.00	1.00	1.00	1.00	
Poultry fat	1.00	1.00	1.00	1.00	
Limestone	0.90	0.90	0.90	0.90	
Dicalcium phosphate	1.21	1.21	1.21	1.21	
Salt	0.50	0.50	0.50	0.50	
DL-methionine	0.09	0.09	0.09	0.09	
L-lysine	0.07	0.07	0.07	0.07	
Choline chloride	0.20	0.20	0.20	0.20	
Vitamin premix ¹	0.10	0.10	0.10	0.10	
Mineral premix ²	0.10	0.10	0.10	0.10	
Coccidiostat ³	0.05	0.05	0.05	0.05	
Selenium premix⁴	0.10	0.10	0.10	0.10	
Phytase ⁵	0.00	0.0165	0.033	0.066	
Vermiculite ⁶	0.10	0.0835	0.067	0.034	
	100	100	100	100	
Calculated nutrient content ^{7,8}					

Metabolizable energy (kcal g ⁻¹)	2.80
Crude protein	14.13
Calcium	0.70
Available phosphorus	0.35
Total lysine	0.70
Total threonine	0.49

Total methionine+cysteine

Vitamin premix supplied the following per kg of diet: 7,000 IU vitamin A, 2,000 IU vitamin D3, 35 IU vitamin E, 0.02 mg vitamin B12, 0.13 mg biotin, 2 mg menadione (K_3), 2 mg thiamine, 7 mg riboflavin, 11 mg d-pantothenic acid, 4 mg vitamin B6, 55 mg niacin and 1 mg folic acid. ²Mineral premix supplied the following per kg of diet: 0.30 mg manganese, 120 mg zinc, 120 mg iron, 80 mg copper, 10 mg iodine and 2.5 mg cobalt. ³Coccidiostat supplied amprolium at 125 mg kg⁻¹ feed. ⁴Selenium premix provided 0.2 mg Se (as Na₂SeO₃). ⁵Phytase enzyme added at 300, 600 and 1200 FTU kg⁻¹. ⁶Vermiculite was used as an inert filler. ⁷Formulations were based upon the control without added enzyme. Each diet was amended with phytase as a replacement for inert filler in this study. Nutrient values shown above only reflect basal values and not any matrix values added by phytase. ⁸Confirmed by proximate analysis

0.57

Table 3: Layer diets fed from 26-64 weeks of age

Total threonine

Total methionine+cysteine

	Dietary treatments	(FTU kg ⁻¹) (%)		
Ingredients	0	300	600	1200
Corn	65.55	65.55	65.55	65.55
Soybean meal	17.08	17.08	17.08	17.08
Wheat bran	6.04	6.04	6.04	6.04
Corn gluten meal	1.00	1.00	1.00	1.00
Poultry fat	1.00	1.00	1.00	1.00
Limestone	6.91	6.91	6.91	6.91
Dicalcium phosphate	0.00	0.00	0.00	0.00
Salt	0.50	0.50	0.50	0.50
DL-methionine	0.11	0.11	0.11	0.11
L-lysine	0.07	0.07	0.07	0.07
L-threonine	0.03	0.03	0.03	0.03
Choline chloride	0.20	0.20	0.20	0.20
Vitamin premix ¹	0.10	0.10	0.10	0.10
Mineral premix ²	0.10	0.10	0.10	0.10
Coccidiostat ³	0.05	0.05	0.05	0.05
Selenium premix ⁴	0.10	0.10	0.10	0.10
Phytase ⁵	0.00	0.0165	0.033	0.066
Vermiculite ⁶	0.15	0.1335	0.117	0.084
	100	100	100	100
Calculated nutrient content ^{7,8}				
Metabolizable energy (kcal g ⁻¹)	2.80			
Crude protein	14.80			
Calcium	2.70			
Available phosphorus	0.12			
Total lysine	0.75			

Vitamin premix supplied the following per kg of diet: 7,000 IU vitamin A, 2,000 IU vitamin D3, 35 IU vitamin E, 0.02 mg vitamin B12, 0.13 mg biotin, 2 mg menadione (K₃), 2 mg thiamine, 7 mg riboflavin, 11 mg d-pantothenic acid, 4 mg vitamin B6, 55 mg niacin and 1 mg folic acid. ²Mineral premix supplied the following per kg of diet: 0.30 mg manganese, 120 mg zinc, 120 mg iron, 80 mg copper, 10 mg iodine and 2.5 mg cobalt. ³Coccidiostat supplied amprolium at 125 mg kg⁻¹ feed. ⁴Selenium premix provided 0.2 mg Se (as Na₂SeO₃). ⁵Phytase enzyme added at 300, 600 and 1200 FTU kg⁻¹. ⁶Vermiculite was used as an inert filler. ⁷Formulations were based upon the control without added enzyme. Phytase was added as a replacement for inert filler. Nutrient values shown above only reflect basal values and not any matrix values added by phytase. ⁸Confirmed by proximate analysis

0.53

0.62

All of the treatment diets were formulated to be isonitrogenous and isocaloric with similar amino acid levels. Feed was mixed in a horizontal double ribbon mixer (TRDB126-0604, Hayes and Stolz Ind. Mfg. Co., Fort Worth, TX). Corn was ground using a two-pair roller mill (Model C128829, RMS, Tea, SD) and corn particle size was determined⁹. The common starter diet (Table 1) was made with 800 µm corn and contained 17.5% CP and 2.9 kcal g⁻¹ ME. Grower diets (Table 2) and layer diets (Table 3) were made with 1200 µm corn. Conditioned mash (at 85°C for 45 sec) was pelleted (ring die of 4.4 mm by 35 mm, pellet mill Model PM1112-2, California Pellet Mill Co., Crawfordsville, IN), cooled in a counter-flow cooler (Model VK09 × 09KL, Geelen Counterflow USA Inc., Orlando, FL) and then crumbled. This heat treatment was intended to reduce endogenous phytase normally found in these feed ingredients.

Data collection: All males and females were group weighed by pen at placement and individually weighed at 7, 14 and

20 weeks of age. At 20 weeks, 8 males and 64 females were selected to represent the existing BW distribution of each pen and transferred to the laying house. At 26, 32, 40, 48 and 57 weeks of age, 20 randomly selected females per pen were weighed. All males were weighed at each of these ages as well as at 64 weeks of age. Nest and floor eggs were collected and recorded twice daily. Nest eggs were collected and identified by pen, separated from floor eggs and stored in an egg cooler at 17°C and 70% RH. A minimum of 60 eggs per pen were set at least every other week to determine fertility and hatchability. After hatching was completed, eggs that did not hatch were examined macroscopically to determine fertility and/or time of embryonic mortality. Egg samples (20 eggs per pen) were collected at 30, 40, 50 and 64 weeks of age for egg quality assessment. Egg weight, yolk weight, shell weight, albumen weight, shell thickness and percentage shell were determined. At 65 weeks of age, one pen from each treatment with similar egg production was selected. All females in each of selected pens were necropsied to determine absolute and

Table 4: Feeding program from initial placement to sexual maturity

	Feed allocation1 (g/b	oird/day)
Flock		
age (weeks)	Female	Male
1	32.4	38.9
2	32.4	50
3	34	55
4	37	60
5	40	70
6	43	80
7	45	65
8	47	67
9	49	69
10	51	71
11	53	73
12	55	75
13	58	78
14	61	81
15	64	84
16	68	87
17	72	91
18	77	96
19	82	102
20	87	108
21	92	114
22	96	117
23	100	119
24	104	121
25	108	123

¹Females and males were fed on a daily basis to 28 days of age followed by restricted feeding employing a 4/3 feeding to 13 weeks of age (fed Monday, Wednesday, Friday and Saturday). A 5/2 feeding program (Not fed on Wednesday and Sunday) was employed from 14 weeks of age to photostimulation and housing at 21 weeks of age. Thereafter, daily feeding was utilized

Table 5: Feeding to peak program from 5% rate of lay

Rate of lay (%)	Feed allocation ¹ (g/bird/day)
5	108.0
7	109.0
9	110.0
11	111.0
14	112.0
17	113.5
20	115.0
23	117.0
26	119.0
29	121.0
31	124.0
34	127.0
38	130.0
42	134.0
46	138.0
50	142.0
54	146.0
58	150.0
60	155.0

Females received daily feed amounts as shown above beginning at 5% rate of lay, which was at the first day of 25th weeks (169 days) of rearing. The peak feed amount of 155 g/hen/day was maintained for 21 days

relative weights of liver, ovary, oviduct, gizzard and proventriculus. Tibia bones were excised to determine bone

Table 6: Separate sex male feeding program from 26 weeks of age

	Feed allocation' (g/bird/day)				
Flock age					
(weeks)	Male ¹	Female ²			
26	123.0	127.0			
27	123.0	152.0			
28	125.0	155.0			
29	125.0	155.0			
30	127.1	150.0			
33	127.1	147.0			
36	129.8	144.0			
39	132.6	141.0			
42	135.3	138.0			
45	138.0	135.0			
48	140.7	133.5			
51	143.5	132.0			
54	146.2	130.5			
57	148.9	129.0			
60	151.6	127.5			

Males were fed amounts as shown above on a daily basis in a separate male feeder. Peak feed of 155 g/hen/day was maintained for 21 days (30 weeks of age). Female feed allocation was then decreased by 5 g followed by 1 g/hen/day on a weekly basis to 45 weeks of age and thereafter, decreased by 1 g/hen/day on a bi-weekly basis. This is summarized in an abbreviated format above for the sake of brevity

strength by using a 3-point bending test. Bones were sheared midshaft using a crosshead speed of $30\,\mathrm{mm}\,\mathrm{min^{-1}}$ to minimize splintering 10 .

Statistical analysis: A one-way ANOVA using completely randomized design with 4 replicate pens per dietary treatment was employed. Variables were analyzed on a quartile age period and on overall basis from 24-64 weeks of age. There was a total of four 10 weeks quartile age periods. Data were examined for normality of distributions and homogeneity of variance. Percentage data were subjected to arcsin transformation before analysis. The GLM of SAS¹¹ was used to analyze variables of egg production and BW and differences among means were partitioned by LSMEANS.

RESULTS AND DISCUSSION

Diet formulations were based upon the control (0 FTU kg⁻¹) to which the various levels of phytase were added¹². This was an "on top" application of phytase as has been frequently employed in numerous commercial situations globally for a number of years (personal observations of the authors). The 300 FTU kg⁻¹ dose was assumed to liberate approximately 0.1% AvP and 0.1% Ca.

Cumulative nutrient intakes calculated from Table 1-4 during rearing were 1,218 g CP and 23,277 kcal ME to 21 weeks of age. These nutrient intakes were achieved 7 days later than the previously determined minimum age¹⁻⁵. This insured that the flock had been reared on marginal nutrition.

Table 7: Effect of inclusion level of dietary phytase on broiler breeder body weight

Flock age	Phytase (FTU kg	$g^{-1})^{1,2}$ (g)				
(weeks) and sex	0	300	600	1200	SE	p-value
Female						· · · · · · · · · · · · · · · · · · ·
0	37	38	38	37	1	NS
7	760	768	758	771	3	NS
14	1523	1531	1519	1494	6	NS
20	2163ab	2207ª	2168ab	2129 ^b	10	0.04
26	3062	3097	3096	3048	16	NS
32	3566	3612	3562	3524	15	NS
40	3580	3586	3597	3511	19	NS
48	3851	3793	3748	3801	23	NS
57	4032	4002	3921	3941	19	NS
Male						
0	43.64	43	43.50	43	1	NS
7	1419	1419	1394	1402	7	NS
14	2388ª	2332 ^{bc}	2379 ^{ab}	2303°	12	0.02
20	3136	3165	3166	3073	16	NS
26	3868	3870	3871	3816	36	NS
32	3856	3805	3987	3768	67	NS
40	3943	4120	4037	3860	55	NS
48	4081	4328	4225	4176	61	NS
57	4424	4529	4546	4626	70	NS
65	4621	4765	4691	4720	59	NS

abc Means that possess different superscripts within an age quartile or cumulatively differ significantly (p≤0.05). ¹Phytase used was derived from Buttiauxella spp. and provided 2 FTU activity per g of product. ²Means and SE for four pens of broiler breeders with 80 females and 16 males to 20 weeks of age and 20 females and 8 males per pen after 21 weeks of age

Table 8: Effect of dietary phytase on broiler breeder hen-day egg production

Flock age (weeks)		g ⁻¹) ^{1,2} (% egg production)				
and sex	0	300	600	1200	SE	p-value
25-35	68.1 ^b	73.8ª	71.4 ^{ab}	70.9ab	1.19	0.04
36-45	64.1	67.3	68.3	67.7	1.31	NS
46-55	56.3	60.1	63.2	61.9	1.78	0.08
56-64	40.3 ^b	49.8a	50.9ª	47.4ª	2.33	0.03
25-64	58.8 ^b	64.0a	64.7ª	63.3ª	1.33	0.03

^{a,b}Means that possess different superscripts within an age quartile or cumulatively differ significantly (p≤0.05). ¹Phytase used was derived from *Buttiauxella* spp. and provided 2 FTU activity per g of product. ²Means and SE for four pens of broiler breeders with 64 females and 8 males per pen at 21 weeks of age when the flock was moved from rearing to laying quarters and photostimulated. The experimental diets began at 43 days of age

The effect of phytase on male and female BW is shown in Table 7. Females consuming the 300 FTU diet weighed more than those consuming the 1200 FTU diet at 20 weeks of age with the other diets intermediate. Males consuming the control diet weighed more than those consuming the 300 and 1200 FTU diets while the 600 FTU diet was intermediate at 14 weeks of age. No other effects on BW were observed. As BW is one cumulative measure of nutrient intake and balance, these data were interpreted to mean that effective daily ME intake varied very little across the treatments. However, this would not be unexpected as broiler breeders restricted to different ME intakes have been reported to exhibit quite similar BW⁵.

The effect of phytase on percentage hen-day egg production is shown in Table 8 and is summarized in Table 9. Hen-day egg production was improved (p<0.05) by the

300 FTU kg⁻¹ diet relative to the control during the initial 25-35 weeks of age quartile with the 600 and 1200 FTU kg⁻¹ dosages intermediate. This suggested that the 300 FTU diet elaborated sufficiently greater ME to affect egg number but not BW (Table 7) or egg weight (Table 10). Additional phytase produced no additional benefit. Thus, the dietary requirement for AvP, on a percentage basis, during lay was clearly met by the control diet. Kashani et al.¹³ reported the performance of laying hens fed a diet with 0.10-0.13% AvP in the presence of 100-300 units phytase to be comparable to hens fed an AvP level of 0.4-0.55%. During the final 56-64 weeks quartile and overall (25-64 weeks), all phytase dosages improved (p<0.05) rate of lay relative to the control. Francesch et al.14 reported improved rate of lay by hens fed a low non-phytate P (NPP) diet supplemented with phytase when compared to non-supplemented hens. All phytase dosages also produced a greater number of eggs per hen housed (p<0.10) (Table 9). Thus, the effect of phytase was evident at the end of lay where it would be expected, if there was an increased effective cumulative intake of ME during rearing of the broiler breeder female^{4,5}. The absence of a dose response was most likely due to some negative effect or lack of greater dietary requirement, beyond the 300 FTU range. Nusairat *et al.*¹² reported that a phytase dosage of 1200 FTU kg $^{-1}$ probably fully disrupted the phytate present in the broiler breeder digestive tract as evidenced by an increased fecal liquid portion. This full disruption would be expected to have elaborated maximum nutritional value from the feed if such were possible. Indeed, Nusairat *et al.*¹² reported increased fecal P but not Ca, at 600 and 1200 FTU as compared to 300 FTU. Thus, there must be an optimum dosage in the 300 FTU range.

The effects of phytase on all live production variables are summarized in Table 9. Heat stress caused some mortality at the end of the first quartile age period that was not treatment related (Table 9). There were no significant overall effects of phytase on fertility (Table 9), which suggested that the effective cumulative CP intake for the females had not been significantly altered nor had the males been affected. Walsh and Brake^{2,3} demonstrated the relationship between

cumulative CP during rearing of females and subsequent fertility. Hatchability of total eggs set (Table 9) was not significantly affected by phytase. Hatchability of fertile eggs was decreased in the control hens relative to the 600 and 1200 FTU kg⁻¹ groups with the 300 FTU kg⁻¹ group intermediate during the last quartile age period (86.1, 88.2, 89.7 and 89.2%, respectively). However, there was no cumulative effect (Table 9). There were also no significant cumulative effects of dietary phytase on percentage early dead, late dead and pipped embryos (Table 9).

The effects of dietary phytase on egg and egg characteristics at 30, 40, 50 and 64 weeks of age are shown in Table 10-13. No effects were observed at 30 or 64 weeks of age (Table 10 and 13). At 40 weeks of age (Table 11), percentage yolk was greater for the 300 and 1200 FTU kg⁻¹ groups as compared to the control with the 600 FTU kg⁻¹ group intermediate. Albumen weight was greater in the control group as compared to 300 and 600 FTU kg⁻¹ groups with the 1200 FTU kg⁻¹ group intermediate. Percentage albumen was greater for the control group as compared to the others. Shell weight was greatest in the control and 1200 FTU kg⁻¹ groups as compared to the 300 FTU kg⁻¹ group with the 600 FTU kg⁻¹ group intermediate while percentage

Table 9: Effect of dietary phytase on broiler breeder hen-day egg production, female and male mortality, fertility, hatchability of total and fertile eggs and early dead, late dead and pipped embryos from 25-64 weeks of age

	Phytase (FTU	hytase (FTU kg ⁻¹) ^{1,2} (%)				
Variables	0	300	600	1200	SE	p-value
Hen-day egg production	58.8 ^b	64.0ª	64.7ª	63.3ª	1.3	0.03
Eggs per hen housed	142.7	160.7	155.2	152.8	5.0	NS
Female mortality	16.2	12.1	18.4	17.6	4.3	NS
Male mortality	15.6	21.9	25.0	22.3	6.3	NS
Fertility	95.1	96.4	95.7	94.9	1.1	NS
Hatchability of total eggs set	86.7	89.3	88.5	86.7	1.7	NS
Hatchability of fertile eggs	91.1	92.6	92.5	91.3	1.0	NS
Early dead embryos	5.0	4.0	3.9	4.4	0.5	NS
Late dead embryos	2.9	2.8	2.8	3.0	0.4	NS
Pipped embryos	0.9	1.0	0.8	1.4	0.3	NS

^{a,b}Means that possess different superscripts within an age quartile or cumulatively differ significantly (p≤0.05). ¹Phytase used was derived from *Buttiauxella* spp. and provided 2 FTU activity per g of product. ²Means and SE for 4 pens each of broiler breeders. Each pen had 64 females and 8 males at 21 weeks of age when the flock was moved from rearing to laying quarters and photostimulated. The experimental diets began at 43 days of age

Table 10: Effect of dietary phytase on broiler breeder egg, yolk, albumen and shell weight, shell thickness and percentage yolk, albumen and shell at 30 weeks of age

Variables	Phytase (FTU	3				
	0	300	600	1200	SE	p-value³
Egg weight (g)	55.85	54.67	54.85	55.52	0.36	NS
Yolk weight (g)	15.94	15.63	15.88	15.97	0.13	NS
Albumen weight (g)	34.80	33.96	33.91	34.48	0.28	NS
Shell weight (g)	5.11	5.08	5.05	5.06	0.05	NS
Shell thickness (mm)	0.37	0.37	0.37	0.36	0.01	NS
Yolk (%)	28.56	28.61	28.97	28.79	0.19	NS
Albumen (%)	62.29	62.09	61.79	62.09	0.20	NS
Shell (%)	9.16	9.29	9.22	9.12	0.07	NS

Phytase used was derived from *Buttiauxella* spp. and provided 2 FTU activity per g of product. ²Means and SE for 20 eggs from each of four pens of broiler breeders with 64 females and 8 males per pen at 21 weeks of age when the flock was moved from rearing to laying quarters and photostimulated. The experimental diets began at 43 days of age. ³No significant differences

Table 11: Effect of dietary phytase on broiler breeder egg, yolk, albumen and shell weight, shell thickness and percentage yolk, albumen and shell at 40 weeks of age

Variables	Phytase (FTU					
	0	300	600	1200	SE	p-value
Egg weight (g)	60.68	59.41	59.53	60.37	0.44	NS
Yolk weight (g)	18.38	18.60	18.33	18.65	0.16	NS
Albumen weight (g)	36.28ª	34.95 ^b	35.19 ^b	35.62ab	0.32	0.05
Shell weight (g)	6.03ª	5.86 ^b	6.00 ^{ab}	6.09 ^a	0.06	0.05
Shell thickness (mm)	0.38	0.37	0.37	0.37	0.01	NS
Yolk (%)	30.32 ^B	31.33 ^A	30.84 ^{AB}	30.90 ^A	0.21	0.01
Albumen (%)	59.74ª	58.81 ^b	59.07 ^b	59.00 ^b	0.23	0.05
Shell (%)	9.95	9.86	10.09	10.10	0.08	NS

^{a,b}Means that possess different superscripts within an age quartile or cumulatively differ significantly ($p \le 0.05$). ^{A,B}Means that possess different superscripts within an age quartile or cumulatively differ significantly ($p \le 0.01$). ¹Phytase used was derived from *Buttiauxella* spp. and provided 2 FTU activity per g of product. ²Means and SE for 20 eggs from each of four pens of broiler breeders with 64 females and 8 males per pen at 21 weeks of age when the flock was moved from rearing to laying quarters and photostimulated. The experimental diets began at 43 days of age

Table 12: Effect of dietary phytase on broiler breeder egg, yolk, albumen and shell weight, shell thickness and percentage yolk, albumen and shell at 50 weeks of age

Variables	Phytase (FTU	kg ⁻¹) ^{1,2}				
	0	300	600	1200	SE	p-value
Egg weight (g)	66.47	66.32	66.25	66.66	0.49	NS
Yolk weight (g)	21.17	21.28	21.16	21.42	0.20	NS
Albumen weight (g)	39.23	38.96	39.21	39.09	0.34	NS
Shell weight (g)	6.08 ^a	6.08 ^{ab}	5.89 ^b	6.14ª	0.07	0.05
Shell thickness (mm)	0.38	0.38	0.37	0.38	0.01	0.10
Yolk (%)	31.85	32.11	31.96	32.13	0.21	NS
Albumen (%)	58.99	58.73	59.14	58.62	0.21	NS
Shell (%)	9.16	9.16	8.90	9.29	0.09	0.10

^{ab}Means that possess different superscripts within an age quartile or cumulatively differ significantly (p \leq 0.05). ¹Phytase used was derived from *Buttiauxella* spp. and provided 2 FTU activity per g of product. ²Means and SE for 20 eggs from each of four pens of broiler breeders with 64 females and 8 males per pen at 21 weeks of age when the flock was moved from rearing to laying quarters and photostimulated. The experimental diets began at 43 days of age

Table 13: Effect of dietary phytase on broiler breeder egg, yolk, albumen and shell weight, shell thickness and percentage yolk, albumen and shell at 64 weeks of age

Variables	Phytase (FTU	kg ⁻¹) ^{1,2}				
	0	300	600	1200	SE	p-value ³
Egg weight (g)	71.45	69.91	69.33	69.55	0.69	NS
Yolk weight (g)	23.15	23.09	22.44	22.58	0.27	NS
Albumen weight (g)	42.16	40.82	40.89	41.03	0.49	NS
Shell weight (g)	6.18	6.01	6.00	5.94	0.08	NS
Shell thickness (mm)	0.39	0.40	0.39	0.39	0.01	0.10
Yolk (%)	32.41	33.08	33.42	32.46	0.28	NS
Albumen (%)	58.92	58.32	58.90	59.00	0.27	NS
Shell (%)	8.67	8.60	8.68	8.56	0.09	NS

¹Phytase used was derived from *Buttiauxella* spp. and provided 2 FTU activity per g of product. ²Means and SE for 20 eggs from each of four pens of broiler breeders with 64 females and 8 males per pen at 21 weeks of age when the flock was moved from rearing to laying quarters and photostimulated. The experimental diets began at 43 days of age. ³No significant differences

shell and shell thickness did not differ. At 50 weeks of age (Table 12), the control and 1200 FTU kg⁻¹ groups exhibited greater shell weight than the 600 FTU kg⁻¹ group with the 300 FTU kg⁻¹ group intermediate. There were no physical egg quality differences that could account for the decreased hatchability of fertile eggs observed in the control group at

the end of lay. At 65 weeks of age, absolute and relative organ weights (Table 14) and bone strength of females were not found to be affected by phytase (Table 15). These data suggested that there were no detrimental effects of the dietary formulations employed on the basic health and physiology of the females.

Table 14: Effect of dietary phytase on broiler breeder BW and absolute and relative weights of liver, ovary, oviduct, gizzard and proventriculus at 65 weeks of age

Weights	Phytase (FTU k					
	0	300	600	1200	SE	p-value ³
		(]			
Body	3921	3896	3812	3943	56	NS
Liver	55.20	55.60	57.40	57.90	1.70	NS
Ovary	50.00	52.00	54.00	53.50	3.60	NS
Oviduct	49.00	51.10	56.50	49.90	3.20	NS
Gizzard	44.80	41.70	41.60	44.60	1.20	NS
Proventriculus	7.20	6.80	7.20	7.00	0.20	NS
		(g/100	g BW)			
Liver	1.41	1.43	1.51	1.48	0.04	NS
Ovary	1.27	1.34	1.42	1.37	0.09	NS
Oviduct	1.26	1.33	1.51	1.28	0.09	NS
Gizzard	1.16	1.09	1.10	1.14	0.04	NS
Proventriculus	0.18	0.18	0.19	0.18	0.01	NS

¹Phytase used was derived from *Buttiauxella* spp. and provided 2 FTU activity per g of product. ²Means and SE for 50, 52, 51 and 53 tibia bones, respectively. Each pen had 64 females and 8 males at 21 weeks of age when the flock was moved from rearing to laying quarters and photostimulated. The experimental diets began at 43 days of age. ³No significant differences

Table 15: Effect of dietary phytase on broiler breeder tibia bone breaking strength at 65 weeks of age

	Phytase (FTU kg ⁻¹) ^{1,2} (N)						
Mayialala		200			CE		
Variable	0	300	600	1200)E	p-value	
Breaking strength	556.8	586.5	566.1	601.7	15.0	NS	

¹Phytase used was derived from *Buttiauxella* spp. and provided 2 FTU activity per g of product. ²Means and SE for 50, 52, 51 and 53 tibia bones, respectively. Each pen had 64 females and 8 males at 21 weeks of age when the flock was moved from rearing to laying quarters and photostimulated. The experimental diets began at 43 days of age. ³No significant differences

CONCLUSION AND FUTURE RECOMMENDATIONS

It was concluded that adding phytase "on top" of broiler breeder grower and layer diets at the rate of 300 FTU was sufficient to liberate adequate AvP as well as additional ME but not CP as evidenced by increased egg production without effect on fertility, egg weight or bone strength. A benefit of a dosage greater than 300 FTU was not observed in this study.

SIGNIFICANCE STATEMENT

The extra phosphoric effects of phytase as CP and ME can be interpreted as fertility and egg production responses, respectively, in broiler breeders. Phytase released additional ME during rearing as evidenced by improved persistency of egg production and during the initial laying period as evidenced by improved rate of lay but no additional release of CP was noted as evidenced by a lack of effect on fertility.

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