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Effect of phytase supplementation on fertility, hatchability and egg quality traits in swarnadhara breeders fed diet with different levels of non-phytate phosphorus

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Abstract
A biological trial was conducted to study the effect of phytase supplementation on fertility, hatchability and egg quality traits in Swarnadhara breeders reared from 29 to 48 weeks at the Department of Poultry Science, Veterinary College, Hebbal, Bengaluru. The trial had total 450 birds with 5 treatments and three replicates per treatment, each having 30 birds (90 birds per treatment). Control diet (T1) was formulated according to ICAR (2013) nutrient requirements and the groups T2 to T5 were formulated with changed non-phytate phosphorus levels so as to contain 0.12% in T2, 0.18% in T3, 0.12% NPP with 500 IU/kg phytase in T4 and 0.18% NPP with 500 IU/kg phytase in T5. Fertility, hatchability, albumen index, haugh unit score and yolk index are not significantly (P>0.05) affected by phytase supplementation. The addition of phytase to the diets having 0.12 % NPP significantly (P<0.05) increased the egg weight to the level on par with the control group in phase I, III and IV. NPP level of 0.12% with phytase is optimum for Swarnadhara breeders to maintain egg quality traits. There was no additional advantage, however, of increasing the NPP concentration above 0.12% or adding phytase above this level in Swarnadhara breeders.

Keywords: Phytase, NPP, egg traits, fertility, hatchability

Introduction
Phosphorus (P) has been found to be the second most abundant mineral in the body, not only for humans but for all domestic animals. Only 21% of the total P in corn and 35% in soybean meal (NRC, 1994) [9] were reported to be readily available for absorption, while the remainder was excreted in the faeces due to the lack of sufficient endogenous intestinal phytase. Berry *et al*., (2003) [1] reported that fertility and hatchability in broiler breeders are not sensitive to dietary NPP (0.1and 0.3%) and supplemental phytase (300 FTU/kg). Egg weight and egg specific gravity too was not significantly affected by phytase supplementation. Nusairat *et al.*, (2018) [10] reported that in broiler breeders fed laying diets containing 2.7 % Ca and 0.7% available P with phytase at different levels 0,300,600 and 1200 FTU/Kg has no significant effect on egg weight and egg quality traits such as shell weight, yolk and albumen weights. Faham *et al.*, (2016) [4] observed that the Arbor ACRES strain broiler breeder 54 weeks old fed diet supplemented with 200 g/ton phytabex plus a multi-enzyme mixture recorded higher fertility and hatchability percentages during experimental period (20 weeks) than other treatments (p<0.01). However, 1200 FTU/kg phytase increased the moisture content of the faeces and lead to sanitation problem in hatching eggs and broiler chick quality. The effect of phytase supplementation on fertility, hatchability and egg quality traits of Swarnadhara breeders fed different levels of NPP diets with supplementation of phytase is yet to be investigated. Therefore, the present study was undertaken to determine the effect of phytase supplementation with different levels of NPP on fertility, hatchability and egg quality traits of Swarnadhara breeders.

Materials and Methods
Birds, Diet and Management
Swarnadhara breeders, 29 weeks of age, were randomly distributed into 5 treatment groups of 90 each and housed in deep litter system. Considering 30 birds a replicate, 3 such replicates

were randomly allotted to each dietary treatment. Five isonitrogenous and isocaloric diets (Table 1) were formulated to contain 0.12, 0.18 and 0.30 % NPP diet with the two lowest NPP (0.12 and 0.18%) diets supplemented with microbial phytase (Adventis Biolab, Bengaluru) with the activity of 500 IU per kg diet. Each experimental diet was offered @160gm/bird/day for 20 weeks. A continuous 500 lux 16 h light per day was provided using incandescent bulbs. All the birds were maintained under uniform manage mental conditions throughout the experimental period of 29 to 48 weeks of age, which is divided into 5 phases of 4 weeks duration each, each phase duration was 4 weeks. All treatments and procedures performed during the trial were approved by the Institutional Animal Ethics Committee of the university (KVAFSU, Bidar, Karnataka).

Table 1: Percent composition of feed ingredients in the basal layer diet

Ingredients	Layer Mash (Kg/100kg)
Maize	62.50
Soybean meal (46 %CP)	21.30
De-oiled rice bran	07.40
Common Salt	0.40
Oyster Shell grit	7.20
Dicalcium Phosphate	0.95
Mineral Mixture*	0.08
DL-Methionine	0.07
Vitamin premix**	0.10
Total	100.0
Nutrient Composition	
ME (Kcal/Kg) [a]	2749
Crude Protein (%) [b]	16.06
Calcium (%) [b]	3.05
Phosphorus (%) [b]	0.574
Lysine (%) [a]	0.872
Methionine (%) [a]	0.425

* Mineral mixture: Each 100gm contains Magnesium oxide-1.48 g., Ferrous sulphate-6 g., Copper sulphate-0.05 g., Manganese sulphate-0.04 g., Potassium iodide-0.001 g., Zinc sulphate-1 g. and Potassium chloride-17.09 g.

** Vitamin-mineral premix: Each 100gm contains Vitamin AD3(Vitamin A-10,00,000 IU/Kg,Vitamin D-2,00,000 IU/Kg)-0.165 g,Vitamin K3-0.103 g,Vitamin E-2.4 g,Thiamine mono nitrate-0.206 g, Riboflavin-0.513 g, Pyridoxine hydrochloride-0.309 g,Cyanocobalamine-0.00031 g, Folic acid-0.103 g,Niacin-4.124 g, Ca-D-Pantothenate-1.031 g, Biotin-1.5 g,Maltodextrine-89.545 g.

[a] Calculated value, [b] Analysed values.

Response criteria

Egg quality characteristics

Eggs laid during the last 3 consecutive days of each phase (26, 27 and 28th day of each phase) were pooled and 20 eggs from each replicate (60 eggs per treatment) were subjected to internal and external quality characteristics. The eggs collected for quality analysis were weighed with a precision of 0.01g using analytical balance and broken on the same day to assess the albumen index, yolk index as per the procedure of Romanoff and Romanoff (1949) [11] and Haugh unit score according to Haugh (1937) [6].

Fertility and hatchability

Fertile eggs collected from experimental birds reared in deep litter pens were labelled to identify treatment and replicate. Fertility and hatchability were calculated by the formula,

a. Fertility percentage

Fertility (%) =Number of eggs fertile/Total number of eggs set x100

b. Hatchability (%) on total eggs set

Hatchability (%) on total eggs set =Number of chicks hatched out/Total number of eggs set x100

c. Hatchability (%) on fertile eggs set

Hatchability (%) on fertile eggs set = Number of chicks hatched out/Total number of fertile eggs set x 100

Statistical analysis

The data collected on various parameters were subjected to Statistical analysis by one-way ANOVA using SPSS 20 software. Difference between the means were tested using tukeys test at 5% confidence level.

Results and Discussion

During Phase II and V, Egg weight (Table.2) between the treatments differed insignificantly (P>0.05). The addition of phytase to the diets having 0.12 % NPP significantly (P<0.05) increased the egg weight to the level on par with the control group in phase I and III. However, egg weight was significantly (P<0.05) higher in phase IV in phytase supplemented treatment groups (T4 and T5) compared to unsupplemented groups (T1, T2 and T3). Thus, the lowest NPP content (0.12 %) with 500 IU/kg phytase was sufficient/optimum to support the normal egg weight in phase I, III and IV. Similar findings have also been reported in the literature by Boling *et al.*, (2000) [3], Scott *et al.*, (1999) [12], Yossef *et al.*, (2001) [16] and Keshavarz, (2000) [7]. Albumen index (Table.3), Yolk index (Table.4) and haugh unit (Table.5) score are influenced by neither NPP level nor phytase supplementation. However, current study showed that lowest NPP level of 0.12% is sufficient to maintain above indices similar to control and phytase supplemented groups. Fertility (Table.6) and hatchability (Table.7) are not significantly (P<0.05) influenced by Phytase supplementation in the present study may be due to Optimum phosphorus (0.12% NPP), calcium (3%) and similar protein and energy levels in all the diets. There were no significant overall effects of phytase on fertility, which suggested that the effective crude protein intake for the females had not been significantly increased. Walsh and Brake (1997 and 1999) [14, 15] demonstrated that low cumulative crude protein intake during rearing of females and reduction in fertility, inspite of 0.45%NPP without phytase enzyme in the diet. The results of this study indicated that, even at 0.45 and 0.50% NPP level in the diet, fertility and hatchability are not sensitive to dietary NPP. For this reason, differences in fertility due to phytase supplementation would not be expected. Hatchability, like fertility, did not appear to be sensitive to dietary NPP levels as fed in this experiment. Contrary to the findings of the present study, Berry *et al.*, (2003) [1] reported reduced fertility with 300FTU/kg phytase at 0.1% NPP in broiler breeders. Hassanien *et al*., (2015) [5] showed that fertility % was increased from 82.2 to 85.1% and hatchability from 71.9 to 77.9% in groups of Hubbard breeders supplemented with 500 g/ton of enzymes mixture. In agreement to the current findings Bhanja *et al.*, (2005) [2] stated that the hatchability was not influenced by either increasing the NPP content or supplementing the enzyme phytase.

Swarnadhara breeders on 0.12% dietary NPP without phytase were able to package sufficient resources for embryonic development. Our findings indicated that fertility and hatchability in Swarnadhara breeders were not responsive to either dietary phytase supplementation or NPP levels. The reason can be explained by the fact that the key nutrients supporting hatchability (protein, vitamins and micro minerals) are supplemented at levels as per the requirement of breeders. Therefore, enzyme supplementation or dietary NPP may not improve the hatchability to a great extent.

Table 2: Effect of Phytase supplementation on egg weight in Swarnadhara breeders

Treatment	Phase I	Phase II	Phase III	Phase IV	Phase V
T1	57.99±0.60ab	59.88±0.66	62.40±0.59ab	62.81±0.63b	57.99±0.60
T2	56.31±0.51b	57.85±0.57	57.16±0.59 c	62.03±0.55 b	57.11±0.56
T3	58.06±0.56ab	57.91±0.60	60.82±0.69 b	62.37±0.60 b	58.06±0.56
T4	59.05±0.71a	59.76±0.70	63.96±0.78 a	65.99±0.69 a	59.05±0.71
T5	57.24±0.62ab	59.62±0.74	60.16±0.64 b	65.88±0.96 a	57.24±0.62

Means with dissimilar superscripts (a, b, c…) indicate significant difference between treatments within phase. (P>0.05)

Table 3: Effect of Phytase supplementation on albumen index in Swarnadhara breeders

Treatment	Phase I	Phase II	Phase III	Phase IV	Phase V
T1	0.079±0.00ab	0.085±0.00	0.095±0.00 a	0.090±0.00	0.079±0.002ab
T2	0.080±0.00ab	0.081±0.00	0.088±0.00 ab	0.087±0.00	0.082±0.002ab
T3	0.075±0.00b	0.080±0.00	0.082±0.00 b	0.086±0.00	0.075±0.002b
T4	0.083±0.00 a	0.081±0.00	0.085±0.00 ab	0.089±0.00	0.083±0.001a
T5	0.082±0.00ab	0.081±0.00	0.084±0.00 b	0.092±0.00	0.082±0.003a

Means with dissimilar superscripts (a, b, c…) indicate significant difference between treatment within phase. (P>0.05)

Table 4: Effect of Phytase supplementation on yolk index in Swarnadhara breeders

Treatment	Phase I	Phase II	Phase III	Phase IV	Phase V
T1	0.43±0.004	0.45±0.003	0.46±0.004	0.47±0.003a	0.43±0.003
T2	0.43±0.005	0.44±0.004	0.45±0.004	0.46±0.006ab	0.43±0.004
T3	0.43±0.005	0.44±0.002	0.44±0.003	0.47±0.003a	0.43±0.005
T4	0.43±0.004	0.44±0.002	0.45±0.006	0.46±0.002a	0.43±0.004
T5	0.43±0.005	0.44±0.003	0.45±0.006	0.44±0.006b	0.43±0.002

Means with dissimilar superscripts (a, b, c…) indicate significant difference between treatment within phase. (P>0.05)

Table 5: Effect of Phytase supplementation on Haugh Unit Score in Swarnadhara breeders

Treatment	Phase I	Phase II	Phase III	Phase IV	Phase V
T1	74.11±0.75	78.04±0.72	81.40±1.06	86.53±0.71	74.11±0.76
T2	74.37±0.88	74.49±0.55	79.80±0.81	84.89±0.56	74.38±0.88
T3	74.06±0.85	75.82±0.55	80.27±0.97	86.40±0.63	74.07±0.85
T4	74.42±0.86	76.78 ±0.77	80.71±0.93	86.16±0.54	74.42±0.87
T5	73.77±0.78	76.49±0.88	79.49±0.92	85.24±1.01	73.78±0.79

Abcd: between treatment within phase

Table 6: Effect of Phytase supplementation on Fertility in Swarnadhara breeders

Treatment	Phase I	Phase II	Phase III	Phase IV	Phase V
T1	94.11±0.25	94.37±1.28	92.19±0.11	92.22±2.24	91.95±0.51
T2	93.95±1.46	94.28±0.28	93.48±2.19	93.08±1.03	94.58±1.06
T3	93.56±1.58	97.36±2.00	94.87±0.87	90.36±2.45	92.34±1.57
T4	89.34±5.96	93.98±1.17	94.26±1.03	92.25±2.39	93.87±2.40
T5	91.42±2.60	92.25±1.07	92.17±1.83	91.84±3.59	92.50±1.11

Table 7: Effect of Phytase supplementation on Hatchability (%) on Total Eggs Set in Swarnadhara breeders

Treatment	Phase I	Phase II	Phase III	Phase IV	Phase V
T1	85.53±1.70	86.35±2.47	84.64±0.33	80.57±3.33	77.56±3.14
T2	88.11±1.85	87.40±6.06	86.08±4.84	82.29±1.68	78.64±2.19
T3	86.53±7.04	91.47±2.93	89.45±1.63	79.47±1.73	80.77±8.77
T4	82.64±7.51	95.21±7.18	87.59±2.10	79.82±3.09	82.58±2.63
T5	80.19±3.21	83.76±2.35	85.26±3.63	83.87±5.51	79.46±2.40

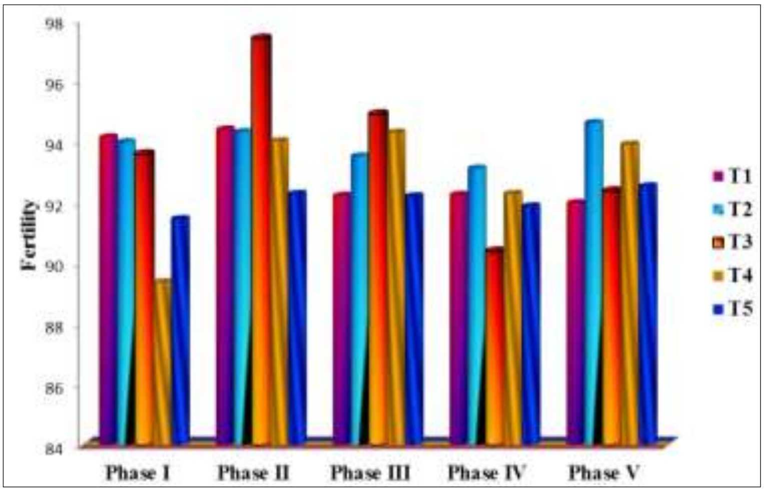

Fig 1: Effect of phytase supplementation on fertility in Swarnadhara breeders

Conclusion

Supplementing Swarnadhara breeder hen diets with phytase at 500 IU/kg in a diet containing 0.12% available phosphorus is sufficient to support egg production. Phytase supplementation can reduce addition of inorganic phosphate (NPP) in Swarnadhara breeder diets down to the 0.12% available phosphorus level, which is optimum to maintain egg production and egg quality, which in turn leads to economic feed formulations, as P is the third costly nutrient next to protein and energy. Based on the results of the experiment, it can be concluded that 180 mg of NPP /bird/day is adequate for Swarnadhara breeders to maintain optimum egg quality, fertility and hatchability.

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