J. Agric. Sci. Mansoura Univ., 29 (4): 1665 - 1676, 2004

EFFECTS OF DIFFERENT DIETARY LEVELS OF AVAILABLE PHOSPHORUS AND MICROBIAL PHYTASE ON THE PRODUCTIVE PERFORMANCE OF MAMOURAH LAYING HENS

Abou Khashaba, H. A.; M. A. Ibrahim. and M. A. M., sayed.
Animal Production Research Institute, Agriculture Research Center, Ministry of Agriculture, Dokki, Cairo, Egypt.

ABSTRACT

A 3 x 3 factorial experiment was conducted to determine the effects of different available phosphorus (AP) levels in diets of Mamourah laying hens with or without dietary microbial phytase (MP) supplementation on productive performance (egg production rate, egg weight, egg mass and feed conversion), egg quality (some exterior and interior parameters and Ca and P contents of egg shell), and some blood constituents (levels of plasma total protein, albumin and globulin, and activities of the transaminase enzymes: GOT and GPT). Tibia bone length and width and its content of Ca and P were also determined. Three isocaloric (ME of about 2760 kcal/kg)-isonitrogenous (CP of about 16 %) diets, containing AP levels of 0.405, 0.269 or 0.135 % were formulated with three levels of supplemental MP (0, 500 or 1000 U/kg diet). All birds had a free access to feed and water throughout the experimental period from 30 to 50 weeks of age. The obtained results can be summarized as follows:

1- Regardless of MP supplementation, reducing dietary AP level from 0.405 to 0.135% caused a significant increase in feed intake with concomitantly less efficient feed conversion, and significant reductions in eggshell weight and thickness, tibia bone width, and P content of eggshell, but increased the activities of plasma transaminases (GOT and GPT). In addition, egg production of the layers was decreased; but not significantly, with reducing dietary AP level.
2- Independently of the effect of dietary AP level, the dietary supplementation with MP resulted in significant improvements in egg production rate, egg weight, egg mass, feed conversion, and eggshell weight and thickness, as well as significant increases in tibia length and Ca content of the eggshell. Concentrations of total protein, albumin, Ca and P in the plasma of layers were significantly increased, whereas activities of GOT and GPT were reduced in response to dietary MP supplementation.
3- There were significant interactions between the effects of dietary AP and supplemental MP levels on most of the estimated parameters.

It can be concluded that supplementation of laying hen diets; in particular those containing low available phosphorus, with microbial phytase at a supplementary level of 500 U/kg diet, may improve the performance of laying hens for egg production and feed conversion, with no detrimental effect on egg quality.

INTRODUCTION

Generally, corn and soybean meal are the major feedstuffs in the diets of poultry. Unfortunately, many grains, oil seed meals and other plant ingredients contain high concentrations of phytic acid, amounting to 60% to 80% of their total phosphorus (Simons *et al.*, 1990). The phytate phosphorus is not available to poultry because they lack the endogenous digestive enzymes needed to hydrolyse phytate into inorganic phosphorus and inositol.

This limited ability of birds to utilize phytate P creates two problems to poultry nutritionists. The first problem concerns the formulation of diets that satisfy the birds' physiological need for phosphorus, and the second involves the environmental pollution by an increased quantity of dietary P which is not absorbed and being excreted by birds in their droppings. The use of exogenous phytase enzyme has been applied to reduce the need for inorganic phosphorus supplementation and to improve the utilization of the phosphorus present in plant feedstuffs (Sebastian *et al.*, 1998). Supplementation of laying hen diets with 250 U of phytase/kg diet has been found to be equivalent to 0.8 g of P from monocalcium phosphate (Van der Klis *et al.*, 1994). In a study with laying hens fed a corn-soybean basal diet, a supplemental level of 500 U phytase/kg diet was shown to have an effect equivalent to 1.0 g of inorganic P (Peter and Jeroch, 1993). Gordon and Roland (1997) reported that hens fed on a low nonphytate P (NPP)-diet with supplemental phytase, performed as well as hens fed diets containing higher levels of NPP without supplementary phytase. It is well established also that supplementing animal feeds with microbial phytase increases P availability. These beneficial effects of dietary phytase supplementation have been reported in broilers (Sohail and Roland, 1999). However, El Deeb *et al.* (2000) observed no effect of supplemental phytase on some physical measurements of bone (tibia length and width) in broiler chicks of 3 or 6 weeks of age. Comparatively, little work has been published concerning the response of laying hens performance to dietary supplementation with phytase. Carlos and Edwards (1998) and Um and Paik (1999) reported some beneficial effects of supplemental phytase on most of production parameters of laying hens, and indicated that minerals' availability was improved and retention of Ca, P, Mg, Fe, and Zn was significantly increased by dietary supplementation with phytase.

Many reports indicated that the minimum requirement of P for laying hens is below 0.50% of the diet (Day *et al.*, 1987). Abdallah *et al.* (1993) found that increasing total P in the diet of laying hens from 0.55 to 0.90% had no significant effect on egg production, shell weight, or feed intake. Feeding of pullets with diets containing non-phytate P (NPP) at levels ranged from 0.45 to 0.15% of the diet through three stages of a laying cycle from 25 to 72 weeks of age, had no significant effect on their laying performance compared with that of their control counterparts fed a 0.454% NPP-diet continuously (Scheiderler and Sell, 1986; Sell *et al.*, 1987). Supplementing microbial phytase to corn-soya diets containing low P level improved P and amino acids digestibility (Rutherfurd *et al.*, 2002) and increased Ca, Mg and Zn availability (Viveros *et al.*, 2002). Bragg *et al.* (1971) studied the transfer of calcium from the diet to the egg shell using ^{45}Ca as a tracer. These investigators suggested that 60 to 70% of egg shell calcium was derived from the diet.

This experiment was conducted to determine the effect of dietary supplementation with microbial phytase on performance, egg production, exterior and interior egg quality, bone parameters and some blood constituents in Mamourah laying hens fed different dietary levels of available phosphorus.

MATERIALS AND METHODS

This study was carried out at El-Gimmizah Poultry Station, Animal Production Researches Institute, Ministry of Agriculture. A total number of 180, 30-week-old Mamourah laying hens and 18 cockerels were randomly distributed into nine equal groups, each of which composed of 2 replicates of 10 females and 1 male each. Birds were housed in floor pens with free access to feed and water, and were subjected to a total of 16 hr continuous light/day. Three corn-soy basal diets were formulated as shown in Table 1.

Table (1): Composition and chemical analyses of the basal diets.

Ingredients (%)	Diet (1) normal 0.405%AP	Diet (2) 0.269% AP	Diet (3) 0.135% AP
Yellow corn	66.50	66.50	66.00
Soybean meal, 44 %	23.75	23.50	23.05
Wheat bran	00.00	0.60	1.90
Limestone	7.50	7.90	8.30
Dicalcium phosphate	1.60	0.85	0.10
Common salt (NaCl)	0.30	0.30	0.30
Vit. & Min. mix.*	0.25	0.25	0.25
Methionine	0.10	0.10	0.10
Total	100	100	100
Calculated values**:			
Crude protein, %	16.35	16.33	16.30
ME,Kcal/kg	2760.97	2763.20	2753.31
Calcium, %	3.33	3.32	3.31
Available phosphorus, AP %	0.40	0.27	0.13
Lysine, %	0.81	0.81	0.80
Methionine,%	0.37	0.36	0.34
Methionine + cysteine %	0.64	0.64	0.64
Determined values***: (Dry matter basis)			
Dry matter, %	89.48	88.68	89.24
Crude protein,%	15.98	15.78	15.46
Crude fiber,%	3.12	3.17	3.27
Ether Extract,%	2.72	2.73	2.75
NFE%	70.82	70.11	70.15
Ash,%	7.36	8.21	8.37

*Each 3kg contains: 10,000,000 IU Vit. A; 2,000,000 IU Vit D3 10,000 mg Vit. E;1,000mg Vit. K; 1,000mg Vit. B1; 5,000mg Vit. B2; 1,500mg Vit B6; 10mg Vit. B12; 50mg; Niacin, 20 gm ; Panatothenic acid, 1gm, Biotin;1,000mg Folic acid;250,000mg choline chloride; 80g manganese; 40g iron; 40g zinc; 2g copper; 2g iodine; 1gm Seleinium and 2g cobalt.
** Calculated according to NRC (1984).
*** Determined according to the methods of A.O.A.C (1980)

These diets were formulated to be isocaloric (ME of about 2760 kcal/kg) and isonitrogenous (crude protein of about 16 %), and to contain three dietary available phosphorous levels (0.405, 0.269 or 0.135%) with three supplemental phytase levels (0, 500 or 1000 U/kg diet). These experimental diets were given to the birds during the period from 30 to 50 weeks of age. Pullets were weighed at the beginning and at the end of the experimental period. Eggs number, egg weight and egg mass were recorded daily through the experimental period (20 weeks). Feed consumption was recorded weekly and then feed conversion (g feed/g egg) was calculated. Egg quality was evaluated at 4-week intervals. In each test, 10 eggs from each dietary treatment were collected, individually weighed and broken-out in order to separate their shells, yolks and albumens. The individual weights of yolk, albumen and shell (with membranes) were recorded and calculated as percentages of egg weight. Also, Ca and P concentrations in eggshells ash were determined (A.O.A.C,1980).

At the end of study, three hens from each treatment were slaughtered in order to measure the length and width of right tibia. The bones were dried, crushed and then, ashed at 600 °C for 16 hours in a muffle furnace. Calcium and phosphorus were determined according to A.O.A.C (1980). Blood samples were individually collected during slaughtering in heparinized tubes, then plasma samples were obtained by centrifugating the tubes at 4000 rpm for 15 minutes and stored at –20 °C until analysis. Commercial kits were used to measure the levels of total protein (TP) (Doumas, 1975) and albumin (A) (Doumas *et al.*, 1971) in blood plasma, then globulin (G) concentration, and albumin/globulin (A/G) ratio were calculated. Levels of plasma Ca and inorganic P and activities of plasma glutamic oxaloacetic transaminase (GOT) and glutamic pyruvic transaminase (GPT) were determined (Reitman and Frankel, 1957). Concentrations of Ca (Moorhead and Biggs, 1974) and inorganic P (Glodenberg and Fernandez, 1966) in blood plasma of the experimental laying hens were also determined.

A completely randomized design, with a factorial arrangement of treatments (3x3), was used. Data were analyzed using the General Linear Model (GLM) procedure of Statistical Analysis System (SAS, 1994). Significant differences among means were separated by Duncan"s multiple range test (Duncan, 1955).

RESULTS AND DUSCISION

Performance for egg production and feed conversion:

Results in Table 2 indicated that irrespective of the effect of supplemental phytase (MP), layers fed the lowest dietary level (0.135%) of available phosphorus (AP) put on a significantly ($P \leq 0.05$) less body weight gain compared to those fed either 0.269 or 0.405% AP-diets. Regardless of the effect of dietary AP level, layers fed on diets supplemented with the microbial phytase (MP) at inclusion rate of either 500 or 1000 U/kg, attained similar weight gains, but significantly ($P \leq 0.05$) higher than that of layers fed on diets without supplementary MP (0.0 U/kg). Both effects of dietary AP and supplemental MP were interacted on layers body weight gain. On the contrary, Yossef *et al.* (2001) reported that there was no significant effect of dietary phosphorus (AP) level, with or without phytase supplementation, on body weight of laying hens. However, it should be pointed out that hens do not necessarily have to gain weight throughout the laying period, rather than maintaining their body weight, in order to perform satisfactorily. In this respect, Summers and Leeson (1977) stated that any gain in body weight after commencement of egg production should be minimal as the hen is essentially at her mature body weight.

Table (2): Effects of different dietary levels of available phosphorus and microbial phytase on the performance of Mamourah laying hens for egg production and feed conversion, throughout the experimental period from 30 to 50 weeks of age.

Variables	Initial body weight; kg	Final body weight; kg	Body weight gain; kg	Daily feed intake; (g)	Egg Number	Hen-day egg production (%)	Egg weight; (g)	Daily egg mass; (g)	Feed conversion (g feed/g egg)
Dietary available phosphorus levels(AP)	NS	*	NS	*	NS	NS	NS	NS	*
(0.405%)	1.44	1.77a	0 33	112 92b	74 98	53.56	53.03	28.40	3 98b
(0.269%)	1.43	1 71ab	0 28	116.24ab	73 75	52.78	52.68	27.75	4.19b
(0.135%)	1 43	1.67b	0.24	122.58a	70 31	50.22	50.22	25.22	4.86a
SEM	0 01	0.01	0.02	1.50	1.32	0.94	0.28	0.81	0.10
Supplemental phytase levels (MP)	NS	*	*	NS	*	*	*	*	*
(0 U/kg)	1.44	1.65b	0.21b	114.03	66.38b	47.41b	51.69b	24.51b	4.65a
(500 U/kg)	1.43	1.73a	0.30a	118.35	75.27a	53.76a	52.88ab	28.43a	4.09b
(1000 U/kg)	1.43	1.78a	0.33a	121.38	77 39a	55.28a	53.27a	29.45a	4.12b
SEM	0.01	0 01	0.02	1.50	1.32	0.94	0.28	0.81	0.10
APxMP interaction	NS	*	*	**	**	**	NS	**	**
0.405%*0 U/kg	1.44	1.69	0.25	112.71	67.44	48.17	52.26	25.17	4.48
0.269%*0 U/kg	1.43	1.68	0.24	108.58	66 70	47 84	51.59	24.58	4.42
0.135%*0 U/kg	1.44	1.59	0.17	120.81	64 99	48.42	51.23	23 73	5.08
0.405%*500 U/kg	1.43	1.76	0.33	114.53	77 78	55.58	53.09	29.49	3.88
0.269%*500 U/kg	1.42	1 74	0.31	115.65	74.07	53.55	53.02	28.39	4.07
0.135%*500 U/kg	1.44	1 69	0.26	118.88	73.06	52 18	52.52	27.41	4.34
0.405%*1000 U/kg	1.44	1.83	0.39	111.53	79.72	56.94	51.59	29.38	3.79
0.269%*1000 U/kg	1 43	1.73	0.29	124.81	79 80	56.84	53.72	30.62	4.08
0.135%*1000 U/kg	1 43	1.71	0.29	128.05	72 87	52.05	52.35	27.25	4.70
SEM	0.08	0.05	0.02	1 50	1 32	0.94	0 28	0 85	0.07

a-b : For each criterion, means in the same column bearing different superscripts differ significantly

Irrespective of the effect of MP supplementation, dietary AP level had no significant effect on eggs number (EN), hen-day egg production rate (EPR), egg weight (EW) or daily egg mass (EM). However, daily feed intake (FI) and feed conversion (FC) of layers were significantly ($P \leq 0.05$) affected by the dietary AP level. Layers fed on the 0.135% AP-diets consumed more feed and exhibited a significantly ($P \leq 0.05$) poorer feed conversion compared to those fed the 0.269 or 0.405% AP-diets (Table 2). Independently from the effect of dietary AP level, results in Table 2 showed that layers fed on diets supplemented with MP at inclusion rate of either 500 or 1000 U/kg performed equally for EN, EPR, EW, EM and FC, and achieved a significantly ($P \leq 0.05$) better performance than layers fed diets without supplementary MP (0.0 U/kg). There were significant interactions between the effects of dietary AP and supplemental MP levels on EW, EPR, EM, FI and FC (Table 2).

These results are in agreement with those reported by Summers *et al.* (1976) who found that feeding the laying hens on low AP-diet had no significant effect on egg weight. Yossef *et al.* (2001) found that eggs number and egg mass were increased in response to phytase supplementation to low AP-diets of laying hens. Gordon and Roland (1997) reported that feeding of laying hens with 0.1% AP-diet decreased egg production and feed consumption compared to 0.2 to 0.5% AP-diet, but 0.1 % AP-diet supplemented with 300 U of phytase/kg diet completely corrected the adverse effects. Other reports indicated that the adverse effect of low AP-diets on laying hens performance can be completely alleviated in the presence of phytase in the diet (Punna and Roland, 1999; Keshavarz, 2000). Even though, in the present study no differences were detected in the performance of layers fed diets supplemented with MP at levels of 500 or 1000 U/kg, other reports in the literature indicated that, dietary supplementation with phytase at a level of 300 U/kg, was more effective than the lower supplementary phytase level (150 U/kg diet) in restoring the performance of laying hens fed low-P (0.25 - 0.15% AP) diets to that of hens fed a control diet (0.45% AP) (Keshavarz, 2003).

Egg Quality Traits:

Regardless of the effect of supplemental MP, results in Table 3 showed that increasing dietary level of AP to 0.405% significantly ($P \leq 0.05$) improved most of the external and internal egg quality traits. With the exception of albumen and yolk percentages, layers fed the 0.135% AP-diets produced eggs of significantly ($P \leq 0.05$) lower values for egg shape index, shell weight %, shell thickness and yolk index, compared to those produced by layers on the 0.405% AP-diets. On the other hand, layers fed on the 0.269% AP-diets produced eggs of significantly ($P \leq 0.05$) higher yolk weight %, and egg shape and yolk indices, but had similar shell weight % and shell thickness compared to those laid by layers on the 0.135% AP-diets.

Table (3): Effects of different dietary levels of available phosphorus and microbial phytase on external and internal egg quality traits[1] of mourah laying hens.

Variables	Egg shape index (%)	Shell weight (%)	Shell thickens mmx100	Albumen weight (%)	Yolk weight (%)	Yolk index (%)	Haugh Units
Dietary available phosphorus levels(AP)	*	*	*	*	*	*	NS
(0.405%)	77.89a	13.72a	37.08a	54.79a	31.83b	43.02a	82.59
(0.269%)	75.56b	13.04b	34.44b	51.21b	33.88a	43.62a	83.35
(0.135%)	74.27c	12.78b	34.74b	58.42a	30.79b	39.96b	81.71
SEM	0.23	0.08	0.19	0.74	0.39	0.33	0.46
Supplemental phytase levels (MP)	*	*	*	NS	*	*	NS
(0 U/kg)	74.31b	12.73b	34.75b	53.28	32.09ab	43.24a	81.67
(500 U/kg)	76.58a	13.49a	35.80a	53.29	33.46a	41.60b	82.98
(1000 U/kg)	76.82a	13.32a	35.69a	55.85	30.94b	41.75ab	83.02
SEM	0.23	0.08	0.19	0.74	0.39	0.33	0.46
APxMP Interaction	*	**	**	*	**	*	*
0.405%*0 U/kg	77.66	13.30	38.85	54.87	32.02	42.80	80.96
0.269%*0 U/kg	73.07	12.68	33.76	44.48	36.88	46.40	85.50
0.135%*0 U/kg	72.20	12.21	33.65	60.40	27.38	40.53	76.56
0.405%*500 U/kg	78.00	14.03	37.42	54.82	31.88	43.07	84.02
0.269%*500 U/kg	76.80	13.45	35.28	51.37	35.18	42.13	82.52
0.135%*500 U/kg	74.93	12.99	34.72	53.68	33.32	39.60	82.34
0.405%*1000 U/kg	78.00	13.82	38.98	54.59	31.1	43.20	82.81
0.269%*1000 U/kg	76.90	13.01	34.29	57.77	29.55	42.33	82.01
0.135%*1000 U/kg	75.67	13.13	35.82	5.19	31.68	39.73	84.23
SEM	0.23	0.08	0.19	0.75	0.33	0.33	0.46

a-c : For each criterion, means in the same column bearing different superscripts differ significantly. 1: Means represent an average of five egg quality tests.

Dietary AP level had no significant effect on Haugh Units score. Independently from the effect of dietary AP level, supplemental MP at inclusion rate of 500 or 1000 U/kg significantly (P≤0.05) increased the egg shape index, and eggs shell weight % and thickness, but had no significant effect on albumen weight % or Haugh Units score. Erratic significant (P≤0.05) differences were observed, however, in yolk weight % and yolk index due to the effect of supplementary MP level. Effects of dietary AP and supplemental MP levels on egg quality traits were also interrelated.

These results are in partial agreement with those reported by Gordon and Roland (1998). On the other hand, Yossef *et al.* (2001) fed Gimmizah pullets on diets containing 0.25 or 0.40% AP supplemented with phytase at levels of 0, 600 or 1000 U/kg, and found that interior egg quality and shell thickness were not affected by either dietary AP or supplemental phytase levels. Lim *et al.* (2003) concluded that, supplementation of microbial phytase at a level of 300 U per kg diet of laying hens can Improve egg production, and decrease the percentage of broken and soft-shell eggs, but the effects of phytase supplementation were modified by the dietary levels of Ca (0.3 and 0.4%) and NPP (0.15 and 0.25%), during an experimental period from 21 to 41 weeks of age.

Blood Parameters:

The effects of dietary AP and supplemental MP levels on some blood constituents of 50-week-old Mamourah laying hens are presented in Table 4. Independently of the effect of supplemental MP, no significant differences were observed in the levels of plasma total protein (TP), albumin (A), globulin (G) and A/G ratio, or in plasma Ca and P concentrations due to the effect of dietary AP level. However, the activities of plasma GOT and GPT were significantly (P≤0.05) increased with reducing dietary AP level to 0.135% of the diet. Regardless of the effect of dietary AP level, MP supplementation at either 500 or 1000 U/kg diet significantly (P≤0.05) increased the levels of plasma TP, A, Ca and P, but significantly (P≤0.05) reduced the activities of plasma GOT and GPT. There were significant interactions between the effects of dietary AP and supplemental MP levels, only for plasma TP, G, A/G ratio, Ca and P levels.

These results are in partial keeping with those of Yossef *et al.* (2001) who reported that plasma Ca and P concentrations were significantly increased with addition of phytase to laying hen diets. On the contrary, Triyuwanta *et al.* (1992) reported an increase in plasma Ca as a result of feeding laying hens diets containing various levels of available P ranged from 0.44 to 0.64%. Furthermore, several studies conducted with laying hens, had indicated that plasma P is positively correlated with dietary P level, (Zumbado and Britton; 1983, Keshavarz, 1986; Triyuwanta *et al.*, 1992). On the other hand, Abdallah *et al.* (1993) and Pan *et al* (1998) reported that concentration of Ca or P in plasma of laying hens could be influenced by the level of dietary inorganic P supplementation. Edwards (1993) and Roberson and Edwards (1994) reported that supplementation of chicks diet with microbial phytase had no significant effect on plasma Ca. Generally, discrepancies may exist among results of the different studies concerning the changes which may occur in blood constituents of birds, as it was observed in the present case.

These discrepancies could be attributed to differences in quantitative and qualitative composition of diets, nutritional status of birds, feed intake, strain and age of birds, time of blood sampling, levels and types of dietary Ca, P and supplemental phytase, or stage of egg production.

Table (4): Effects of different dietary levels of available phosphorus and microbial phytase on blood plasma constituents in 50-week-old Mamourah laying hens.

Variables	Total protein (g/dl)	Albumin (g/dl)	Globulin (g/dl)	A/G ratio	GOT (U/L)	GPT (U/L)	Calcium (mg/dl)	Phosphorus (mg/dl)
Dietary available phosphorus levels(AP)	NS	NS	NS	NS	*	*	*	NS
(0.405%)	4.66	1.87	2.79	0.70	181.24b	22.25b	23.09a	6.35
(0.269%)	4.21	1.85	2.40	0.76	188.07ab	22.98ab	16.68b	5.75
(0.135%)	4.05	1.76	2.50	0.71	189.28a	23.80a	12.59b	5.03
SEM	0.14	0.05	0.10	0.02	1.49	0.25	1.27	0.29
Supplemental phytase levels (MP)	*	*	NS	NS	*	*	*	*
(0 U/kg)	3.70b	1.66b	2.38	0.72	198.77a	24.61a	13.23b	4.41b
(500 U/kg)	4.52a	1.84ab	2.63	0.72	181.81b	22.33b	18.37ab	6.06a
(1000 U/kg)	4.71a	1.96a	2.68	0.74	180.01b	22.09b	20.77a	6.67a
SEM	0.14	0.05	0.10	0.02	1.49	0.25	1.27	0.29
APxMP Interaction	*	NS	*	*	NS	NS	*	*
0.405%*0 U/kg	3.84	1.76	2.08	0.86	190.56	24.41	16.85	4.41
0.269%*0 U/kg	3.81	1.59	2.4	0.63	199.87	24.89	12.12	4.69
0.135%*0 U/kg	3.46	1.62	2.59	0.52	199.87	24.53	10.71	4.13
0.405%*500 U/kg	5.06	1.83	3.23	0.58	177.98	21.46	23.40	6.90
0.269%*500 U/kg	4.16	1.89	2.82	0.81	182.56	22.07	18.18	5.92
0.135%*500 U/kg	4.33	1.81	2.39	0.77	184.90	23.47	13.51	5.35
0.405%*1000 U/kg	5.09	2.03	3.05	0.66	175.19	20.89	29.01	7.75
0.269%*1000 U/kg	4.67	1.97	2.45	0.80	181.77	21.99	19.73	6.65
0.135%*1000 U/kg	4.37	1.85	2.52	0.75	183.06	23.40	13.55	5.61
SEM	1.14	0.01	0.10	0.02	2.82	0.40	0.12	0.02

a-b : For each criterion, means in the same column bearing different superscripts differ significantly.

Bone minerals:

Data of the effects of dietary AP and supplemental MP levels on tibia length and width and its Ca and P contents in the 50-week-old experimental layers are summarized in Table 5. Regardless of the effect of dietary AP levels, no significant differences were observed in tibia length or in its Ca and P contents due to the effect of dietary AP level. However, a significant decrease in tibia width was occurred with lowering AP levels in the diets. Independently of the effect of dietary AP level, supplementation of MP to the layers diets significantly ($P \leq 0.05$) increased their tibia length. However, no significant differences were observed in layers tibia width or its Ca and P contents due to the effect of dietary MP supplementation. There were significant interactions between the effects of dietary AP and supplemental MP levels, only for tibia length and tibia width.

Table (5): Effects of different dietary levels of available phosphorus and microbial phytase on Ca and P contents of tibia bone and egg shell ash of Mamourah laying hens.

Variables	Tibia				Egg Shell Ash	
	Tibia length (cm)	Tibia Width (mm)	Ca (%)	P (%)	Ca (%)	P (%)
Dietary available phosphorus levels(AP)	NS	*	NS	NS	NS	*
(0.405%)	12.37	8.58a	24.02	10.41	28.18	0.46a
(0.269%)	11.88	8.81ab	23.68	9.56	26.14	0.34b
(0.135%)	11.70	8.08b	22.67	9.32	24.89	0.31b
SEM	0.17	0.09	0.51	0.29	0.17	0.03
Supplemental phytase levels (MP)	*	NS	NS	NS	*	NS
(0 U/kg)	11.40b	8.08	22.07	9.13	21.65b	0.29
(500 U/kg)	12.17ab	8.42	24.23	9.88	28.89a	0.39
(1000 U/kg)	12.38a	8.60	24.08	10.29	28.64a	0.42
SEM	0.17	0.08	0.51	0.29	0.17	0.03
APxMP Interaction	*	*	NS	NS	*	NS
0.405%*0 U/kg	11.60	8.25	22.94	9.21	22.33	0.30
0.269%*0 U/kg	11.50	8.25	22.53	9.09	21.38	0.30
0.135%*0 U/kg	11.10	7.75	20.75	9.08	21.25	0.28
0.405%*500 U/kg	12.20	8.75	24.48	10.47	30.92	0.48
0.269%*500 U/kg	12.00	8.50	24.23	8.91	29.33	0.38
0.135%*500 U/kg	12.00	8.25	23.99	9.25	26.44	0.32
0.405%*1000 U/kg	13.00	8.75	24.64	11.56	31.24	0.59
0.269%*1000 U/kg	12.15	8.50	24.28	9.88	27.72	0.33
0.135%*1000 U/kg	12.00	8.50	23.26	9.62	26.97	0.33
SEM	0.17	0.02	0.51	0.29	1.17	0.02

a-b : For each criterion, means in the same column bearing different superscripts differ significantly.

Egg shell minerals:

Results in Table 5 indicated that, regardless of dietary MP supplementation a significant ($P \leq 0.05$) decrease in egg shell Ca and P contents was observed with decreasing dietary AP levels. Irrespective of the effect of dietary AP level, dietary MP supplementation significantly ($P \leq 0.05$) increased the Ca content of eggshell. Also, eggshell P content was increased, but not significantly, due to the effect of dietary MP supplementation. A significant interaction was observed between the effects of dietary AP and supplemental MP levels for egg shell Ca contents.

It can be concluded that supplementation of laying hens diets; in particular those containing low available phosphorus, with microbial phytase at a supplementary level of 500 U/kg diet, may improve the performance of laying hens for egg production and feed conversion, with no detrimental effects on egg quality.

REFERENCES

Abdallah, A.G.; R.H. Harms; and O. El-Husseiny, (1993). Performance of hens laying eggs with heavy or light shell weight when fed diets with different calcium and phosphorus levels. Poultry Sci. 72:1881-1891.

A.O.A.C; Association of Official Analytical Chemists, (1980). Official Methods of Analysis. 13th Washington, D.C.USA.

Bragg, D.B.; J. Floyd and E.L. Stephenson (1971). Factors affecting the transfer of Calcium (^{45}Ca) from the hens diet to the egg shell. Poultry Sci 50:167-178.

Carlos, A.B, and H.M. Edwards, Jr. (1998). The effects of 1,25 dihydroxycholecalcaiferol and phytase on the natural phytate phosphorus utilization by laying hens. Poultry. Sci. 77:850-858.

Day, E.J; B.C. Dilworth and S, Omar (1987). Effect of varying levels of phosphorus and live yeast culture in caged layer diets. Poultry. Sci. 66:1402-1410.

Doumas, B.T. (1975). Standards for total serum protein assays. A collaborative study. Clin. Chem. 21: 1159 – 1166.

Doumas, B.T.; W.A. Watson and H.G. Biggs (1971). Albumin standards and the measurement of serum albumin with bromocresol green. Clin. Chim. Acta, 31: 87 – 96.

Duncan, D. (1955). Multiple range and multiple F tests. Biometrics, 11:1-42. edition, Assoc. Offic. Anal. Chem., Arlington, VA. USA.

Edwards, H.M., Jr.(1993). Dietary 1,25-dihydroxycholecalciferol supplementation increases natural phytate phosphorus utilization in chickens. Journal of Nutrition. 123:567-577.

El Deeb, Mariam A.; H.H. Sharara and M.N. Makled (2000). Enhance calcium and phosphorus utilization by enzyme phytase supplemented to broiler diet contained rice bran. Egypt. Poultry Sci, 20:545-566.

Goldenberg, H. and A. Fernandez (1966). Simplified method for estimation of inorganic phosphorus in body fluids. Clin. Chem. , 12:871-882.

Gordon, R.W. and D.A., Roland, Sr.(1997). Performance of commercial laying hens fed various phosphorus levels, with and without supplemental phytase . Poultry Sci. 76:1172-1177.

Gordon, R.W. and D.A., Roland, Sr. (1998). Influence of supplemental phytase on calcium and phosphorus utilization in laying hens. Poultry Sci. 77:290-294

Keshavarz, K. (1986). Interaction between calcium and phosphorus in laying hens. Nurt. Rep. Int. 36:9-20.

Keshavarz, K. (2000). Nonphytate phosphorus requirement of laying hens with and without phytase on a phase feeding program. Poultry Sci 79:748-763.

Keshavarz, K. (2003). The effect of different levels of nonphytate phosphorus with and without phytase on the performance of four strains of laying hens. Poultry Sci, 82:71-91

Lim H.S, Namkung H, Paik I.K. (2003). Effects of phytase supplementation on the performance, egg quality, and phosphorous excretion of laying hens fed different levels of dietary calcium and nonphytate phosphorous. Poultry Sci., 82:92-99.

Moorhead, W.R.and H.G. Biggs (1974). 2-amino-2-methyl-1-propanol as the alkalizing agent in an improved continuous flow cresolphthalein complexone procedure for calcium in serum. Clin. Chem.,20:1458-1460.

National Research Council, (N.R.C, 1994). Nutrient Requirements of Poultry .9th revised ed. National Academy Press., Washington, D.C.,USA.

Pan, C.F.; F.A Igbasan,.; W Guenter,.; and R.R Marquardt,.(1998). The effects of enzyme and inorganic phosphorus supplements in wheat and raye based diets on laying hens performance, energy and availability. Poultry Sci. 77:83-89.

Petre, W. and H. Jeroch, (1993). The effectiveness of microbial phytase addition to layer rations on maiz and wheat basis. Page 206-209 in, Enzymes in Animal Nutrition. Proceeding of the 1st Symposium. Edited by Wenk, C. and M. Boessinger, Roch, kartause Ittingen, Switzerland.

Punna, S. and D.A. Roland, Sr (1999). Influence of supplemental microbial phytase on first cycle laying hens fed phosphorus-deficient diets from day one of age. Poultry Sci 78:1407-1411.

Reitman, S. and S. Frankel (1957). A colorimetric method for the determination of serum glutamic-oxaloacetic and glutamic-pyrovic transaminases. Amer. J. Clin. Path., 28: 56-63.

Roberson, K.D. and H.M Edwards. (1994). Effects of 1,25-dihydroxycholecalciferol and phytase on zinc utilization in broiler chicks. Poultry Sci 73:1312-1326 .

Rutherfurd, SM, T.K. Chung and P.J. Moughan (2002). The effect of microbial phytase on ileal phosphorus and amino acid digestibility in the boiler chicken. Br. Poultry Sci., 43:598-606.

SAS (1994). The SAS system for windows 3.11 Release 6.10. SAS Institute Inc., Cary, NC. Washington, D.C.USA.

Scheideler, S.E.; and J.L. Sell (1986). Effects of calcium and phase-feeding phosphorus on production traits and phosphorus retention in two strains of laying hens. Poultry.Sci.,65: 2110-2119.

Sebastian, S.; S.P. Touchburn and E.R. Chavez (1998). Implications of phytic acid and supplemental microbial phytase in poultry nutrition: A review. World'ss Poultry. Sci. J. 54:27-47.

Sell, J. L.; S.E. Scheideler and B.E. Rahn. (1987). Influence of different phosphorus phase-feeding programs and dietary calcium level on performance and body phosphorus of laying hens. Poultry .Sci. 66:1524-1530.

Simons, P.C.M.; H.A.J. Versteegh .; A.W. Jongbloed .; P.A. Kemme .; P.Slimp ; K.D. Bos.; M.G.E. Wolters. ; R.F. Beudeker and G.J. Verschoor.(1990). Improvement of phosphorus availability by microbial phytase in broilers and pigs. Brit. J. Nutr. 64:525-540.

Sohail, S.S. and D.A. Roland, Sr. (1999). Influence of supplemental phytase on performance of broilers four to six weeks of age. Poultry Sci. 78:550-555.

Summers, J.D.; R. Grandhi,; and S. Leeson, (1976). Calcium and phosphorus requirements of the laying hen. Poultry Sci., 55:402-413.

Summers, J.D. and S. Leeson (1977)." Poultry Nutrition Handbook". Guelph, Ontario, Canada.

Triyuwanta, C. leterrier; and Y. Nys (1992). Dietary phosphorus and food allowance o f d warf b reeders affect reproductive performance of hens and bone development of their progeny. Br. Poultry. Sci., 33:363-379

Um, J.S. and I.K. Paik (1999). Effects of microbial phytase supplementation on egg production, eggshell quality, and mineral retention of laying hens fed different levels of phosphorus. Poultry Sci., 78:75-79.

Van der Klis, J.D., H.A.J. Versteegh and C.W. Scheele, (1994). Practical enzyme use in poultry diets: phytase and NSP enzymes. Pages 113-128 in: BASF Technical Symposium during the Carolina Poultry Nutrition Conference, December 1994, Charlotte, NC.

Viveros, A; A. Brenes; , I Arija and C. Centeno (2002). Effects of microbial phytase supplementation on mineral utilization and serum enzyme activities in broiler chicks fed different levels of phosphorus. Poultry Sci 81(8):1172-1183.

Yossef Mervat, S.E.; M.A. Abaza; H.M. Yakout, A.A. Abdalla and Azza El-Sebai (2001). Influence of microbial phytase supplementation on performance parameters and blood constituents of Gimmizah hens fed different dietary phosphorus levels. Egypt Poultry Sci., 21:997-1020.

Zumbado, M.E.; and W.M Britton, (1983). The influence of feeding increasing levels of dietary phosphorus and sodium on the laying hen shell quality. Poultry Sci., 62:1528. (Abstract).

تأثير التغذية على علائق ذات مستويات مختلفة من الفوسفور المتاح و إنزيم الفيتيز على الأداء الإنتاجي لدجاج المعمورة البياض

حمدى عبد العزيز محمد أبو خشبة ، محمد عبد الباقى ابراهيم ، محمد أحمد سيد أحمد
معهد بحوث الإنتاج الحيوانى و الدواجن –مركز البحوث الزراعية –وزارة الزراعة

صممت هذه التجربة لدراسة تأثيرات استخدام مستويات مختلفة من الفوسفور المتاح و إنزيم الفيتيز فى علائق دجاج المعمورة البياض على المظاهر الإنتاجية (معدل إنتاج البيض- وزن البيض- كتلة البيض- معدل التحويل الغذائى) وكذلك بعض صفات الجودة الخارجية و الداخلية للبيض و محتوى رماد قشرة البيض من الكالسيوم و الفوسفور وبعض مكونات الدم (مستوى البروتين الكلى-الألبيومين- الجلوبيولين- ونشاط إنزيمات الكبد: GOT,GPT) وبعض الخصائص الطبيعية والكيميائية لعظمة الساق (الوزن ، الطول ، العرض ، ومحتوى رماد العظام من الكالسيوم والفوسفور). تم استخدام ٣ علائق متساوية فى محتواها من الطاقة الممثلة (حوالى ٢٧٦٠ ك كالورى/ كجم) ومن البروتين(حوالى ١٦%) وتحتوى على ٣ مستويات من الفوسفور المتاح (٠,٤٠٥% ، ٠.٢٦٩% أو ٠,١٣٥%) و ثلاث مستويات من إنزيم الفيتيز (٠ ، ٥٠٠ أو ١٠٠٠ وحدة/كجم عليقة).و قدمت العلائق التجريبية والمياه بحرية طوال فترة التجربة (٣٠-٥٠ أسبوع من العمر).

ويمكن تلخيص النتائج فيما يلى :

١- دون الأخذ فى الاعتبار مستوى إنزيم الفيتيز المضاف أدى تقليل مستوى الفوسفور المتاح من ٠,٤٠٥% الى ٠,١٣٥% إلى زيادة معنوية فى الغذاء المأكول وبالتالى حدث انخفاض معنوى فى كلا من معدل التحويل الغذائى وكذلك انخفض وزن قشرة البيض وسمك القشرة وعرض عظمة الساق ومحتوى رماد القشرة من الفوسفور . كذلك أدى خفض مستوى الفوسفور المتاح إلى ارتفاع ملحوظ فى نشاط إنزيمات الكبد (GOT,GPT) بينما انخفض إنتاج البيض بدرجة غير معنوية.

٢- بغض النظر عن تأثير مستوى الفوسفور للمتاح فى الغذاء أدت إضافة إنزيم الفيتيز إلى تحسن معنوى فى كلا من معدل إنتاج البيض- كتلة البيض- معدل التحويل الغذائى- وزن القشرة- سمك القشرة وكذلك زيادة معنوية فى طول عظمة الساق ومحتوى رماد القشرة من الكالسيوم وكذلك محتوى سيرم الدم من البروتين الكلى- الألبيومين- الكالسيوم- الفوسفور-بينما انخفض نشاط إنزيمات الكبد(GOT,GPT) معنويا بإضافة الأنزيم.

٣- أظهرت النتائج وجود تداخلات معنوية بين تأثيرات مستويات كلا من الفوسفور المتاح و إنزيم الفيتيز فى معظم المقاييس السابقة.

بناء على النتائج المتحصل عليها يمكن التوصية بأن إضافة إنزيم الفيتيز (بمعدل ٥٠٠ وحدة /كجم عليقة) الى علائق الدجاج البياض خاصة المنخفضة فى محتواها من الفوسفور المتاح يمكن أن يحسن من الأداء الإنتاجي للدجاج البياض ومعدل التحويل الغذائى دون حدوث تأثيرات سلبية على جودة البيض.