RESPONSE OF LAYING HENS TO DIETS SUPPLEMENTED WITH MICROBIAL PHYTASE AND DIFFER IN THEIR PROTEIN AND METHIONINE CONTENT.

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ABSTRACT

A total number of 480 Bovans White laying hens 21 weeks of age were used to study the effect of dietary crude protein (CP), methionine (M) and microbial phytase (Phy) levels on laying hen performance, egg shell thickness, nutrient digestibility coefficients and economic efficiency. Hens were randomly divided into 8 equal groups with four replicates of 15 hens each. Two levels each of crude protein, methionine and microbial phytase were used in a 2X2X2 factorial design. The two levels of crude protein were the optimum level, 18% (CP_1) and low level, 16% (CP_2); methionine levels were the optimum level, 0.42% (M_1) and low level, 0.31% (M_2) and microbial phytase levels were 0.0 (Phy_1) and 500 FTU /Kg of the diet (Phy_2). The diet contained CP_1 and M_1 without microbial phytase supplementation represents the control. Hens were kept in cleaned and fumigated cages of wire floored batteries in an open system house under similar conditions of management up to 40 weeks of age. Water and feed were offered *ad-libitum* under a total of 16 hours light /day regimen.

The overall results showed that laying hens fed diets containing the optimum level of crude protein (18%) recorded significantly ($P<0.05$) higher egg production and egg weight and better feed conversion ratio compared to those fed low level of crude protein (16%). While, the average values of feed intake significantly ($P<0.05$) increased with feeding laying hens diets containing the lower level of crude protein. Feeding laying hens diets containing the optimum level of methionine (0.42%) significantly ($P<0.05$) increased egg production values comparing to those fed diets containing low level of methionine (0.31%). While, the average values of egg weight, feed intake and feed conversion ratio were not affected by using different levels of dietary methionine. At the same time, supplemental microbial phytase at 500 FTU/Kg of laying hen diets significantly ($P<0.05$) improved the average values of egg production, egg weight and feed conversion ratio. Laying hens fed diets either containing optimal levels of crude protein or supplemented with microbial phytase recorded significantly ($P<0.05$) higher values of egg shell thickness. Also, either the optimum level of dietary crude protein or microbial phytase addition to laying hens diets significantly ($P<0.05$) improved digestibility coefficient of organic matter, crude protein and ether extract. Economic efficiency values improved by feeding laying hens diets containing lower levels of either crude protein or methionine with supplemental microbial phytase. Accordingly, the results of this study indicated that adding microbial phytase can spare part of dietary crude protein or methionine in laying hen diets without adverse effects on laying hen performance, egg quality or economic efficiency.

Key words: Laying hens, performance, crude protein, methionine, microbial phytase, shell thickness)

INTRODUCTION

Feeding cost in poultry production is considered the most expensive item particularly dietary crude protein and amino acids, which are the most expensive components in poultry diets. Therefore, the use of low protein and amino acids diets for poultry has been the subject of numerous investigations **(Abd-Elsamee, 2002; Keshavarz and Austic, 2004; Ali, 2006; and Azazi *et al.*, 2006)** who reported that promising results can be obtained by the use of low protein and amino acids diets for various kinds of poultry. However, in a majority of these reports, productive performance still remained inferior to the control groups that were fed diets with conventional crude protein and amino acids levels. The results of a previous experiments conducted by **Keshavarz and Jackson (1992)** indicated that egg production and egg weight were not significantly different when hens were fed low protein diet as compared with the control group, which were fed recommended crude protein diet. On the other hand, poultry feedstuffs especially plant protein sources have a relatively high content of phosphorus. However, up to 80% of the phosphorus is presented as phytic acid which reduces the availability of minerals, protein, amino acids and digestive enzymes such as pepsin and trypsin **(Kornegay, 1996; Kornegay *et al.*, 1998;** and **Selle *et al.*, 2000)**. Thus, it is necessary to supplement phytase as feed additive to improve the utilization of nutrients which are bound with phytate molecule **(Ravindran, 1999)**. Some investigators reported that adding microbial phytase to poultry diets had positive impact on protein and amino acids availability of poultry diets and improved poultry performance **(Biehl and Baker, 1997; Sebastian *et al.*, 1997; Kies *et al.*, 2001** and **Adeola and Sands, 2003).**

The present study was carried out to determine the effect of different levels of dietary crude protein and methionine on laying hen performance. Seemingly, it aimed to determine the effect of adding microbial phytase, under such conditions, to laying hen diets on their performance, egg quality, nutrients digestibility and economic efficiency.

MATERIALS AND METHODS

The present work was carried out at the Poultry Nutrition Research Unit, Faculty of Agriculture, Cairo University. The analytical part of this study was performed at the Laboratories of Animal Production Department, Faculty of Agriculture, Cairo University. A total number of 480 Bovans White laying hens 21 weeks of age were used to study the effect of different levels of crude protein, methionine and microbial phytase on laying hen performance, egg quality, nutrients digestibility coefficients and economic efficiency of egg production. Birds were randomly distributed into 8 treatments, each containing 60 hens in 4 replicates of 15 hens each. Two levels of crude protein (CP), methionine (M) and microbial phytase (Phy) were used in a 2X2X2 factorial design. The tested crude protein levels were the optimum level, 18% (CP_1) and low level, 16% (CP_2). Methionine levels were the optimum level, 0.42% (M_1) and low level, 0.31% (M_2). The two levels of microbial phytase supplementation were 0.0 (Phy_1) and 500 FTU /Kg of the diet (Phy_2). Natuphos was used as a source of microbial phytase, each gram of Natuphos contain 2500 phytase units (FTU). Therefore, Natuphos was added to the tested diets at levels of 0.0 and 200 g/ton (500 FTU/Kg of the diet). The first diet containing 18% crude protein (CP_1) and 0.44% methionine (M_1) without microbial phytase supplementation (Phy_1) represents the control group. Composition of the experimental diets and their calculated analysis are presented in Table (1). In all experimental diets, metabolizable energy, minerals and vitamins were adjusted according to the

strain recommended catalog. Hens were allocated in cleaned and fumigated cages of wire floored batteries in an open system house under similar conditions of management. Water and feed in mash form were offered *ad-libtium* allover the experimental period (20 weeks, 5 periods of 4 weeks each) from 21 to 40 weeks of age, under a total of 16 hours light/day regimen.

Table (1). Composition and chemical analysis of the experimental diets

Ingredients	Diets								Price/ton LE ***
	1	2	3	4	5	6	7	8	
Yellow corn	55.00	55.00	55.00	55.00	61.51	61.50	61.50	61.50	900
Soybean meal (44%)	29.50	29.50	29.62	29.62	23.50	23.50	23.50	23.50	1700
Vegetable oil	3.50	3.50	3.50	3.50	2.50	2.50	2.50	2.50	3000
Bone meal	3.05	3.03	3.05	3.03	3.37	3.36	3.49	3.47	800
Lime stone	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	50
NaCl	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	100
Vit Min-mix*	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	7000
DL-Methionine	0.15	0.15	0.03	0.03	0.17	0.17	0.06	0.06	16000
L-Lysine HCl	0.00	0.00	0.00	0.00	0.15	0.15	0.15	0.15	16000
Natuphos	0.00	0.02	0.00	0.02	0.00	0.02	0.00	0.02	60000
Total	100	100	100	100	100	100	100	100	
Calculated analysis**									
CP	18.02	18.02	18.00	18.00	16.15	16.15	16.09	16.09	
ME K.Cal/kg	2808	2808	2811	2811	2804	2804	2804	2804	
CF	3.36	3.36	3.37	3.37	3.07	3.07	3.07	3.07	
EE	2.33	2.33	2.33	2.33	2.53	2.53	2.53	2.53	
Ca	4.04	4.03	4.04	4.03	4.12	4.11	4.15	4.15	
`Av. P	0.52	0.51	0.52	0.51	0.55	0.54	0.56	0.56	
Lysine	1.00	1.00	1.00	1.00	0.98	0.98	0.98	0.98	
Methionine	0.42	0.42	0.31	0.31	0.42	0.42	0.31	0.31	
Price/ton (LE)	1176	1186	1159	1169	1133	1143	1116	1125	

* Vitamin and mineral premix at 0.3 % of the diet supplies the following per Kg of the diet: Vit. A 10000 IU, Vit. D3 3000 IU, Vit. E 20 mg, Vit. K3 3 mg, Vit. B1 2 mg, Vit. B2 6 mg, Pantothenic acid 10 mg, Folic acid 1 mg, Biotin 5 mcg, Choline chloride 500 mg, Niacin 66 mg, Vit. BB6 5 mg, Vit. B12 20 mcg, Mn 100 mg, Fe 100 mg, Zn 75 mg, Cu 8 mg, I 45 mcg, Co 10 mcg and Se 10 mcg

** According to **NRC (1994).**

*** According to prices of the used ingredients at the experimental time

During the experimental period, daily egg production, feed intake and egg weight averages were calculated per hen every four weeks intervals. Records of egg production, feed intake and egg weight were used to calculate the values of feed conversion ratio (g feed consumed/g egg mass). Every four weeks, a total number of 40 eggs were taken from each treatment (10 eggs from each replicate) for testing egg quality as measured by shell thickness, using a digital dial pipe gauge. After the end of the feeding trial, four hens of each treatment (one from each replicate) were randomly chosen and individually housed in metabolic cages to determine the digestibility coefficient of nutrients. The analyses of feed and dried excreta were done according to the official methods **(A.O.A.C., 1990)**. Nitrogen–free extract (NFE) was calculated according to **Abou-Raya and Galal (1971)**. Fecal nitrogen was determined according to **Jacobson *et al.* (1960).** The data obtained were statistically examined by using **SAS (1994)** procedure.

Differences among treatment means were separated by Duncan's new multiple–range test **(Duncan, 1955)**. Finally, all dietary treatments were economically evaluated by calculating the net revenue per unit of total cost.

RESULTS AND DISCUSSION

Laying hen performance:

Egg production (%):

The effect of CP, M and Phy levels on egg production is presented in Table (2). Results showed that the average values of egg production significantly ($P<0.05$) decreased with feeding low dietary crude protein (16%) compared to those fed diets containing optimum level of crude protein (18%). The same trend was observed when laying hens were fed diets containing low level of methionine at 0.31% compared to those fed the optimum level of methionine at 0.42%. This means that performance of laying hen fed diets containing either 16% CP or 0.31% methionine was not as satisfactory as the performance of a group fed 18% CP or 0.42% methionine. These results are in agreement with those obtained by **Harms and Russell (1993); Keshavarz and Austic (2004)** and **Abd-Elsamee (2005)** who founed that the proper ratio between essential amino acids in low–protein diets may have been the reason for the inferior performance of hens fed diets containing low level of either crude protein or amino acids. However, **Keshavarz and Jackson (1992)** found that egg production was not significantly affected when laying hen were fed a sequence of 14-13-12% CP as compared with the control groups, which were fed a sequence of 18-16-15% CP. Regardless of dietary crude protein and methionine, data in Table (2) showed that the addition of microbial phytase at 500 FTU/Kg of the laying hen diet significantly ($P<0.05$) improved egg production values compared to those fed diet without microbial phytase supplementation. The improvement in egg production was mainly due to improving nutrients utilization due to adding microbial phytase to laying hen diets. Similar results were obtained by **Boling *et al.* (2000); Abd-Elsamee (2002)** and **Keshavarz and Austic (2004)** who found that adding phytase to laying hen diets improved egg production values. The interaction effect of CP, M and Phy on egg production is presented also in Table (2). Data showed that laying hens fed diet containing optimum levels of crude protein and methionine with supplemental microbial phytase (T2) recorded the highest value of egg production (92%). While, the lowest value (87.0%) was observed when laying hens were fed diets containing low levels of crude protein and methionine without supplemental microbial phytase (T7). Results indicated that there were no significant differences in egg production values between control group (T1) which recorded 90.6% and those fed diet containing low level of methionine and crude protein with supplemental microbial phytase (T8) which recorded 89.9%.

Table (2). Effect of treatments on egg production (%)

Treatments				Experimental period (weeks)					
No.	CP	M	Phy	21-24	25-28	29-32	33-36	37-40	Overall
Main effect of CP									
	CP1			84.9[a]	88.7[a]	92.2 [a]	94.1[a]	93.4[a]	90.7[a]

	CP2			83.5^{b}	86.5^{b}	90.3^{b}	92.6^{b}	91.1^{b}	88.8^{b}
Main effect of Meth.									
		M1		84.7^{a}	88.2^{a}	91.7^{a}	94.0^{a}	92.8^{a}	90.3^{a}
		M2		83.8^{b}	87.1^{a}	90.9^{b}	92.9^{b}	91.6^{b}	89.3^{b}
Main effect of Phytase									
			Phy1	83.3^{b}	86.7^{b}	90.2^{b}	92.5^{b}	91.2^{b}	88.8^{b}
			Phy2	85.2^{a}	88.5^{a}	92.3^{a}	94.4^{a}	93.2^{a}	90.7^{a}
1	**CP1**	**M1**	**Phy1**	84.9^{bc}	88.5^{bc}	92.1^{bc}	94.2^{bc}	93.4^{b}	90.6^{bc}
2	**CP1**	**M1**	**Phy2**	86.2^{a}	90.1^{a}	93.4^{a}	95.3^{a}	95.0^{a}	92.0^{a}
3	**CP1**	**M2**	**Phy1**	83.5^{cd}	87.2^{c}	90.9^{c}	92.5^{c}	91.3^{c}	89.1^{bc}
4	**CP1**	**M2**	**Phy2**	85.3^{ab}	89.1^{ab}	92.5^{ab}	94.4^{ab}	93.8^{ab}	91.1^{ab}
5	**CP2**	**M1**	**Phy1**	83.2^{c}	86.2^{d}	89.8^{d}	92.1^{cd}	90.9^{cd}	88.4^{cd}
6	**CP2**	**M1**	**Phy2**	84.6^{cd}	87.8^{cd}	91.5^{cd}	94.1^{bc}	92.1^{c}	90.0^{bc}
7	**CP2**	**M2**	**Phy1**	81.7^{e}	84.9^{e}	88.2^{e}	91.2^{d}	89.2^{d}	87.0^{d}
8	**CP2**	**M2**	**Phy2**	84.6^{cd}	87.2^{d}	91.9^{c}	93.8^{c}	92.2^{c}	89.9^{bc}

a , b , c … Means in each column, within each item, bearing the same superscripts are not significantly different (P<0.05).

Egg weight (g):

The effect of experimental treatments on egg weight (g) is listed in Table (3). Results showed that feeding laying hens the diets containing optimum level of crude protein (CP_1) significantly ($P \leq 0.05$) improved egg weight values compared to those fed diets containing low level of crude protein (CP_2). The same trend was observed with using microbial phytase (Phy_2) comparing to without microbial phytase supplementation (Phy_1). While, there was no significant effect due to using different levels of methionine on egg weight values. The positive effect of dietary crude protein on egg weight may be explained by improving the formation of interior egg content such as yolk and albumen with using optimum level of crude protein as compared with low level of crude protein. In this connection, **Keshavarz and Austic (2004)** found that egg weight values improved when they fed laying hen diets containing 16% CP compared to those fed diets containing 13% CP. Also, **Um and Paik (1999)** reported that egg weight significantly increased with supplemental phytase to laying hen diets. In contrast, **Abd-Elsamee (2005)** noticed that egg weight values significantly increased with increasing dietary methionine level.

The interaction effect of CP, M and Phy on egg production is presented in Table (3). Data showed that laying hens fed diet containing optimum levels of crude protein and methionine with supplemental microbial phytase (T2) recorded the highest value of egg weight (59.1g). While, the lowest value (57.4g) was observed when laying hens were fed diets containing low levels of crude protein without supplemental microbial phytase (T5 and T7). Results indicated that there were no significant differences in egg weight values between control group (T1) and those fed diet containing low level of methionine and crude protein with supplemental microbial phytase (T8), being 58.6 and 58.5g, respectively.

Table (3). Effect of treatments on egg weight (g)

Treatments				**Experimental period (weeks)**					
No.	**CP**	**M**	**Phy**	**21-24**	**25-28**	**29-32**	**33-36**	**37-40**	**Overall**
Main effect of CP									
	CP1			52.4^{a}	56.2^{a}	60.2^{a}	61.7^{a}	62.8^{a}	58.7^{a}

	CP2			51.5^{b}	55.5^{b}	59.6^{b}	60.9^{b}	62.1^{b}	57.9^{b}
Main effect of Meth.									
		M1		52.1^{a}	55.9^{a}	59.9^{a}	61.5^{a}	62.5^{a}	58.4^{a}
		M2		51.8^{a}	55.9^{a}	59.9^{a}	61.2^{a}	62.4^{a}	58.2^{a}
Main effect of Phytase									
			Phy1	51.7^{b}	55.5^{b}	59.5^{b}	60.9^{b}	62.1^{b}	57.9^{b}
			Phy2	52.2^{a}	56.2^{a}	60.3^{a}	61.7^{a}	62.8^{a}	58.7^{a}
1	**CP1**	**M1**	**Phy1**	52.2^{a}	56.1^{a}	60.1^{a}	61.9^{ab}	62.7^{a}	58.6^{a}
2	**CP1**	**M1**	**Phy2**	52.9^{a}	56.5^{a}	60.6^{a}	62.4^{a}	63.1^{a}	59.1^{a}
3	**CP1**	**M2**	**Phy1**	52.1^{ab}	56.1^{a}	60.1^{a}	61.2^{b}	62.5^{a}	58.4^{a}
4	**CP1**	**M2**	**Phy2**	52.4^{a}	56.2^{a}	60.2^{a}	61.5^{b}	62.8^{a}	58.6^{a}
5	**CP2**	**M1**	**Phy1**	51.4^{cb}	54.9^{b}	58.9^{b}	60.2^{c}	61.5^{b}	57.4^{b}
6	**CP2**	**M1**	**Phy2**	51.8^{cb}	56.1^{a}	60.2^{a}	61.5^{b}	62.7^{a}	58.5^{a}
7	**CP2**	**M2**	**Phy1**	51.1^{c}	55.1^{b}	59.1^{b}	60.3^{c}	61.6^{b}	57.4^{b}
8	**CP2**	**M2**	**Phy2**	51.7^{cb}	56.1^{a}	60.1^{a}	61.6^{b}	62.8^{a}	58.5^{a}

See footnote in Table 2.

Feed intake (g):

The effect of experimental treatments on feed intake (g/hen/day) is shown in Table (4). Results showed that feed intake values significantly ($P \leq 0.05$) increased when the laying hens were fed diets containing lower level of crude protein (CP_2) compared to those fed diets containing optimum level of crude protein (CP_1). This may be explained on the basis that hens tend to consume more feed to cover the requirement of crude protein, which have a favorable effect on laying hen performance. Data indicated that using either optimum level of methionine (M_1) or microbial phytase (Phy_2) slightly increased feed intake values but without significant differences as compared to those fed diets containing lower level of methionine (M_2) or without microbial phytase supplementation (Phy_1). Similar results were reported by **Abd-Elsamee (2005)** who found that the average values of feed intake were not significantly different due to various dietary methionine levels. Also, **Van Der Klis *et al.* (1997)** noticed no significant differences in feed intake values when laying hens were fed diets supplemented with phytase. In contrast, **Gordon and Roland (1997)** and **Abd-Elsamee (2002)** found that supplemental phytase to laying hen diets significantly increased feed intake.

The interaction effect of CP, M and Phy on feed intake is presented also in Table (4). Data showed that laying hens fed diet containing lower level of crude protein and the two levels of methionine with supplemental microbial phytase (T6 and T8) recorded the highest value of feed intake (120.8 and 120.6g, respectively). While, the lowest values (116.1 and 116.4g) were observed when laying hens were fed diet containing the optimum level of crude protein and each of methionine levels without supplemental microbial phytase (T3 and T1, respectively).

Table (4). Effect of treatments on feed intake (g/hen/day)

Treatments				**Experimental period (weeks)**					
No.	**CP**	**M**	**Phy**	**21-24**	**25-28**	**29-32**	**33-36**	**37-40**	**Overall**
Main effect of CP									
	CP1			110.0^{b}	114.7^{b}	118.4^{b}	119.9^{b}	121.2^{b}	116.8^{b}

	CP2			113.5[a]	118.0[a]	121.7[a]	123.2[a]	124.4[a]	120.2[a]
Main effect of Meth.									
		M1		111.9[a]	116.4[a]	120.1[a]	121.6[a]	122.9[a]	118.6[a]
		M2		111.6[a]	116.3[a]	120.0[a]	121.5[a]	122.7[a]	118.4[a]
Main effect of Phytase									
			Phy1	111.4[a]	115.8[a]	119.5[a]	120.9[a]	122.1[a]	117.9[a]
			Phy2	112.1[a]	116.9[a]	120.6[a]	122.2[a]	123.5[a]	119.1[a]
1	**CP1**	**M1**	**Phy1**	109.8[c]	114.2[c]	117.9[c]	119.4[c]	120.5[c]	116.4[c]
2	**CP1**	**M1**	**Phy2**	110.5[bc]	115.4[bc]	119.1[bc]	120.6[bc]	122.1[bc]	117.5[bc]
3	**CP1**	**M2**	**Phy1**	109.5[c]	114.0[c]	117.7[c]	119.2[c]	120.3[c]	116.1[c]
4	**CP1**	**M2**	**Phy2**	110.2[bc]	115.3[bc]	119.0[bc]	120.5[bc]	122.0[bc]	117.4[bc]
5	**CP2**	**M1**	**Phy1**	113.3[a]	117.5[ab]	121.2[a]	122.7[ab]	123.9[ab]	119.7[ab]
6	**CP2**	**M1**	**Phy2**	114.0[a]	118.7[a]	122.3[a]	123.8[a]	125.1[a]	120.8[a]
7	**CP2**	**M2**	**Phy1**	113.0[ab]	117.4[ab]	121.1[a]	122.6[ab]	123.8[ab]	119.6[ab]
8	**CP2**	**M2**	**Phy2**	113.7[a]	118.5[a]	122.2[a]	123.8[a]	124.9[a]	120.6[a]

See footnote in Table 2.

Feed conversion ratio (FCR):

The effect of dietary crude protein, methionine, microbial phytase and their interaction on feed conversion ratio is listed in Table (5). Data showed that laying hens fed diets containing optimum level of dietary crude protein (CP_1) had significantly ($P<0.05$) better feed conversion ratio compared to those fed diets containing low level of crude protein (CP_2). Also, supplementing laying hen diets with microbial phytase (Phy_2) significantly ($P<0.05$) improved feed conversion ratio as compared with those fed diets without microbial phytase (Phy_1) supplementation. While, when laying hens were fed diets containing optimum level of methionine (M_1), the average values of feed conversion ratio slightly improved was without significant differences compared to those fed diets containing lower level of methionine (M_2). Such improvement perhaps is due to increasing both egg production and egg weight with using optimum level of crude protein and methionine or supplemental microbial phytase to laying hen diets. Similar results were obtained by **Keshavarz and Austic (2004)** who found that feed conversion ratio was significantly improved when laying hens were given diets containing conventional crude protein level compared to those fed diets containing low crude protein level. Also, **Abd-Elsamee (2005)** reported that feed conversion ratio was improved when laying hens were fed diets containing high levels of methionine. Similarly, **Abd-Elsamee (2002)** showed that supplemental microbial phytase to laying hen diets improved feed conversion ratio. The interaction effect of CP, M and Phy on feed conversion is presented also in Table (5). Data showed that laying hens fed diet containing optimum levels of crude protein and methionine with supplemental microbial phytase (T2) recorded better value of feed conversion (2.17). While, the worst feed conversion (2.40) was observed when laying hens were fed diets containing low levels of crude protein and methionine without supplemental microbial phytase (T7).

Table (5). Effect of treatments on feed conversion ratio (FCR)

Treatments				**Experimental periods**					
No.	**CP**	**M**	**Phy**	**21-24**	**25-28**	**29-32**	**33-36**	**37-40**	**Overall**
Main effect of CP									
	CP1			2.47[b]	2.29[b]	2.13[b]	2.06[b]	2.07[b]	2.20[b]

	CP2			2.64[a]	2.45[a]	2.26[a]	2.18[a]	2.20[a]	2.35[a]
Main effect of Meth.									
		M1		2.54[a]	2.36[a]	2.18[a]	2.11[a]	2.12[a]	2.26[a]
		M2		2.57[a]	2.39[a]	2.20[a]	2.14[a]	2.15[a]	2.29[a]
Main effect of Phytase									
			Phy1	2.59[a]	2.40[a]	2.22[a]	2.15[a]	2.16[a]	2.30[a]
			Phy2	2.52[b]	2.35[b]	2.17[b]	2.10[b]	2.11[b]	2.25[b]
1	**CP1**	**M1**	**Phy1**	2.48[de]	2.30[cd]	2.13[c]	2.05[cd]	2.06[de]	2.20[de]
2	**CP1**	**M1**	**Phy2**	2.42[f]	2.27[d]	2.10[c]	2.03[d]	2.04[e]	2.17[e]
3	**CP1**	**M2**	**Phy1**	2.52[d]	2.33[c]	2.15[c]	2.11[bc]	2.11[cd]	2.24[cd]
4	**CP1**	**M2**	**Phy2**	2.46[e]	2.30[cd]	2.14[c]	2.07[cd]	2.07[de]	2.21[de]
5	**CP2**	**M1**	**Phy1**	2.65[ab]	2.48[a]	2.29[a]	2.22[a]	2.22[ab]	2.37[a]
6	**CP2**	**M1**	**Phy2**	2.60[b]	2.41[b]	2.22[b]	2.14[b]	2.17[bc]	2.31[b]
7	**CP2**	**M2**	**Phy1**	2.71[a]	2.51[a]	2.32[a]	2.23[a]	2.25[a]	2.40[a]
8	**CP2**	**M2**	**Phy2**	2.59[c]	2.42[b]	2.21[b]	2.14[b]	2.16[bc]	2.30[bc]

See footnote in Table 2.

Egg shell thickness:

The effect of treatments on egg shell thickness (mm) as an indicator of egg quality is presented in Table (6). Results showed that feeding laying hens either the optimal level of crude protein (CP_1) or supplemental microbial phytase (Phy_2) significantly ($P<0.05$) increased shell thickness values. While, there were no significant differences in average values of shell thickness due to different levels of dietary methionine. Data showed that the highest value of shell thickness (0.377mm) was recorded when laying hens were fed diet containing optimal level of both crude protein and methionine with supplemental microbial phytase (T2). While, the lowest value (0.363mm) was observed by feeding laying hens the diet containing lower level of crude protein and methionine without supplemental microbial phytase (T7). These results may be attributed to the influence on utilization of minerals due to supplementing microbial phytase to laying hen diets. Results of shell thickness obtained herein confirmed those previously found by **Abd-Elsamee (2002)** who reported that eggshell thickness was significantly improved with adding microbial phytase to laying hen diets.

Digestibility coefficient:

The effect of treatments on nutrient digestibility coefficients are summarized in Table (7). Results indicated that using the optimum level of crude protein (CP_1) and supplemental microbial phytase (Phy_2) in laying hen diets significantly ($P<0.05$) improved digestibility coefficient of both organic matter (OM), crude protein (CP) and ether extract (EE). While, digestibility coefficients of either crude fiber (CF) or nitrogen free extract (NFE) were not significantly affected. Also, data in Table (7) showed that there was a slight improvement but without significant effects in favor of the optimum dietary methionine level (M_1). The improvement in nutrient digestibility coefficients due to using optimum level of crude protein along with optimum level of methionine (T2) may be due to the associative effect of diet nutrients. While, the improvement due to adding microbial phytase perhaps is due to the improvement in nutrients absorption especially crude protein which complicate with phytate and its inhibitory effects on proteolytic enzymes such as pepsin and trypsin .In this regard, **Ravindran (1999); Zhang *et al.* (1999);** and **Attia *et al.* (2001)** indicated that phytase

addition improved protein and amino acids utilization of poultry diets by 4-6 % depending on feedstuffs used.

Table (6). Effect of treatments on egg shell thickness (mm)

Treatments				Experimental period (weeks)					
No.	**CP**	**M**	**Phy**	**21-24**	**25-28**	**29-32**	**33-36**	**37-40**	**Overall**
Main effect of CP									
	CP1			0.389[a]	0.381[a]	0.370[a]	0.365[a]	0.355[a]	0.372[a]
	CP2			0.386[b]	0.377[b]	0.366[b]	0.358[b]	0.350[b]	0.367[b]
Main effect of Meth.									
		M1		0.387[a]	0.379[a]	0.368[a]	0.362[a]	0.354[a]	0.370[a]
		M2		0.387[a]	0.378[a]	0.368[a]	0.361[a]	0.351[a]	0.369[a]
Main effect of Phytase									
			Phy1	0.385[b]	0.374[b]	0.364[b]	0.357[b]	0.349[b]	0.366[b]
			Phy2	0.389[a]	0.383[a]	0.372[a]	0.365[a]	0.355[a]	0.373[a]
1	**CP1**	**M1**	**Phy1**	0.386[a]	0.377[ab]	0.366[ab]	0.364[a]	0.353[ab]	0.369[ab]
2	**CP1**	**M1**	**Phy2**	0.392[a]	0.387[a]	0.375[a]	0.368[a]	0.361[a]	0.377[a]
3	**CP1**	**M2**	**Phy1**	0.385[a]	0.374[bc]	0.365[ab]	0.363[a]	0.352[ab]	0.368[ab]
4	**CP1**	**M2**	**Phy2**	0.391[a]	0.384[ab]	0.374[a]	0.365[a]	0.355[ab]	0.374[ab]
5	**CP2**	**M1**	**Phy1**	0.385[a]	0.375[bc]	0.363[b]	0.352[b]	0.347[b]	0.366[bc]
6	**CP2**	**M1**	**Phy2**	0.387[a]	0.379[a]	0.368[ab]	0.365[a]	0.354[ab]	0.370[abc]
7	**CP2**	**M2**	**Phy1**	0.384[a]	0.371[c]	0.362[b]	0.351[b]	0.346[b]	0.363[c]
8	**CP2**	**M2**	**Phy2**	0.389[a]	0.382[a]	0.371[ab]	0.363[a]	0.352[ab]	0.370[abc]

See footnote in Table 2.

Economic efficiency:

Economic efficiency of different formulated diets and money return per hen at the end of experimental period are shown in Table (8). Egg production (egg no./hen) and feeding cost are generally among the most important factors involved in the achievement of maximum efficiency of egg production. The economic efficiency values were calculated according to the prevailing market (selling) price of egg, which was 0.3 LE on average at the experimental time. Results indicated that the average values of net revenue and economic efficiency decreased when laying hens were fed diets containing low levels of both crude protein (CP_2) and methionine (M_2) without supplemental microbial phytase (T7). However, it is clear that supplemental microbial phytase to such diets (containing lower levels of both CP and M) improved both net revenue and economic efficiency values (T8). This improvement in economic efficiency could be attributed to improvement in both egg production and feed conversion ratio with adding microbial phytase to laying hen diets. These results are in agreement with those obtained by **Abd-Elsamee (2002)** who indicated that adding microbial phytase to laying hen diets improved economic efficiency values.

On the basis of the results of this study, it may be concluded that laying hens can be fed diets containing lower levels of both crude protein and methionine with supplemental 500 FTU phytase /Kg of the diet without adverse effects on laying hen performance, egg shell thickness and economic efficiency

Table (7). Effect of treatments on nutrient digestibility coefficients (%)

Treatments				Digestibility coefficients (%)				
No.	**CP**	**M**	**Phy**	**OM**	**CP**	**EE**	**CF**	**NFE**
Main effect of CP								

	CP1			82.7[a]	88.0[a]	78.2[a]	26.8[a]	85.5[a]
	CP2			81.7[b]	87.0[b]	77.0[b]	26.7[a]	85.4[a]
Main effect of Meth.								
		M1		82.4[a]	87.7[a]	77.8[a]	26.9[a]	85.6[a]
		M2		82.0[a]	87.3[a]	77.5[a]	26.7[a]	85.4[a]
Main effect of Phytase								
			Phy1	81.9[b]	86.9[b]	77.2[b]	26.9[a]	85.3[a]
			Phy2	82.5[a]	88.1[a]	78.1[a]	26.7[a]	85.6[a]
1	**CP1**	**M1**	**Phy1**	82.6[a]	87.6[ab]	77.9[ab]	26.5[a]	85.5[a]
2	**CP1**	**M1**	**Phy2**	83.2[a]	88.8[a]	78.8[a]	27.3[a]	85.8[a]
3	**CP1**	**M2**	**Phy1**	82.4[ab]	87.3[ab]	77.5[bc]	26.3[a]	85.3[a]
4	**CP1**	**M2**	**Phy2**	82.7[a]	88.4[ab]	78.7[a]	27.1[a]	85.4[a]
5	**CP2**	**M1**	**Phy1**	81.5[bc]	86.7[bc]	76.8[c]	26.9[a]	85.3[a]
6	**CP2**	**M1**	**Phy2**	82.2[ab]	87.9[ab]	77.6[bc]	27.2[a]	85.7[a]
7	**CP2**	**M2**	**Phy1**	81.1[c]	86.1[c]	76.5[c]	26.4[a]	85.2[a]
8	**CP2**	**M2**	**Phy2**	81.9[bc]	87.4[bc]	77.2[bc]	26.8[a]	85.6[a]

See footnote in Table 2.

Table (8). Effect of treatments on economic efficiency

Treatments				**Fixed Cost* (LE)**	**FI/ Hen (Kg)**	**Price/ Kg feed (LE)**	**Cost of FI (LE)**	**Total cost (LE)**	**EP No./ hen**	**Price of egg (LE)**	**Net revenue**	**EEF****	**Relative EEF *****
No.	**CP**	**M**	**Phy**										
Main effect of CP													
	CP1			1.5	16.3	1.17	19.1	20.6	126	37.8	17.2	0.83	
	CP2			1.5	16.8	1.13	18.9	20.4	124	37.2	16.8	0.82	
Main effect of Meth.													
		M1		1.5	16.6	1.16	19.2	20.7	126	37.8	17.1	0.83	
		M2		1.5	16.5	1.14	18.8	20.3	124	37.2	16.9	0.82	
Main effect of Phytase													
			Phy1	1.5	16.5	1.15	18.9	20.4	124	37.2	16.8	0.82	
			Phy2	1.5	16.6	1.15	19.1	20.6	126	37.8	17.2	0.83	
1	**CP1**	**M1**	**Phy1**	1.5	16.3	1.18	19.2	20.7	126	37.8	17.1	0.82	**100**
2	**CP1**	**M1**	**Phy2**	1.5	16.4	1.19	19.5	21.0	129	38.7	17.7	0.84	**102**
3	**CP1**	**M2**	**Phy1**	1.5	16.2	1.16	18.8	20.3	124	37.2	16.9	0.83	**101**
4	**CP1**	**M2**	**Phy2**	1.5	16.4	1.17	19.2	20.7	127	38.1	17.4	0.84	**102**
5	**CP2**	**M1**	**Phy1**	1.5	16.8	1.13	18.9	20.4	123	36.9	16.5	0.81	**98**
6	**CP2**	**M1**	**Phy2**	1.5	16.9	1.14	19.3	20.8	126	37.8	17.0	0.82	**100**
7	**CP2**	**M2**	**Phy1**	1.5	16.7	1.12	18.7	20.2	121	36.3	16.1	0.79	**96**
8	**CP2**	**M2**	**Phy2**	1.5	16.9	1.13	19.0	20.5	125	37.5	17.0	0.83	**101**

* Rearing cost per hen. ** Net revenue per unit cost. *** Relative to control

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استجابة الدجاج البياض للعلائق المضاف إليها انزيم الفيتيز الميكروبى وتختلف فى محتواها من البروتين والمثيونين

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أجريت هذه الدراسة على عدد ٤٨٠ دجاجة بياضة عمر ٢١ أسبوع من نوع البوفانز الأبيض قسمت الطيور إلى ٨ مجاميع متساوية العدد (قسمت كل مجموعة إلى أربع مكررات بكل منها ١٥ طائر) وذلك لدراسة تأثير المستويات المختلفة من بروتين العليقة والمثيونين وأنزيم الفيتيز الميكروبى على الأداء الإنتاجي للدجاج البياض وسمك قشرة البيضة ومعاملات هضم المركبات الغذائية والكفاءة الاقتصادية لإنتاج البيض. تم استخدام مستويين من البروتين الخام ومستويين من المثيونين ومستويين من إنزيم الفيتيز الميكروبي وذلك فى تصميم عشوائي متداخل ٢x٢x٢. كان مستوى بروتين العليقة عبارة عن المستوى الأمثل (١٨%) والمستوى المنخفض (١٦%) وكان مستوى مثيونين العليقة عبارة عن المستوى الأمثل (٠.٤٢%) والمستوى المنخفض (٠.٣١ %) أما مستوى إنزيم الفيتيز الميكروبي فكان صفر، ٥٠٠ وحدة فيتيز/كجم من العليقة أخذا" فى الاعتبار مجموعة المقارنة التى تغذت على العليقة المحتوية على المستوى الأمثل من كل من البروتين الخام والمثيونين بدون إضافة إنزيم الفيتيز الميكروبي .تم تربية الطيور تحت نفس الظروف من الرعاية والمعاملات البيطرية طوال فترة التجربة حتى الأسبوع ٤٠ من العمر.

أوضحت نتائج هذه الدراسة أن تغذية الدجاج البياض على علائق تحتوى على المستوى الأمثل من البروتين (١٨%) أدى إلى زيادة معنوية لكل من إنتاج البيض ووزن البيضة كما تحسن معنويا الكفاءة التحويلية للغذاء مقارنة بمجموعة الطيور المغذاة على المستوى المنخفض من البروتين (١٦%). بينما أدى تغذية الدجاج البياض على علائق تحتوى على المستوي المنخفض من البروتين إلى زيادة معنوية فى قيم الغذاء المأكول. أدى استخدام المستوى الأمثل من المثيونين (٠.٤٢%) إلى زيادة معنوية فى قيم إنتاج البيض مقارنة بمجموعة الطيور المغذاة على علائق تحتوى على المستوى المنخفض من المثيونين (٠.٣١%). بينما لم يكن هناك تأثير معنوي فى قيم كل من وزن البيضة والغذاء المأكول وكفاءة تحويل الغذاء عند تغذية الدجاج البياض على علائق مختلفة فى محتواها من المثيونين. فى نفس الوقت أدى إضافة ٥٠٠ وحدة إنزيم فيتيز/كجم من العليقة إلى تحسن معنوي فى قيم كل من إنتاج البيض ووزن البيضة وكفاءة تحويل الغذاء. عند تغذية الدجاج البياض على علائق تحتوى إما على المستوى الأمثل من البروتين أو إضافة إنزيم الفيتيز الميكروبي أدى هذا إلى تحسن معنوي فى قيم سمك القشرة كما أدى أيضا إلى تحسن معنوي فى قيم معامل هضم كل من المادة العضوية والبروتين الخام والدهن الخام . وبوجه عام كان هناك تأثير واضح لإضافة إنزيم الفيتيز الميكروبي إلى علائق الدجاج البياض المحتوية على مستوى منخفض سواء من البروتين الخام أو المثيونين على التقييم الاقتصادي للمعاملات الغذائية حيث أدى إلى تحسن قيم الكفاءة الإقتصادية. من هذه الدراسة يتضح إمكانية تغذية الدجاج البياض على علائق منخفضة فى محتواها من البروتين الخام والمثيونين مع إضافة إنزيم الفيتيز الميكروبي دون التأثير السلبى على الأداء الإنتاجي أو صفات قشرة البيضة أو الكفاءة الاقتصادية لإنتاج البيض.