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Effect of Enzyme Supplementation to Laying Hens Diets Containing Different Levels of Sunflower Seed Meal on Performance and Egg Quality

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ABSRACT

An experiment was conducted to determine the effect of enzyme supplementation to laying hens diets containing different levels of sunflower seed meal on performance and egg quality. The experiment was used 128 laying hens (Hy-Line W36) at 44 week of age throughout 84 days. Sixteen diets consisting of four levels of sunflower seed meal (0, 5, 10 and 20 % diet) and four levels enzyme (0, 500, 1000 and 2000 mg/ kg diet) in 4 x 4 factorial arrangement were used with four replicates of two birds each. During the experiment, feed and water were used as ad libitum. The effect of enzyme supplementation to laying hens diets containing different levels of sunflower seed meal did not significantly effect on egg production, egg weight, egg mass, feed intake, feed conversion ratio, egg shell weight, egg shell thickness and egg shell breaking strength (P>0.05). The result of this study that containing 20% sunflower seed meal to laying hens diets can be used without adversely affecting performance and egg quality.

1. Introduction

Soybean meal is the major protein source for the world's industrial poultry production. Other oil seed sources such as cottonseed, rapeseed and sunflower meals (SFM) are frequently used as alternatives (Swick, 1996). Sunflower contains about 45% of oil and a valuable amount of protein and B-complex vitamins. However, the use of SFM in poultry diet is limited by variations in its chemical composition and the two main components restricting its use are high fiber and low lysine contents (Rezaei & Hafezian, 2007; De Morais Oliveira et al., 2016). Fiber of SFM has been shown to mainly consist of non-starch polysaccharides (NSP) with cellulose, xylose, pectin and lignin contents (Carre & Brillouet, 1986). Sunflower meal is high fiber content, of 45.19 neutral detergent fiber (NDF) and 21.35 acid detergent fiber (ADF) limits its inclusion of the meal in diets for those birds (NRC 1994). Most of these compounds are not available for poultry since they lack the related enzymes in their gastrointestinal tract.

Most of the studies evaluated the use of sunflower in poultry feeding and reported that a high level of SFM can be used in laying hens' diet without any negative effect on egg performance and quality (Tsuzuki et al., 2003; Casartelli et al., 2006; Laudadio et al., 2014). However, levels higher than 5% in diet require supplementation of lysine. Amino acid composition of SFM is also variable, with lysine and methionine levels ranging from 0.56 to 0.66% and from 0.33 to 0.50%, respectively. The SFM is a good protein source and its content depends by dehulling, air-classification, and oil extraction processes (Laudadio et al., 2013). McNaughton & Deaton (1981) and Roth-Maier (2000) reported that SFM could be included up to 30% in layer diets, thereby totally replacing soybean meal, without adversely affecting body weight, egg production or egg weight. However, feed intake increased as the dietary inclusion rate of SFM increased. A similar result was obtained by Vieira et al. (1992) when high-fibre SFM (27% fiber) was included from 14% to 41% in non-isocaloric layer diets. Şenköylü & Dale (1999) reviewed that SFM could successfully be included in layer, broiler and water-fowl diets to replace 50-100 % of soybean meal, depending on the type of diet and the nature of the other ingredients. Substituting soybean meal with SFM could reduce feeding cost to layers quite significantly. Serman et al. (1997) verified that inclusion of sunflower meal in layer diets decreased feed intake, egg mass and weight gain. However, in that study, the diets did not contain synthetic amino acids, which may explain those results. Vieira et al. (1992) replaced all the soybean meal by sunflower meal (40.5%) and added lysine in layers diets without compromising production parameters. However, feed

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conversion ratio (FCR) and feed intake showed positive linear behavior as a function of the inclusion of sunflower meal in the diets (Vieira et al., 1992). Casartelli et al. (2006) reported that inclusion of up to 12.0% sunflower meal in layer feeds did not affect productive parameters or egg internal quality parameters. Additionally, the inclusion of sunflower meal in diets improved eggshell thickness. Studies found that it was possible to include up to 5.6% ground whole sunflower seeds in layer diets without affecting performance or egg quality (Tsuzuki et al., 2003). Working with sunflower meal under partial oil extraction, Senkoylu et al. (2004) included up to 20.0% in layer diets and did not observe any differences in egg mass or feed intake values.

The utilization of exogenous enzymes could enhance fiber digestibility or phytic phosphorus solubilization in sunflower meal, thereby reducing the negative effects on layer performance. (Tsuzuki et al., 2003). Exogenous enzymes have been used in the poultry industry to reduce the anti-nutritional factors effects and to improve the utilization of dietary energy and protein, thus leading to enhanced poultry production (Sateri et al., 2017). It is, however, understood that the responses to exogenous enzymes in poultry species with respect to performance has been variable and depends on many factors, such as bird age and quality or type of diet used (Tufarelli et al., 2007; Lee et al., 2014; Sateri et al., 2017). Since SFM contains substantial concentrations of cell-wall material and a high fiber level that could affect the nutritive value of meal, the use of exogenous enzyme may be justified to improve the accessibility of cell contents to digestive enzymes (Alagawany et al., 2015). Khan et al. (2006) reported that supplementation of SFM-based diets including a multi enzyme improved feed conversion ratio and egg weight. Therefore, the use of multi-enzymes in diet may increase digestibility of the protein improving laying hens performance. Recently, Junqueira et al. (2010), evaluating the economic viability of the inclusion of enzyme phytase and sunflower meal in layer diets, concluded that the addition of phytase reduced phosphorus excretion and improved the economic parameters. However, the maximum level of sunflower meal inclusion that did not negatively affect productive parameters was 4%.

The objective of this study was to determine the effects of multi enzyme supplementation to dietary substitution of soybean meal by different levels of SFM on performance and eggshell quality traits of laying hens.

2. Materials and Methods

One hundred and twenty-eight commercial layers (Hy-Line W36) were housed in a layer house equipped with 72 metal battery cages (50 x 50 x 40 cm, W x L x L, respectively). A completely randomized design was used, with four replications of eight birds each, two hens per cage. The experiment started when the hens were 44 weeks of age, and consisted of three periods of 84 days. Light was provided for 16 hours per day during the experimental period. Feed and water were offered ad libitum throughout the experiment (44-56 week of age). Housed in individual layer cages were environmentally controlled room (23-25 ^{o}C).

The treatments were distributed using a factorial arrangement (4x4), which consisted of the combination of four sunflower meal (0, 5, 10, 20 %) and four enzyme inclusion levels (0, 500, 1000 and 2000 IU/kg). Soybean meal and corn of the total diet was replaced with sunflower meal. The experimental diets (total 16 diets; Table 1) were formulated to be isolocaloric and isonitrogenous and met the NRC nutrient recommendations (NRC, 1994). The enzyme (Farmazyme 3000 PROENX®) supplement contained fungal xylanase (1,800,000 IU/kg), β-glucanase (300,000 IU/kg), cellulase (4,000,000 IU/kg), pectinase (200,000 IU/kg), protease (1,200,000 IU/kg), α-amylase (200,000 IU/kg) and it was added into the experimental diets.

Body Weight (BW) was obtained by weighting hens at the beginning and end of the experiment. Feed Intake (FI) and Egg Weight (EW) were recorded biweekly. Egg Production (EP) was recorded daily and Egg Mass (EM) was calculated from collecting data of EP and EW at biweekly via: EM= (EP x EW) / Period (days). Feed conversion ratio (FCR; g of feed g of egg) was calculated via: FCR = FI (g of feed/hen/period) / EM (g of egg/hen/period).

The eggs were subjected to determine characteristics of eggshell quality parameters (shell breaking strength, shell weight and shell thickness) on all collected eggs produced at the last two days of each period during the experiment. Eggshell breaking strength was measured using a cantilever system by applying increased pressure to the broad pole of the shell using an instrument (06-UM-001, Version B; Orka Food Tech. Ltd, Hong Kong, China). Eggs were then broken, and eggshell, albumen, and yolk were separated and weighed. Eggshells were rinsed running water and dried in oven at 60°C for 12 h, to determine eggshell thickness (including the membrane) in three points on the eggs (one point on air cell or the randomized two points of equator) using a micrometer (Mitutoyo, 0.01 mm, Japan). Eggshells were weighed using a 0.001g precision scale. Eggshell ratio was calculated via: Eggshell weight (g/100 g egg) = [Eggshell weight (g) / Egg weight (g)].

Data were subjected to ANOVA by using Minitab (2000). Duncan's multiple range tests were applied to separate means (MstatC, 1980). Statements of statistical significance are based on probability of $P<0.05$.

Table 1
Composition of experimental diets

	Sunflower meal levels of diets (%)			
Ingredients (%)	**0**	**5**	**10**	**20**
Corn	55.38	53.00	50.70	45.84
Barley	6.00	6.00	6.00	6.00
Wheat	4.00	4.00	4.00	4.00
Soybean meal	19.50	16.50	13.33	7.17
Sunflower meal	0.00	5.00	10.00	20.00
Limestone	9.77	9.75	9.76	9.74
Vegetable oil	2.76	3.20	3.70	4.74
Salt	0.35	0.35	0.35	0.35
Di-Calcium phosphate	1.88	1.84	1.80	1.76
Vitamin Premix[1]	0.16	0.16	0.16	0.16
Mineral Premix[2]	0.08	0.08	0.08	0.08
Lysine	0.00	0.00	0.04	0.08
Methionine	0.12	0.12	0.08	0.08
Calculated nutrients				
Metabolizable Energy (Kcal/kg)	2900	2899	2899	2901
Crude protein (%)	14.55	14.59	14.54	14.57
Calcium (%)	4.142	4.138	4.143	4.141
Available phosphorus (%)	0.439	0.441	0.440	0.441
Crude cellulose %	2.113	2.625	3.133	4.152
Lysine (%)	0.770	0.744	0.753	0.740
Methionine (%)	0.367	0.364	0.359	0.362
Methionine +Cystine, %	0.632	0.640	0.640	0.640

[1] Vitamin premix provided the following per kg of diet: vitamin A, 8.800 IU; vitamin D3, 2.200 IU; vitamin E, 11 mg; nicotinic acid, 44 mg; Cal-D-Pantothenate, 8.8 mg; riboflavin 4.4 mg; thiamine 2.5 mg; vitamin B12, 6.6 mg; folic asit, 1 mg; D-Biotin, 0.11 mg; choline, 220 mg.
[2] Mineral premix provided the following per kg of diet: manganese, 80 mg; copper, 5 mg; iron, 60 mg; zinc, 60 mg; cobalt, 0.20 mg; iodine, 1 mg; selenium, 0.15 mg.

3. Results and Discussion

The performance and egg quality parameters are shown in Table 2 and Table 3, respectively. The treatments had no effect on egg production, egg weight, egg mass, feed intake and feed conversion ratio (Table 2), eggshell weight, eggshell thickness and eggshell breaking strength (Table 3).

In the present study, no significant differences on egg production, feed intake, feed conversion ratio, egg weight and egg mass were observed among dietary treatments. Our findings agree those of Shi et al. (2012) reported that diets supplemented with 8.26, 16.52, and 24.84% SFM had no significant effect on body weight gain, egg production, egg mass, feed intake or feed conversion in layers. Yalçın et al. (2000) and Rezaei & Hafezian (2007) reported that SFM supplementation to laying hens diets had no effect on body weight change. Şenköylü et al. (2004) reported that containing different levels (0, 15 and 20 %) of SFM to diets with enzyme supplementation had no significantly effect on egg production in laying hens. Supporting to results were reported Francesch et al. (1995), Tsuzuki et al. (2003), Viera et al. (1992) and Yalçın et al. (2008). Also, these researchers reported that SFM and enzyme supplementation to diets in laying hens had no significantly effect on egg weight, egg mass, feed intake and feed conversion ratio. Vieira et al. (1992) reported that dietary sunflower meal levels up to 40.5% did not affect layer performance. However, current commercial layer strains are more nutrient-demanding, and therefore, high levels of SFM in the diet would compromise their performance. Other authors have also reported the inclusion of high SFM levels (20.0%) in commercial layer diets (Senkoylu et al., 2004). However, the sunflower meal utilized in the diets was submitted only to partial oil extraction, and therefore nutritional value was much higher than that of the SFM utilized in the present study. Most of the results reported in the literature recommend the utilization of SFM levels close to those included here (Tsuzuki et al., 2003; Casartelli et al., 2006; Rezaei & Hafezian 2007; Junqueira et al., 2010). Furthermore, Uwayjan et al. (1983) found that the inclusion of 30% SFM in diet did not affect feed conversion in hens, but feed consumption was reduced, which might be due to the increase in the energy content of diet.

Table 2
Effect of enzyme supplementation to laying hens diets containing different levels of sunflower seed meal on performance from 44 to 56 weeks of age (Mean ± SE)

Diets		Egg production (%)	Feed intake (g/hen/day)	Feed conversion ratio (g feed/g egg mass)	Egg weight (g)	Egg mass (g/hen/day)
SFM (%)	***Enzyme (IU/kg)***					
	0	90.9 ± 1.29	109.0 ± 1.32	2.0 ± 0.04	55.8 ± 0.84	61.4 ± 0.20
0	500	91.2 ± 2.84	108.2 ± 1.97	2.0 ± 0.08	54.8 ± 1.86	60.1 ± 0.32
	1000	88.8 ± 1.67	108.8 ± 2.10	2.0 ± 0.02	53.9 ± 1.11	60.7 ± 0.13
	2000	87.7 ± 1.10	105.9 ± 3.49	2.1 ± 0.10	51.4 ± 1.53	60.2 ± 0.44
	0	91.1 ± 2.43	103.6 ± 2.07	1.9 ± 0.04	54.5 ± 1.45	59.9 ± 0.19
5	500	88.1 ± 2.57	101.5 ± 4.30	2.0 ± 0.06	51.6 ± 2.10	60.1 ± 0.07
	1000	90.1 ± 2.34	111.3 ± 0.59	2.4 ± 0.33	49.6 ± 5.95	61.2 ± 0.29
	2000	90.1 ± 0.92	109.0 ± 2.37	2.1 ± 0.08	52.5 ± 2.15	60.5 ± 0.27
	0	88.3 ± 2.26	105.5 ± 2.81	2.0 ± 0.07	53.3 ± 1.19	60.4 ± 0.47
10	500	90.4 ± 2.15	106.9 ± 3.75	2.0 ± 0.03	54.1 ± 1.94	59.8 ± 0.78
	1000	90.3 ± 0.88	107.0 ± 0.60	2.0 ± 0.02	54.7 ± 0.62	60.5 ± 0.18
	2000	88.3 ± 1.62	105.0 ± 1.51	2.0 ± 0.07	52.7 ± 1.90	60.9 ± 0.64
	0	85.9 ± 1.84	106.4 ± 1.56	2.1 ± 0.08	50.1 ± 2.49	60.7 ± 0.21
20	500	89.8 ± 3.52	107.0 ± 2.20	2.0 ± 0.10	54.7 ± 1.96	60.9 ± 0.29
	1000	89.7 ± 1.65	110.3 ± 0.78	2.0 ± 0.03	54.2 ± 0.82	60.4 ± 0.41
	2000	88.8 ± 1.72	108.6 ± 0.54	2.0 ± 0.03	53.8 ± 0.97	60.5 ± 0.64

Table 3
Effect of enzyme supplementation to laying hens diets containing different levels of sunflower seed meal on egg quality from 44 to 56 weeks of age (Mean ± SE)

Diets		Eggshell weight (g/100 g egg)	Eggshell thickness (mm)	Eggshell breaking strength (kg)
SFM (%)	***Enzyme (IU/kg)***			
	0	9.0 ± 0.15	0.35 ± 0.002	3.7 ± 0.10
0	500	8.9 ± 0.09	0.35 ± 0.005	3.6 ± 0.10
	1000	9.3 ± 0.10	0.36 ± 0.001	4.0 ± 0.10
	2000	8.8 ± 0.14	0.36 ± 0.003	3.7 ± 0.06
	0	9.3 ± 0.14	0.36 ± 0.002	4.1 ± 0.12
5	500	9.1 ± 0.18	0.36 ± 0.003	3.9 ± 0.17
	1000	9.1 ± 0.24	0.35 ± 0.007	3.9 ± 0.13
	2000	9.2 ± 0.15	0.35 ± 0.005	3.8 ± 0.15
	0	9.1 ± 0.20	0.35 ± 0.002	3.8 ± 0.16
10	500	8.8 ± 0.29	0.35 ± 0.007	3.7 ± 0.12
	1000	9.1 ± 0.16	0.35 ± 0.001	3.9 ± 0.14
	2000	9.2 ± 0.13	0.36 ± 0.004	4.0 ± 0.07
	0	9.0 ± 0.11	0.35 ± 0.001	3.7 ± 0.05
20	500	9.2 ± 0.10	0.36 ± 0.002	3.9 ± 0.24
	1000	8.9 ± 0.10	0.35 ± 0.002	3.7 ± 0.10
	2000	9.2 ± 0.16	0.36 ± 0.002	3.9 ± 0.14

Baghban-Kanani et al. (2018) reported that the supplementation of SFM up to 20% in diet with multi-enzyme complex in laying hens did not appear to cause any adverse effects on egg production and quality in laying hens. Çiftçi et al. (1999), Tsuzuki et al. (2003) and Yalçın et al. (2008) reported that containing different levels of SFM to diets with enzyme supplementation had no significantly effect on egg shell weight, egg shell thickness and eggshell breaking strength in laying hens.

The result of this study that containing 20 % sunflower seed meal without the addition of enzyme to laying hens diets can be used without adversely affecting performance and eggshell quality.

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