

## Effects of dietary administration of multi-enzyme on productive performance of laying hens fed different levels of safflower meal

A. Ehsani, A. H. Mahdavi\*, A. H. Samie and B. Dolatkah

*Department of Animal Sciences, College of Agriculture, Isfahan University of Technology, Isfahan, Iran*

---

### Abstract

The present study aimed to investigate the effects of dietary supplementation of multi-enzyme on productive performance of laying hens fed different levels of safflower meal. A total of 250 Leghorn laying hens (Hy-Line W-36) 47 weeks-old were randomly assigned into 10 experimental treatments with 5 replicates of 5 birds each. Experimental treatments consisted of five safflower meal levels (0.0, 2.5, 5.0, 7.5 and 10.0 g/100g of diet) with and without multi-enzyme (Bergazym P®, 0.0 and 0.1 g/100g of diet) as a 5 × 2 factorial arrangement that fed during a 10 weeks feeding trial. During the experimental period, eggs were collected and weighed daily, and feed consumption was weekly recorded. Then, the productive performance indices including feed consumption, egg production percentage, egg weight, egg mass and feed conversion ratio (FCR) were reported as 14 d intervals and during entire trial period. Feeding laying hens 2.5% and 5% safflower meal showed tendency to increased egg mass in the entire experimental period. This improvement was largely due to the marginal increase in egg production percentage. The best FCR was found in birds fed on diets contained in 0.0% to 5.0 % safflower meal. Feed conversion ratio increased significantly after feeding at least 7.5 % safflower meal. Increase in FCR was due to the trend in decreased egg production and contemporaneous increase in feed consumption. On the other hand, using multi-enzyme did not improve FCR. Adding multi-enzyme increased the egg production percentage and egg mass. Therefore, our results indicated that safflower meal can be included in diets of laying hens up to 5% with no adverse effect on performance. Also, administration of cocktail commercial enzyme to the diets of laying hens would have beneficial effects on productive performance, especially in terms of egg production.

**Key words:** safflower meal, multi-enzyme, laying hens, performance

## **Introduction**

Feed represents the major cost of poultry production, constituting up to 70 percent of the total. Therefore, selection of feedstuffs is very important (Ravindran, 2012) and could have the greatest impact on the net profit (Kwee, 1970). One of the ways for reducing feed costs is finding alternative sources of protein such as soybean meal, which is now referred as a gold standard. One of the available feedstuff for this purpose is safflower meal that is derived from the extraction of safflower oil. Leftover has 12 to 16 percent of fiber and about 25 to 36 percent protein (Kuzmicky and Kohler, 1968; Mailer et al., 2008). Safflower has some anti-nutritional factors such as fiber and so, it has some limitations in poultry nutrition and could reduce the availability of nutrients if it is used in large amounts in rations (Kuzmicky and Kohler, 1968; Abughazaleh et al., 2005). Some types of fibers may alter gastric pH and buffering capacity (Webster, 1986) or they may reduce the metabolizable energy of food (Bach Kundsén, 2001). Furthermore, if it used as the only source of dietary protein, safflower meal has poor percentage of the amino acid lysine, and can cause minor deficiencies in amino acids arginine, methionine, glycine and cysteine (Kratzer and Willimams, 1951). Nonetheless, Halloran (1961) indicated that if one-third of dietary protein be supplied by fish meal, the rest of the protein requirements can be provided by safflower meal without any problem. Also, Valadez et al. (1965) concluded that 50% of soybean meal can be replaced with safflower meal in corn-soybean meal-based diets.

In order to reduce the harmful effects of safflower meal enzymes can be used. Two major groups of enzymes that are currently used in poultry feed are phytase and degrading enzymes of non-starch polysaccharides (NSP's; Wenk and Boessinger, 1993). Enzymes break down compounds that are not easily digested and absorbed, and reduce adhesion in the gastrointestinal tract (Brenes et al., 1993). They increase the digestibility and absorption of nutrients and the efficiency of feed consumption (Airall et al., 1995). These functions result in reduced costs and increased net profit (Classen, 1998). Bergazym P<sup>®</sup> (Berg+Schmidt GmbH & Co. KG. Ander Alster 81, Hamburg, Germany) is a multi-enzyme consisting of xylanase,  $\beta$ -glucanase,  $\alpha$ -amylase and protease. Therefore, this enzyme could have enzymatic activity against more than one substrate. Because most livestock rations are generally a mixture of several feedstuffs multi enzymatic activity is desirable. Using a multiple enzyme could be more effective than a pure enzyme (Campbell and Bedford, 1992).

The present study was conducted to evaluate the effects of multi-enzyme supplementation in the safflower meal-included diets on performance characteristics of laying hens such as average daily feed intake, egg production percentage, egg weight, egg mass and FCR.

## Materials and Methods

### Bird management and experimental design

Two hundred and fifty 47-week old white leghorn hens (Hy-Line, W36) were assigned to a  $5 \times 2$  factorial arrangement of treatments including five levels of safflower meal (0.0%, 2.5%, 5.0%, 7.5% and 10%) and two levels of multi-enzyme (0.0% and 0.1%). Treatments were randomly assigned to the cages (5 replicates) containing 5 birds each scattered in bird house according to a completely randomized design. Treatments were a sample of the population to which we could make inferences. Every experimental unit has the same probability of receiving any treatment. Randomization is performed using a random number table. Throughout the 70 days of the experiment, birds were housed in the environmentally controlled cage system (45 × 50 cm) with 16-hours light to 8-hours dark lighting program.

### Experimental diets and performance indices

All treatment groups received the normal isocaloric and isonitrogenous diets formulated to meet or exceed the nutrient requirements of laying hens as recommended by Hy-Line W-36 manual (Table1). Hens had free access to water and to the experimental diets during 10-week experimental period and body weights were recorded at the beginning and the end of experiment.

During the present study, laid eggs were collected daily (at 8:00) and immediately weighed. These data (egg numbers and weights) were used to calculate egg mass. Feed consumption, egg production percentage, egg weight, egg mass and FCR were reported as 14-day intervals and during entire trial period.

### Statistical analyses

All data were analyzed for variance (ANOVA) using the Generalized Linear Model procedure (PROC GLM) of SAS software (SAS Institute, 2001). The following model was assumed in the analysis of all traits.  $Y_{ijk} = \mu + A_i + B_j + AB_{ij} + e_{ijk}$ , where  $Y_{ijk}$  = observed value for a particular character,  $\mu$  = overall mean,  $A_i$  = effect of the  $i^{\text{th}}$  level of safflower meal,  $B_j$  = effect of the  $j^{\text{th}}$  level of dietary multi-enzyme,  $AB_{ij}$  = the respective interaction between  $i^{\text{th}}$  and  $j^{\text{th}}$  levels of safflower meal and multi-enzyme, and  $e_{ijk}$  = random error associated with the  $ijk^{\text{th}}$  recording. In the present study, a threshold of significance was set at  $P < 0.05$  and trends were declared at  $0.05 < P < 0.10$ .

**Table 1.** The composition and nutrient content of experimental diets (47 to 56 weeks of age)

Item	Dietary treatments <sup>1</sup>				
	A	B	C	D	E
<i>Ingredient (%)</i>					
Soybean meal	21.1	20.2	19.3	18.4	17.5
Soybean oil	2.50	2.50	2.50	2.50	2.50
Fat powder	2.50	2.50	2.50	2.50	2.50
Safflower meal	0.00	2.50	5.00	7.50	10.0
Wheat bran	5.36	4.04	2.72	1.31	0.00
Chip wood	2.00	1.50	1.00	0.50	0.00
Oyster shell	5.00	5.00	5.00	5.00	5.00
Monocalcium phosphate	1.50	1.49	1.48	1.46	1.45
Limestone	5.52	5.51	5.51	5.50	5.50
Common salt	0.22	0.22	0.22	0.21	0.21
L-Lysine HCL	0.03	0.04	0.05	0.06	0.07
DL- Methionine	0.17	0.17	0.16	0.16	0.15
Na bicarbonate	0.70	0.73	0.76	0.80	0.82
Mineral premix <sup>2</sup>	0.25	0.25	0.25	0.25	0.25
Vitaminepremix <sup>3</sup>	0.25	0.25	0.25	0.25	0.25
Crude fat (%)	7.34	7.34	7.33	7.33	7.32
Crude fiber (%)	5.24	5.28	5.32	5.36	5.41
Crude protein (%)	14.6	14.6	14.6	14.6	14.6
Lysine (%)	0.75	0.75	0.75	0.75	0.75
Methionine (%)	0.42	0.42	0.41	0.41	0.41
Methionine+cysteine (%)	0.65	0.65	0.65	0.65	0.65
Calcium (%)	4.17	4.17	4.17	4.17	4.17
Available phosphorus (%)	0.44	0.44	0.44	0.44	0.44
Na (%)	0.30	0.31	0.32	0.33	0.33
Electrolyte balance (mEq/Kg)	249.8	250.0	250.0	249.9	250.0

<sup>1</sup>A: control group, B: 2.5% safflower meal treatment, C: 5% safflower meal treatment, D: 7.5% safflower meal treatment, E: 10% safflower meal treatment.

<sup>2</sup>Mineral premix provided per kilogram of diet: manganese, 80 mg; copper, 10 mg; iodine [from Ca (IO<sub>3</sub>)<sub>2</sub>·H<sub>2</sub>O], 0.8 mg; cobalt, 0.25 mg; selenium, 0.3 mg; zinc, 80 mg; iron, 80 mg.

<sup>3</sup>Vitamin premix provided per kilogram of diet: vitamin A (from vitamin A acetate), 10000 IU; vitamin D<sub>3</sub>, 2500 IU; vitamin E (from dl- $\alpha$ -tocopheryl acetate), 10 IU; vitamin B<sub>1</sub>, 2.2 mg; vitamin B<sub>2</sub>, 4 mg; pantothenic acid, 8 mg; vitamin B<sub>6</sub>, 2 mg; niacin, 30 mg; vitamin B<sub>12</sub>, 0.015 mg; folic acid, 0.5 mg; biotin, 0.15 mg; choline chloride, 200 mg.

## Results and Discussion

### Feed consumption

Average feed consumption in different experimental groups is presented in Table 2. Feed consumption significantly ( $P < 0.01$ ) increased after feeding different levels of safflower meal during the first and second two weeks. Highest and lowest feed consumption were observed in hens fed diet containing 10% safflower meal and control group, respectively. However, feed consumption was not affected by different levels of safflower meal in the third to fifth two-week periods. Using at least 2.5% of safflower meal increased feed consumption at a marginal level ( $P = 0.08$ ) for the entire experimental period.

Increased feed consumption in groups fed high levels of safflower meal compared with control group was not due to the higher levels of crude fiber or its effect on intestinal passage rate. Chemical analysis of diets showed that this index was numerically very close to each other. The increased feed consumption in

**Table 2.** Effects of different levels of safflower meal and multi-enzyme on average daily feed intake of laying hens (g/hen/day)

		Weeks					
		1-2	3-4	5-6	7-8	9-10	1-10
Safflower meal (%)							
0.0		107.0 <sup>c</sup>	109.6 <sup>c</sup>	108.7	108.7	107.4	108.3
2.5		109.2 <sup>bc</sup>	133.6 <sup>bc</sup>	112.8	111.2	111.0	111.5
5.0		112.9 <sup>ab</sup>	115.5 <sup>ab</sup>	115.7	111.9	113.4	113.9
7.5		112.1 <sup>ab</sup>	114.5 <sup>ab</sup>	113.9	112.0	113.1	113.1
10.0		115.4 <sup>a</sup>	118.3 <sup>a</sup>	116.1	112.9	103.7	113.3
SE		1.25	1.49	1.86	1.65	1.43	1.48
0.0		110.9	113.8	112.8	110.8	107.6	111.2
0.1		111.7	114.9	114.1	111.9	111.8	112.9
SE		1.01	1.12	1.34	1.07	2.67	1.03
0.0	0.0	106.1	109.6	107.5	104.0	104.8	106.4
0.0	0.1	107.9	109.7	109.8	113.4	110.2	110.2
2.5	0.0	109.9	114.8	114.6	113.5	112.9	113.1
2.5	0.1	108.4	112.4	110.9	108.8	109.1	109.9
5.0	0.0	114.1	116.3	115.3	114.1	116.2	115.2
5.0	0.1	111.7	114.8	116.2	109.8	110.6	112.6
7.5	0.0	110.1	109.7	109.6	109.6	110.9	109.9
7.5	0.1	114.1	119.4	118.3	114.4	115.3	116.3
10.0	0.0	114.5	118.6	116.8	112.8	116.0	115.7
10.0	0.1	116.2	118.1	115.3	113.1	114.2	115.4
SE		1.65	1.84	2.06	2.07	1.78	1.70
				<i>P</i> -value			
Safflower meal		0.01	0.01	0.08	0.35	0.65	0.08
Multi-enzyme		0.56	0.43	0.45	0.42	0.36	0.22
Interaction		0.48	0.04	0.25	0.01	0.41	0.11

<sup>a-c</sup>Means with no common superscript within each column are significantly different ( $P < 0.05$ ).

these groups was likely due to the palatability of safflower meal. Peterson et al. (1957) reported that feed consumption was same in the chickens that were fed 0, 9.5, 15 and 20 percent of safflower meal. Their study showed that fiber of safflower meal did not have any effect on feed consumption and feed efficiency (Peterson et al., 1957).

In the present study, adding the multi-enzyme numerically but not significantly increased feed intake compared with similar groups without enzyme. Feed consumption was influenced by dietary multi-enzyme and safflower meal interaction during the second and fourth two weeks ( $P < 0.05$ ). However, feed consumption did not differ for the entire experimental period ( $P = 0.11$ , Table 2). The increase in safflower meal level in the diet caused a considerable increase in feed intake when the diets were not supplemented by dietary multi-enzyme ( $P < 0.01$ ).

Dipeolu et al. (2005) showed that dietary multi-enzyme could improve egg production percentage by increasing the digestibility of dietary fiber and metabolizable energy. Kocher et al. (2001) reported that the safflower meal supplementation with a carbohydrase multi-enzyme slightly increased feed consumption of broiler chicks in through of the husbandry period. Slominski and Campbell (1990) demonstrated that enzymatic processing of rapeseed meal diets improved feed consumption in broilers. They concluded that improved feed consumption was due to the higher digestibility of rapeseed fiber in the enzymatic diets. Enzymes disintegrate the fiber-bound proteins and non-starch polysaccharides of the cell wall, and improve digestibility and bioavailability of fibers, proteins and NSP's (Slominski and Campbell, 1990).

**Table 3.** Safflower meal and multi-enzyme interactions on feed intake at the second and fourth two weeks of experimental period

Safflower meal (%)	Second two weeks		Fourth two weeks	
	No enzyme	Enzyme	No enzyme	Enzyme
0.0	109.6 <sup>b</sup>	109.7	104.0 <sup>b</sup>	113.4
2.5	114.8 <sup>ab</sup>	112.4	113.5 <sup>a</sup>	108.8
5.0	116.3 <sup>a</sup>	114.8	114.1 <sup>a</sup>	109.8
7.5	109.7 <sup>b</sup>	119.4	109.6 <sup>ab</sup>	114.4
10.0	118.6 <sup>a</sup>	118.1	112.8 <sup>a</sup>	113.1
		<i>P</i> -value		
	0.01	0.06	0.01	0.28

<sup>ab</sup>Means with no common superscript within each column are significantly different ( $P < 0.05$ ).

**Table 4.** Effects of different levels of safflower meal and multi-enzyme on egg production percentage of laying hens (47 to 56 weeks of age)

Safflower meal (%)		Weeks					
		1-2	3-4	5-6	7-8	9-10	1-10
0.0		74.7	79.6	80.0	79.0 <sup>b</sup>	80.7	78.8
2.5		77.0	81.6	82.3	82.7 <sup>ab</sup>	85.1	81.7
5.0		76.7	85.3	86.4	88.5 <sup>a</sup>	85.6	84.7
7.5		78.3	79.6	79.6	79.5 <sup>b</sup>	79.5	79.3
10.0		76.1	80.6	81.4	79.1 <sup>b</sup>	79.3	79.3
SE		2.68	1.88	2.03	2.22	2.57	1.68
0.0		74.3	80.0	80.0 <sup>b</sup>	80.5	80.0	79.0 <sup>b</sup>
0.1		78.8	82.6	83.8 <sup>a</sup>	83.0	84.0	82.4 <sup>a</sup>
SE		1.57	1.19	1.29	1.60	1.68	1.05
0.0	0.0	73.7	78.8	79.1	76.8	79.7	77.6
0.0	0.1	75.7	80.3	80.8	81.1	81.7	79.9
2.5	0.0	70.8	81.1	81.7	83.7	83.8	80.2
2.5	0.1	83.1	82.0	82.8	81.7	86.3	83.2
5.0	0.0	78.8	84.3	85.7	88.3	87.4	84.9
5.0	0.1	74.6	86.3	86.8	88.6	83.7	84.0
7.5	0.0	77.7	76.3	73.1	75.1	71.3	74.7
7.5	0.1	78.8	82.8	86.0	84.0	87.7	83.9
10.0	0.0	70.6	79.4	80.3	78.6	78.0	77.4
10.0	0.1	81.7	81.7	82.6	79.7	80.6	81.2
SE		2.81	2.51	2.55	3.07	3.29	3.65
			<i>P</i> -value				
Safflower meal		0.89	0.24	0.15	0.04	0.25	0.07
Multi-enzyme		0.05	0.14	0.04	0.25	0.09	0.02
Interaction		0.12	0.86	0.20	0.56	0.10	0.32

<sup>ab</sup>Means with no common superscript within each column are significantly different ( $P < 0.05$ ).

### Egg production percentage

As shown in Table 4, egg production percentages were increased in the second and fourth two-week ( $P < 0.05$ ) after feeding 2.5% and 5% of safflower meal. This improvement could be attributed to increased feed consumption, and implies adaptation ability of laying hens to high proportion of dietary fiber (Wyatt and Goodman, 1993). In this regard, further experiments will be required to confirm this hypothesis.

**Table 5.** Effects of different levels of safflower meal and multi-enzyme on egg weight (g, 47 to 56 weeks of age)

		Weeks					
		1-2	3-4	5-6	7-8	9-10	1-10
Safflower meal (%)							
0.0		63.7	64.8	65.7 <sup>a</sup>	65.7	66.4 <sup>ab</sup>	65.3
2.5		63.3	64.5	65.8 <sup>a</sup>	65.4	66.9 <sup>a</sup>	65.2
5.0		62.5	64.0	64.6 <sup>ab</sup>	64.9	64.4 <sup>b</sup>	64.1
7.5		62.1	62.9	63.3 <sup>b</sup>	64.5	64.5 <sup>b</sup>	63.5
10.0		61.5	64.0	64.6 <sup>ab</sup>	64.1	65.1 <sup>ab</sup>	63.9
SE		0.58	0.52	0.50	0.69	0.60	0.49
0.0		62.6	64.2	65.0	65.2	65.5	64.5
0.1		62.6	63.9	64.6	64.7	65.5	64.3
SE		0.39	0.35	0.37	0.45	0.44	0.34
0.0	0.0	64.4	65.7	66.7	66.3	67.1	66.0
0.0	0.1	63.1	63.9	64.7	65.2	65.7	64.5
2.5	0.0	62.8	64.6	65.7	65.9	66.1	65.0
2.5	0.1	63.8	64.5	65.9	64.9	67.7	65.4
5.0	0.0	62.8	64.3	64.4	65.7	64.9	64.4
5.0	0.1	62.3	63.7	64.7	64.1	63.9	63.8
7.5	0.0	61.6	62.6	62.9	62.9	64.1	62.9
7.5	0.1	62.6	63.1	63.6	66.1	64.9	64.1
10.0	0.0	61.6	64.0	65.2	64.9	65.2	64.2
10.0	0.1	61.4	64.0	63.9	64.4	65.0	63.6
SE		0.80	0.73	0.70	0.93	0.85	0.69
		<i>P</i> -value					
Safflower meal		0.09	0.19	0.01	0.48	0.03	0.06
Multi-enzyme		0.98	0.47	0.39	0.51	0.95	0.60
Interaction		0.61	0.69	0.35	0.11	0.48	0.43

<sup>ab</sup>Means with no common superscript within each column are significantly different ( $P < 0.05$ ).

Absence of simultaneity trend between improvement of egg production and increasing levels of safflower meal was probably due to a higher amount of anti-nutritional factors (such as NSP's and holocellulose) at the levels of 7.5% and 10 % safflower meal. It is probable that administrating of multi-enzyme could suppress the anti-nutritional effects of these components, thereby leading to egg production improvement by about 3.5 percent in whole of the experimental period ( $P < 0.05$ ). Ravindran et al. (2007) reported that using  $\beta$ -glucanase in barley-based diets improved metabolizable energy in broilers via reduction of digestive tract viscosity. There was no interaction between different levels of safflower meal and multi-enzyme on egg production percentage, so that in each level of safflower meal, after supplementation of multi-enzyme increased egg production percentage.

### Egg weight

The effects of different levels of safflower meal and multi-enzyme on egg weight are shown in Table 5. Feeding at least 5% of safflower meal decreased egg weight during the third and fifth two-week ( $P < 0.05$ ) as well as a trend for the entire experimental period ( $P = 0.06$ ). Furthermore, using dietary multi-enzyme had no significant effect on egg weight during experimental weeks. This finding is in agreement with the results of Yu et al. (2007) who indicated that using beta-manase had no remarkable effect on egg weight. However, Jackson et al. (1999) reported that supplementing hemicell enzyme at the age of 18 to 30 weeks, caused an

increase in egg weight in low energy groups so that they didn't have any difference with high energy ones. The interactions between multi-enzyme and safflower meal were not significant in whole of the experimental period.

### Egg mass

Feeding laying hens 2.5% and 5% safflower meal showed tendency to increased egg mass in the entire experimental period ( $P = 0.07$ , Table 6).

Dietary safflower meal by multi-enzyme interactions was not significant for egg mass so that, using 2.5% and 5% safflower meal with or without multi-enzyme increased egg mass. This enhancement was largely due to the increase in egg production. Some studies have used different meals as protein source in poultry diets and concluded that egg mass index is not influenced by using different sources of meals, if nutritional requirements of poultry are adequately provided. For example, Perez et al. (2000) showed that palm kernel meal had no significant effect on mean of egg production efficiency; whereas using 50 percent of palm kernel meal decreased the average of egg production efficiency.

**Table 6.** Effects of different levels of safflower meal and multi-enzyme on egg mass (g/d per hen)

		Weeks					
		1-2	3-4	5-6	7-8	9-10	1-10
Safflower meal (%)							
0.0		47.5	51.4	52.4	51.9 <sup>b</sup>	53.6	51.4
2.5		48.5	52.7	54.2	53.9 <sup>ab</sup>	56.5	53.2
5.0		48.0	54.5	55.8	57.3 <sup>a</sup>	55.1	54.2
7.5		48.6	50.0	50.5	50.9 <sup>b</sup>	51.4	50.3
10.0		46.7	51.6	52.6	50.5 <sup>b</sup>	51.7	50.6
SE		1.80	1.17	1.43	1.51	1.75	1.13
Multi-enzyme (%)							
0.0		46.4	51.4	52.0	52.3	52.5	50.9 <sup>b</sup>
0.1		49.3	52.7	54.3	53.6	54.8	52.9 <sup>a</sup>
SE		1.07	0.75	0.91	1.07	1.17	0.74
Interaction							
0.0	0.0	47.2	51.5	52.7	50.9	53.5	51.2
0.0	0.1	47.9	51.2	52.7	52.9	53.7	51.8
2.5	0.0	44.0	52.5	53.8	54.9	55.3	52.1
2.5	0.1	52.9	52.9	54.7	53.0	57.7	54.2
5.0	0.0	49.5	54.2	55.3	57.9	56.7	54.7
5.0	0.1	46.5	54.9	56.3	56.9	53.5	53.6
7.5	0.0	48.0	47.8	46.0	47.3	45.8	47.0
7.5	0.1	49.3	52.2	55.0	54.7	56.9	53.6
10.0	0.0	43.6	50.9	52.4	50.6	51.0	49.7
10.0	0.1	49.8	52.3	52.9	50.5	52.3	51.6
SE		2.33	1.58	1.82	2.05	2.22	1.42
		<i>P</i> -value					
Safflower meal		0.93	0.14	0.12	0.03	0.21	0.07
Multi-enzyme		0.07	0.24	0.08	0.37	0.14	0.04
Interaction		0.12	0.69	0.15	0.25	0.08	0.18

<sup>ab</sup>Means with no common superscript within each column are significantly different ( $P < 0.05$ ).



**Table 7.** Effects of different levels of safflower meal and multi-enzyme on feed conversion ratio of laying hens (47 to 56 weeks of age)

		Weeks					
		1-2	3-4	5-6	7-8	9-10	1-10
Safflower meal (%)							
0.0		2.28	2.14 <sup>bc</sup>	2.08	2.10 <sup>ab</sup>	2.01	2.12 <sup>b</sup>
2.5		2.38	2.16 <sup>abc</sup>	2.09	2.07 <sup>ab</sup>	1.97	2.14 <sup>b</sup>
5.0		2.44	2.12 <sup>c</sup>	2.08	1.95 <sup>b</sup>	2.06	2.13 <sup>b</sup>
7.5		2.36	2.29 <sup>ab</sup>	2.30	2.23 <sup>a</sup>	2.26	2.29 <sup>a</sup>
10.0		2.59	2.32 <sup>a</sup>	2.23	2.28 <sup>a</sup>	1.97	2.28 <sup>a</sup>
SE		0.11	0.05	0.06	0.06	0.09	0.04
0.0		2.48	2.23	2.19	2.15	2.05	2.22
0.1		2.34	2.18	2.11	2.10	2.06	2.16
SE		0.07	0.03	0.04	0.05	0.07	0.03
0.0	0.0	2.29	2.14	2.05	2.05	1.97	2.10
0.0	0.1	2.27	2.15	2.11	2.14	2.06	2.15
2.5	0.0	2.67	2.20	2.14	2.09	2.05	2.23
2.5	0.1	2.09	2.13	2.03	2.06	1.90	2.04
5.0	0.0	2.33	2.15	2.09	1.97	2.05	2.12
5.0	0.1	2.55	2.08	2.07	1.94	2.07	2.14
7.5	0.0	2.33	2.30	2.43	2.36	2.50	2.38
7.5	0.1	2.40	2.28	2.17	2.09	2.03	2.19
10.0	0.0	2.79	2.37	2.27	2.29	1.71	2.29
10.0	0.1	2.39	2.26	2.19	2.27	2.23	2.27
SE		0.11	0.06	0.08	0.09	0.11	0.06
		<i>P</i> -value					
Safflower meal		0.33	0.04	0.06	0.02	0.34	0.01
Multi-enzyme		0.14	0.19	0.18	0.44	0.98	0.09
Interaction		0.06	0.94	0.53	0.52	0.05	0.16

<sup>abc</sup>Means with no common superscript within each column are significantly different ( $P < 0.05$ ).

Dietary safflower meal by multi-enzyme interactions was not significant for egg mass so that, using 2.5% and 5% safflower meal with or without multi-enzyme increased egg mass. This enhancement was largely due to the increase in egg production. Some studies have used different meals as protein source in poultry diets and concluded that egg mass index is not influenced by using different sources of meals, if nutritional requirements of poultry are adequately provided. For example, Perez et al. (2000) showed that palm kernel meal had no significant effect on mean of egg production efficiency; whereas using 50 percent of palm kernel meal decreased the average of egg production efficiency.

As shown in Table 6, dietary supplementation of multi-enzyme in high safflower meal diet groups (7.5% and 10%) increased the egg mass. This improvement was significant at the end of experiment ( $P < 0.05$ ). Increased egg mass was mainly due to the increase in egg production. It is probable that enzyme might decompose non-starch polysaccharides of safflower meal and increase bioavailability of metabolizable energy. Bayram et al. (2008) indicated that the enzyme was able to compensate reduced egg production in the diets after 3% reduction in energy, but could not compensate 3.5% reduction in dietary energy. Also, Yu et al. (2004) demonstrated that beta-manase supplementation could improve egg mass markedly.

## Feed conversion ratio

Feed conversion ratio (FCR) was increased significantly ( $P < 0.05$ ) after feeding at least 7.5% safflower meal at the second and fourth two-week as well as whole of the experiment (Table 7). Increase in the FCR was due to the trend in decreased egg production ( $P = 0.07$ ). The best FCR were found in birds fed on diets contained in 0.0% -5.0% safflower meal. However, Peterson et al. (1957) reported that using diets with high levels of safflower meal (2.5% increase in fiber) decreased broilers growth performance, Kohler et al. (1966) noted that appropriate levels of safflower meal can be used instead of soybean meal.

Reinforced to previous reports, our data indicated that using safflower meal up to 5% had no undesirable effect on FCR. Additionally, the presence of anti-nutritional factors such as cyanide, oxalate and trypsin inhibitor in safflower meal is another possible reason for increased FCR in diets with high levels of safflower meal compared with soybean meal (Ingale and Shrivastava, 2011).

## Conclusion

In conclusion, the present results indicated that safflower meal can be included in diets of laying hens up to 5% with no adverse effects on performance. Also, administration of cocktail commercial enzyme to the diets of laying hens showed beneficial effects on productive performance, especially in terms of egg production.

## References

- Abughazaleh, A. A., M. B. Riley, E. E. Thies, and T. C. Jenkins. 2005. Dilution rate and pH effects on the conversion of oleic acid to trans C18:1 positional isomers in continuous culture. *Journal of Dairy Science*, 88: 4334-4341.
- Airall, M., M. Francesch, M. Anna, and E. EsteveGagrcia. 1995. The difference in intestinal viscosity product by barley and beta-glucanase alter digesta enzyme actives and ideal nutrient digestibility more in broiler chicks than in cocks. *Journalof Nutrition*, 125: 947-955.
- Bach Kundsén, K. H. 2001. The nutritional significance of dietary fiber analysis. *Animal Feed Science and Technology*, 90: 3-20.
- Bayram, I., I. SadiCetingul, A. BurhaneddinAkkaya, and C. Uyarlar. 2008. Effects of bacterial xylanase on egg production in the laying guail (*Coturnixcoturnix japonica*) diets based on corn and soybean meal. *Archivos de Zootecnia*, 11: 69-74.
- Brenes, A., M. Smith, W. Guenter, and R. R. Marquardt. 1993. Effects of enzyme supplementation on the performance and digestive tract size of broiler chickens feed wheat and barley based diets. *Poultry Science*, 72: 1731-1739.
- Campbell, G. L., and M. R. Bedford. 1992. Enzyme application for monogastric feeds: A review. *Canadian Journal of Animal Science*, 72: 449-466.
- Classen, H. L. 1998. Enzymes in action. *Feed Mix. Enzyme*, Special Issue, Pages 12-16.
- Dipeolu, M. A., D. Eruvbetine, E. B. Oguntona, O. O. Bankole, and K. S. Sowunmi. 2005. Comparison of effects of antibiotics and enzyme inclusion in diets of laying birds. *Archivos de Zootecnia*, 54: 3-11.
- Halloran, H. R. 1961. High protein safflower meal for chickens. *Feedstuffs*, 33: 70-71.
- Ingale, S., and K. S. Shrivastava.. 2011. Chemical and bio-chemical studies of new varieties of safflower. *International Journal of the Bioflux Society*, 3: 2.
- Jackson, M. E., D. W. Fodge, and H. Y. Hsiao. 1999. Effects of b-mannanase in corn-soybean meal diets on laying hen performance. *Poultry Science*, 78: 1737-1741.
- Kocher, A., M. D. Choct, L. Morrisroe, and Y. Broz. 2001. Effect of enzyme supplementation on the replacement value of canola meal for soybean meal in broiler diets. *Australian Journal of Agricultural Research*, 52: 447-452.

- Kohler, G. O., D. D. Kuzmicky, R. Palter, J. Guggolz, and V. V. Hering. 1966. Safflower meal. *The Journal of the American Oil Chemists' Society*, 43: 413-415.
- Kratzer, F. H., and D. Willimams. 1951. Safflower oil meal in rations for chicks. *Poultry Science*, 30: 417-421.
- Kuzmicky, D. D., and G. O., Kohler. 1968. Safflower meal utilization as a protein source for broiler rations. *Poultry Science*, 47(4): 1266-1270.
- Kwee, W. H. 1970. Recovery of nutrients from tomato cannery wastes and vines-their functional and nutritional properties. PhD. dissertation, Univ. of Maryland, College Park, MD.
- Mailer, R. J., T. D., Potter, R., Redden, and J. Ayton. 2008. Quality evaluation of safflower (*Carthamustinctorius* L.) cultivars. 7<sup>th</sup> *International Safflower Conference*. Australia.
- Perez, J. F., A. G. Gernatl, and J. G. Murillo. 2000. The effect of different levels of palm kernel meal in layer diets. *Poultry Science*, 79: 77-79.
- Peterson, C. F., A. C. Wiese, G. J. Anderson, and C. E. Lampman. 1957. The use of safflower oil meal in poultry rations. *Poultry Science*, 39: 3-8.
- Ravindran, V. 2012. Poultry feed availability and nutrition in developing countries. Available online at <http://www.fao.org>, accessed on 10 December 2012.
- Ravindran, V., Z. V. Tilman, P. C. H. Morel, G. Ravindran, and G. D. Coles. 2007. Influence of  $\beta$ -glucanase supplementation on the metabolisable energy and ileal nutrient digestibility of normal starch and waxy barleys for broiler chickens. *Animal Feed Science and Technology*, 134: 45-55.
- SAS Institute. 2001. SAS User's Guide. Version 8.02 ed. SAS Institute Inc., Cary, NC.
- Slominski, B. A., and L. D. Campbell. 1990. None-starch polysaccharides of canola meal: quantification, digestibility in poultry and potential benefit of dietary enzyme supplementation. *Journal of the Science of Food and Agriculture*. 53: 175-184.
- Valadez, S., W. R. Featherston, and R. A. Pickett. 1965. Utilization of safflower meal by the chick and its effect upon plasma lysine and methionine concentrations. *Poultry Science*. 44:909-915.
- Webster, I. 1986. Oats: Chemistry and Technology. *American Association of Cereal Chemists*. Inc. USA.
- Wenk, C., and M. Boessinger. 1993. Enzymes In Animal Nutrition. Pages 13-16 in *Proceeding of the 1<sup>st</sup> symposium kartauseIttingen*. Switzerland.
- Wyatt, C., and T. Goodman. 1993. Utilization of feed enzymes in laying hen rations. *Journal of Applied Poultry Science*, 2: 68-74.
- Yu, B., C. Y. Jan, T. K. Chung, T. T. Lee, and P. W. S. Chiou. 2004. Exogenous phytase activity in the gastrointestinal tract of broiler chickens. *Animal Feed Science and Technology*, 117: 295-303.
- Yu, B., S. T. Wu, C. C. Liu, R. Gauthier, R. Peter, and W. S. Chiou. 2007. Effects of enzyme in a maize-soybean diet on broiler performance. *Animal Feed Science and Technology*, 134: 283-294.