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## Replacing of soybean meal with sunflower meal with and without multi-enzyme on laying performance and egg quality in Hy-Line laying hens

M. Marami<sup>1</sup>, A. Nobakht<sup>1\*</sup>, Y. Mehmannaavaz<sup>1</sup>, F. Mazlum<sup>1</sup>, S. Mahdavi<sup>1</sup>

*Department of Animal Science, Maragheh Branch, Islamic Azad University, Maragheh, Iran*

**ABSTRACT:** This experiment was performed to evaluate the replacement effects of different soybean meal levels with sunflower meal on laying performance and egg quality parameters. In this experiment, 360 laying hens ( $1530 \pm 20$  g) from the age of 47 to 57 weeks, in 9 treatments and 5 replications (8 hens in each replication) were divided into a  $3 \times 3$  factorial arrangement consisting of sunflower meal (0, 30, and 60%) and enzyme additive (0, 100 and 200 g/ton) in a completely randomized design for 10 weeks. The results showed that the replacement of soybean meal with sunflower meal and enzyme has significant effects on the performance of laying hens ( $P < 0.05$ ). So that 60% replacement of soybean meal with sunflower meal increased the amount of daily feed intake of laying hens ( $P < 0.05$ ). The use of 200 gr/ton of multi enzyme in laying hens diet, without having significant effects on other performance parameters, increased the amount of daily feed intake ( $P < 0.05$ ). Concurrently, there was a non-significant trend for egg quality parameters ( $P > 0.05$ ), but weights of egg albumin were affected significantly ( $P < 0.05$ ) substitution of 60% of sunflower meal in diets increased the amount egg albumin weight. In general, in laying hens, it is possible to replace 60% of soybean meal with sunflower meal without adversely affecting the performance and egg quality. The use of a multi-enzyme only increased the amount of feed intake.

**Keywords:** Egg quality, Laying hens, Laying performance, Soybean meal, Sunflower meal

*Corresponding Author:*

A. Nobakht, Department of Animal Science, Maragheh Branch, Islamic Azad University, Maragheh, Iran  
E-mail address: anobakht20@yahoo.com

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## INTRODUCTION

Protein is a major limiting factor and the second most expensive substance in poultry diet composition (Iji et al., 2017). In many developing countries, poultry feed prices rise due to their dependence on soybean meals (Chisoro et al., 2017; Onsongo et al., 2018). Therefore, to reduce this dependence and reduce the cost of the diet, it is necessary to use appropriate protein source alternatives (Giannenas et al., 2017; Ayaşan and Ayaşan, 2020; Sevim and Ayaşan, 2020). Sunflower meal is a source of plant protein (with 32 to 45% crude protein (CP); which, depending on the oil extraction method and peeling), has 14 to 32% crude fiber (CF), and is a source of insoluble non-starch polysaccharides (Ditta et al., 2017; Karkekanov et al., 2020). Sunflower meal has the highest niacin, riboflavin, choline, biotin, pantothenic acid, and pyridoxine levels (Dinusson, 2017; Saleh et al., 2021). Over the past four decades, research efforts have been made to reduce egg cholesterol content by focusing on genetic selection or changing the diet of laying hens with various nutrients, non-nutrients, or drugs (Elkin, 2017).

The liver is where a significant amount of cholesterol is synthesized. The key enzyme in limiting the synthesis of cholesterol is 3-hydroxy-3-methylglutaryl coenzyme A reductase (HMGR). Synthesized cholesterol is either secreted into the bile, regulated by the enzyme Cholesterol 7 alpha-hydroxylase (CYP7A1), or secreted into plasma in the form of very-low-density lipoproteins (VLDL) (Zhu et al., 2018; Abd El-Moneim and Sabic, 2019; Wang et al., 2019; Hu et al., 2020). Laudadio et al. (2014), in a study using sunflower meal at 160 g / kg diet, found that additional sunflower meal leads to significantly reduced yolk and plasma cholesterol levels. Alagwany et al. (2018) reported that plasma cholesterol was reduced by 75% in the diet using sunflower meals. The fiber content of sunflower meals may also stimulate the binding of cholesterol molecules to bile salts (Trisat et al., 2017). These mechanisms may explain the possible effects of sunflower meals on lowering eggs cholesterol. Therefore, this study aimed to evaluate the replacement of different levels of soybean meal with sunflower meal (with and without enzyme) on laying performance and egg quality parameters in laying hens. The project hypothesized that replacing sunflower meal in the diet of laying hens will improve gastrointestinal health and calcium absorption.

## MATERIAL AND METHODS

The animal welfare committee of Islamic Azad University (Maragheh Branch) approved the animal care protocol (no. 1319-IAU. 01.25.2017) used in this experiment. A total of 360 laying hens (1530 ± 20 g) from the age of 47 to 57 weeks in 9 treatments and 5 replications (8 hens in each replicate) were divided into treatments in a 3 × 3 factorial arrangement consisting of sunflower meal (0, 30, and 60%) and enzyme additive (0, 100 and 200 g/ton) in a completely randomized design for ten weeks. The hens had access to *ad-libitum* feed and water, and the lighting program was 16 hours of light and 8 hours of darkness. The hens were fed a practical ration, adjusted to the recommended nutrient requirements of the Hy-Line W36 strains (Sarsour et al., 2021). Received feed and production amount were calculated daily. The percentage of production, weight of eggs, and egg mass were calculated according to the amount of feed intake (FI), feed conversion ratio (FCR) at the end of the experiment. Every 28 days, 3 eggs from each replicate were randomly selected, and after weighing, they were broken on a flat surface, and the yolk color index was determined. Roche unit was used to determine the yolk color index. A page with different colored bars was used in this method, which adds specific scores to them by increasing the colors, respectively. The Haugh unit was then measured in their concentrated white.

The data were analyzed using the general linear model procedure of SAS (2018). Data were log-transformed before analyzing in case of unequal variances Palangi, 2021. The linear model was:

$$Y_{ijk} = \mu + A_i + B_j + AB_{ij} + \varepsilon_{ijkl}$$

where:  $Y_{ijkl}$  = a dependent variable,  $\mu$  = overall mean,  $A_i$  = the effect of sunflower meal replacement,  $B_j$  = the effect of Multi-enzyme,  $AB_{ij}$  = the interaction of factors A and B, and  $\varepsilon_{ijkl}$  = the residual deviation of the observation from the effects in the model. Tukey's test at the 5% level of probability was used to compare means.

## RESULTS

### Laying performance

The effects of replacing soybean meal with sunflower meal (with and without enzyme) in laying hens rations on laying performance are shown in Table 1. Among the measured parameters, only feed intake was affected by experimental treatments ( $P < 0.05$ ). So that increasing the level of sunflower meal or enzyme

led to an increase the amount of feed intake. The 60% sunflower meal  $\times$  200 g/ton multi-enzyme interactions showed the highest amount of feed intake, and 30% sunflower meal  $\times$  100 g/ton multi-enzyme interactions showed the lowest feed intake.

### Egg quality

The effects of replacing soybean meal with sunflower meal in laying hens rations on egg quality are shown in Table 2. High levels of sunflower meal increased albumin weight but reduced shell crust ( $P < 0.05$ ). Other experimental parameters were not affected by treatments ( $P > 0.05$ ).

## DISCUSSION

### Laying performance

As the world's population extends, requisition for eggs will continue to ascent. To meet this requisition will be a big compete because of the high cost of traditional protein sources for layer hen's diets. Furthermore, poultry nutritionists have been working for

decades on sustainability in higher egg production. Using alternative plant protein sources like sunflower meals are innovative solutions for reducing the cost of the diets and improving production (Saleh et al., 2021). Baghban-Kanani et al. (2018) reported that the supplementation of sunflower meal up to 20% in diet with multi-enzyme complex in laying hens did not appear to cause any adverse effects on egg production in laying hens, which were in accordance with our results. Koçer et al. (2021) concluded that productive performance of laying hens improved with 40 or 50 g/kg more CF in feed, mostly from sunflower meal supplementation. Saleh et al. (2021) suggested that the dietary inclusion of sunflower meal, up to 100 g/kg at a late phase of laying, could improve the production performance. The mean reasons for increase the amount of daily feed intake by increase the amount of sunflower meal in layers' diets, related to composition and energy content of this meal. As sunflower meal has high level of crude fiber and low amount of energy, in contrast to soybean meal, so inclusion high level of it in layer diets increased the

**Table 1.** The effect of replacing soybean meal with sunflower meal (with and without enzyme) in laying hens rations on laying performance

TRT	Egg Weight (g)	Production Percent (%)	Mass Production (g of egg/hen/day)	Feed Intake (g/day)	FCR (g feed/g egg)	
<b>Sunflower</b>						
0	59.89	89.45	53.53	101.47 <sup>b</sup>	1.89	
30	61.50	88.95	54.73	100.33 <sup>b</sup>	1.84	
60	61.96	87.50	54.26	103.67 <sup>a</sup>	1.93	
SEM	1.25	0.95	1.26	0.58	0.032	
P Value	0.4803	0.3346	0.7960	0.001	0.1940	
<b>Enzyme</b>						
0	61.66	89.47	55.19	101.60 <sup>ab</sup>	1.85	
100	60.27	87.24	52.53	100.66 <sup>b</sup>	1.92	
200	61.42	89.19	54.79	103.20 <sup>a</sup>	1.89	
SEM	1.25	0.95	1.26	0.59	0.0317	
P Value	0.7062	0.2100	0.2883	0.0145	0.2860	
<b>Sunflower <math>\times</math> Enzyme</b>						
0	0	60.07	91.86	55.18	101.8 <sup>ab</sup>	1.85
0	100	60.43	87.07	52.49	100.6 <sup>ab</sup>	1.92
0	200	59.18	89.43	52.91	102.0 <sup>ab</sup>	1.93
30	0	60.78	90.00	54.77	100.8 <sup>ab</sup>	1.84
30	100	59.04	86.49	51.04	97.8 <sup>b</sup>	1.92
30	200	64.60	90.36	58.37	102.4 <sup>ab</sup>	1.78
60	0	64.05	86.57	55.61	102.2 <sup>ab</sup>	1.87
60	100	61.35	88.14	54.07	103.6 <sup>a</sup>	1.93
60	200	60.49	87.79	53.11	105.2 <sup>a</sup>	1.98
SEM	2.17	1.65	2.18	1.015	0.055	
P Value	0.3767	0.3315	0.3503	0.1886	0.3713	

**Table 2.** The effect of replacing soybean meal with sunflower meal (with and without enzyme) in laying hens rations on egg quality

TRT	Weight (g)	SW (g)	YL (mm)	YH (mm)	YI (mm/mm)	YC	YW (g)	YP (%)	AW (g)	AP (%)	Shell Weight (g)	Shell P (%)	Shell C (g/cm <sup>2</sup> )	AH (mm)	HAUGH
<b>Sunflower</b>															
0	58.81	1.089	14.74	41.19	2.80	3.73	17.08	29.03	35.75 <sup>b</sup>	60.80	5.18	10.17	0.345 <sup>a</sup>	8.42	91.94
30	59.88	1.087	14.69	41.62	2.85	3.73	17.11	28.50	36.75 <sup>ab</sup>	61.69	5.83	9.73	0.335 <sup>b</sup>	8.31	91.05
60	60.07	1.088	14.97	41.69	2.79	3.73	16.99	28.30	37.11 <sup>a</sup>	61.75	5.98	9.95	0.338 <sup>ab</sup>	8.45	91.77
SEM	0.4491	0.00097	0.1445	0.3032	0.035	0.1764	25.09	0.3784	0.3712	0.911	0.1102	0.1656	0.0021	0.0924	0.6037
P Value	0.1138	0.1986	0.3762	0.3130	0.4726	1.0000	0.9432	0.3936	0.0319	0.2010	0.5570	0.1892	0.0131	0.5362	0.5490
<b>Enzyme</b>															
0	59.32	1.088	14.75	41.56	2.82	3.87	17.25	29.04	36.14	60.92	5.95	10.03	0.3357	8.355	91.46
100	59.80	1.088	14.89	41.73	2.80	3.80	17.14	28.67	36.88	61.32	5.93	10.00	0.3404	8.417	91.68
200	59.65	1.087	14.76	41.42	2.81	3.73	16.80	28.19	36.99	62.00	5.85	9.80	0.3421	8.407	91.62
SEM	0.4492	0.00097	0.1445	0.3032	0.0347	0.1784	0.2509	0.3784	0.3813	0.411	0.1106	0.1656	0.0021	0.0324	0.6037
P Value	0.7372	0.6233	0.7413	0.7730	0.9066	0.5777	0.2574	0.2932	0.2888	0.1884	0.6710	0.5823	0.1127	0.8778	0.9666
<b>Sunflower × Enzyme</b>															
0 0	58.43	1.091	15.17	41.61	2.76	3.40	17.43	29.78	35.00	59.94	6.00	10.27	0.344	8.28	91.28
0 100	59.83	1.090	14.69	41.05	2.80	3.60	17.13	28.62	36.52	61.06	6.17	10.32	0.344	8.35	91.32
0 200	58.17	1.088	14.37	40.91	2.85	3.20	16.69	28.69	35.72	61.40	5.76	9.91	0.346	8.63	93.22
30 0	60.05	1.089	14.64	41.66	2.85	4.40	17.47	29.10	36.48	60.75	6.10	10.15	0.331	8.31	91.04
30 100	59.58	1.085	14.76	41.91	2.84	3.60	16.74	28.11	37.26	62.53	5.57	9.35	0.341	8.64	91.86
30 200	60.03	1.087	14.68	41.89	2.85	3.20	17.11	28.51	37.10	61.80	5.82	9.68	0.335	8.18	90.25
60 0	59.47	1.086	14.44	41.41	2.87	3.80	16.79	28.24	36.92	62.08	5.76	9.68	0.332	8.47	92.07
60 100	60.01	1.090	15.23	42.21	2.77	3.60	17.56	29.27	36.24	60.38	6.20	10.34	0.336	8.46	91.84
60 200	60.74	1.087	15.23	41.45	2.73	3.80	16.61	27.38	38.15	62.79	5.97	9.82	0.345	8.41	91.40
SEM	0.7780	0.0017	0.2503	0.2251	0.0601	0.3091	0.4346	0.6559	0.6605	0.7122	0.1916	0.2868	0.0037	0.1601	0.045
P Value	0.757	0.777	0.1381	0.7163	0.4419	0.489	0.4280	0.3316	0.2912	0.1166	0.1178	0.1660	0.2163	0.4373	0.4800

amount of daily feed intake. No significant effects in other performance traits may be related to the amount of high feed intake by high level of sunflower meal replacing. High level of feed intake supply enough amounts of nutrients for egg production, so, reducing in the amount of soybean meal could not negatively affect the performance of laying hens. Increase in the amount of daily feed intake by high amount of multi enzyme application maybe have some reasons such as reducing anti-nutrient contents of meals especially sunflower meal, improve the physically, chemically and microbial population of laying hens digestive tract.

### Egg quality

Sunflower meal contains complex carbohydrates such as pectin, which have groups such as uronic acid and phenolic acid in their structure, known as chelators of calcium, zinc, and magnesium (Wang et al., 2019). This mechanism may have led to reduced calcium availability and thus reduced eggshell crust. As well as adding dietary fat is essential as it accelerates the absorption of pigment and fat-soluble vitamins (Saleh et al., 2021). De Morais Oliveira et al. (2016) indicated that the amount of lipids in the sunflower meal diet

augmented pigment absorption, resulting in improved yolk color. In contrast, Shi et al. (2012) described no positive effect of dietary sunflower meal on the egg yolk color. Aguillón-Páez et al. (2020) noted that the laying hens which receiving sunflower seeds showed significant differences in egg yolk compared with the control group. However, in the present experiment, no significant difference in yolk color was observed, which may be related to the variety of used meals or the experimental poultry. Ceylan, and Cufadar (2018) reported that the effect of enzyme supplementation to laying hens diets containing different levels of sunflower seed meal did not significantly effect on egg quality parameters, which is consistent with the results of this study. As the amount of feed intake increased with replacing high levels of sunflower meal, so the high amount of protein received by hens and transferred to egg and increased the amount of egg albumin.

### CONCLUSION

In conclusion, supplementation of sunflower meal in laying hens diets can reduce production costs in laying poultry rations without negatively impacting production parameters and egg quality.



## CONFLICT OF INTEREST

### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationship that could have appeared to influence the work reported in this paper.

## ETHICS APPROVAL

Protocol (no. 1319-IAU. 01.25.2017) was approved by the experimental animal ethics committee

of Maragheh Islamic Azad University.

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