

Evaluation of Fenugreek (*Trigonella foenum graecum*) Seed Powder and *Moringa oleifera* Leaf Meal on Production Performance and Egg Quality of Laying Hens

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Article

Evaluation of Fenugreek (*Trigonella foenum graecum*) Seed Powder and *Moringa oleifera* Leaf Meal on Production Performance and Egg Quality of Laying Hens

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Abstract: The effect of the addition of fenugreek seed powder and *Moringa oleifera* leaf meal on the production performance and egg quality of Lohmann Brown laying hens was investigated. A total of 108 Lohmann-Brown layers, 48 weeks of age, were used and randomly assigned to six experimental groups of three replicates each in a randomized design. A control diet was supplemented with 3.6 g/hen/day of *Moringa oleifera* leaf meal and fenugreek seed powder alone or in various proportions as follows: control diet (T1), 100% *Moringa oleifera* leaf meal (T2), 25% fenugreek seed powder and 75% *moringa oleifera* leaf meal (T3), 50% fenugreek seed powder and 50% *moringa oleifera* leaf meal (T4), 75% fenugreek seed powder and 25% *moringa oleifera* leaf meal (T5) and 100% fenugreek seed powder (T6) were added on top of the control diet. Data were analyzed using one-way ANOVA by the general linear model method, using Tukey's multiple range tests to differentiate means. According to the results, fenugreek and moringa had a crude protein content of 25.17 and 27.8%, respectively. The groups administered T4 had significantly higher mean feed intake (120.75), feed conversion rate (2.315), egg production (77.22), and egg mass (52.08) than the others. Treatment 4 had a significant ($P<0.05$) improvement in some egg quality traits such as shell weight, shell firmness, albumen weight, yolk weight, yolk color, and Haugh unit compared to the control. In summary, Treatment 4, a mixture of equal parts fenugreek seed powder and *Moringa oleifera* leaf meal, improved egg production performance and egg quality.

Keywords: egg production; egg quality; layers; fenugreek seeds; *Moringa oleifera* leaf

1. Introduction

Chicken production plays an important socioeconomic and nutritional role in the livelihood of rural households in developing countries [1,2]. Among livestock-derived protein source foods, the contribution of the chicken product has been increasing worldwide [3]. The current total number of chickens in Ethiopia is estimated at about 57 million, of which 78.85% are native, 9.11% exotic and

12.03% crossbred chickens [4]. In the countryside, modern poultry production systems are in their infancy, consisting of a small number of large commercial poultry farms and one emerging one "small scale intensive" poultry production system kept for operating on a more commercial basis in urban and peri-urban areas [5]. Feed accounts for 70-75% of the cost of poultry production [6]. According to Negasa [7], Ethiopia lacks self-sufficiency in cereal grains, which form the bulk of the concentrate feeds for poultry feeding. The quality, quantity, and feed cost are the leading obstacles to modern small-scale chicken production in Ethiopia [8]. Therefore, it is necessary to find locally available non-conventional feeds with the potential to improve chicken performance [7]. Fenugreek (*T. foenum-graecum*) and Moringa oleifera leaf meal are among the available non-conventional herbal plants and are used as a supplementary feed additive in chicken production in Ethiopia.

Fenugreek (*Trigonella foenum-graecum*) is an annual herb belonging to the family Leguminosae [9]. Fenugreek seeds have been used as a culinary spice for centuries and as a medicinal product in many traditional / folk medicine systems [10,11]. It is traditionally used as an Ayurvedic medicine for centuries. In recent decades, the therapeutic benefits of fenugreek have been confirmed not only in animal studies but also in human studies [12]. Fresh green leaves are consumed as vegetables, whereas dried seeds are used as a spice in food preparation, and both leaves and seeds are used in flavoring foods [13]. Fenugreek has biologically active substances (protein, amino acids, biogenic elements, lipids, and fatty acids) [14]. Birhane [15] reported that fenugreek seeds contain 29.3% crude protein and other phytochemicals. The seeds are reported to be a natural feed additive and play a role in improving poultry performance [16].

Moringa oleifera Lam. is a fast-growing, drought-resistant tree in tropical and subtropical countries and is a cultivated species of the Moringaceae family [17,18]. It is cultivated throughout India, Africa, Latin America, Mexico, Hawaii, Asia, and Southeast Asia [19]. It is the most promising and perennial multipurpose tree that has been used for nutritional benefits, medicinal properties, environmental protection, and consumption. "Cabbage tree," "horseradish tree," "drumstick tree," "ben oil tree" and "mother's best friend tree" are all names for Moringa oleifera [20]. According to an investigation by Ajantha et al. [19], Moringa oleifera has the potential to be harnessed as animal feed due to its high content of protein, carotenoids, various minerals, and certain phytochemicals [21]. It could be used as a feed additive modulating the production performance of chicken due to its rich source of biologically active ingredients [22]. Despite its high nutritional and phytochemical potential, there is scarce information about its utilization in chicken feeding as a feed additive in the layer ration under feed deficit countries like Ethiopia [23].

On top of the high-cost problem of commercial concentrates [24], synthetic feed additive aimed at improving productivity has been extremely expensive and limited in the local market. Hence, the use of herbal feed additives in poultry nutrition has been gaining considerable attention. A study by Fasuyi [25] reported that supplementation of small quantities of green leaves rich in functional active ingredients (*Moringa oleifera*) [21] and seeds (Fenugreek) [14] on cereal-based poultry feed improves productivity by enhancing nutrient digestion and utilization by the animal.

Although there is a lot of information on the utilization of fenugreek and moringa for chickens, however, there is limited information on the effect of supplementing the combination of fenugreek seed powder and moringa oleifera leaf meal on production performance and egg qualities of Lohmann Brown layers. Therefore, the objective of this study was to evaluate the effect of supplementing a combination of different levels of fenugreek seed powder (FSP) and Moringa oleifera leaf meal (MOLM) on the production performance and egg qualities of Lohmann Brown hens.

2. Materials and Methods

2.1. Description of the Study Areas

The study was conducted at Jimma University, Institute of Technology located at Kito Furdisa Development Enterprise Poultry farm. It lies in the climatic zone locally known as "Weina Dega" (1,500-2,400m above sea level). The study area is characterized by a mean annual maximum and minimum temperature of 30°C and 14°C respectively. The annual rainfall ranges from 1138-to 1690

mm. *Moringa oleifera* leaves were collected from Yayo District. Yayo is a district in southwestern Ethiopia, in the Ilu Aba Bora zone of the Oromia National Regional State. Fenugreek seeds of the “Chala” variety were obtained from Bishoftu Agricultural Research Center.

2.2. Ethical approval and letter of consent

Prior to the start of the experiment, ethical approval was issued from the Ethical Committee of Jimma University College of Agriculture and Veterinary Medicine, Jimma, Ethiopia, and informed consent were also received from co-authors involved in the experiment.

2.3. Collection and Preparation of MOLM and FSP

Adequate quantities of fresh *Moringa oleifera* leaves were collected from the Yayo district, Oromia Regional State. The collected leaves were spread out on a plastic sheet and air-dried under a shade for one week. During the drying process, hands were turned to obtain uniformly dried material. The dried leaves were milled to pass through a 1mm sieve size. Fenugreek seeds of the “Chala” variety were obtained from Bishoftu Agricultural Research Center. The cleaned seeds were soaked in potable water for 12hrs using a seed: water ratio of 1:5 (w/v). The soaking water was intermittently changed every 6 hours. After 12 hours of soaking the water was discarded and the seeds were spread on plastic sheets and allowed to dry. The dried seeds were milled to pass through a 1mm sieve to produce FSP. Both the processed MOLM and FSP were stored in airtight plastic containers until required for laboratory chemical analysis and feeding trial.

2.4. Chemical Analysis

The nutrient compositions of commercial diet, FSP, and MOLM were analyzed in JUCAVM Animal Nutrition Laboratory (Table 1). The wet/fresh leaves of *Moringa oleifera* were weighed using a sensitive balance and dried in an oven at 650c for 72 hrs. A representative sample of the entire test feed materials was grounded to pass through a 1mm sieve size. Dry matter, crude protein, crude fiber, total ash, and ether extract contents were determined according to [26]. Nitrogen was determined by the Kjeldahl procedure and CP was calculated by multiplying N content by 6.25. For the extraction of ether content from the known weight of the powdered sample with petroleum ether (400c to 600c) in a Soxhlet extractor for 4hrs; the residue after evaporation of the solvent was then weighed and the fat content determined. The crude fiber was determined by subjecting the residual powdered sample from ether extraction to successive treatment with boiling 0.25M H₂SO₄ acid and 0.312M NaOH, alkaline. The insoluble residue was washed with hot water until it is base-free. It was then dried to constant weight in an oven at 1050c, cooled in desiccators, and weighed. The weighed sample was incinerated in a furnace at 5500c for 2hrs until a constant weight was obtained. The crude fiber was calculated as the loss in weight of the ash. The organic residue was the crude fiber. The nitrogen-free extract was calculated by difference (% NFE = 100 - % (Moisture + EE + CF + CP + Ash)) according to Henneberg et al. [27]. The metabolizable energy content of the feed ingredients was calculated using the formula suggested by Ponzenga [28]. ME (kcal/kg) = 37*% CP + 81.8*%EE + 35.5*NFE. Where; ME = Metabolizable Energy; CP= Crude protein; EE= Ether Extract, NFE=Nitrogen Free Extract.

Table 1. Nutritional composition and nutrient analysis (% dry matter) of experimental diets.

Dietary ingredients	T1	T2	T3	T4	T5	T6	<i>Moringa oleifera</i> leaf meal	Fenugreek seed powder
Ingredients								
Toasted soybean meal	20	20	20	20	20	20		
Maize grain	49	49	49	49	49	49		
Meat and bone meal	2	2	2	2	2	2		
Noug* seed cake	14	14	14	14	14	14		
Wheat bran	8	8	8	8	8	8		

Vitamin and mineral premix	2	2	2	2	2	2		
Limestone flour	4.5	4.5	4.5	4.5	4.5	4.5		
Salt	0.5	0.5	0.5	0.5	0.5	0.5		
Moringa oleifera leaf meal	0	3.6	2.7	1.8	0.9	0		
Fenugreek seed powder	0	0	0.9	1.8	2.7	3.6		
Nutrient composition								
Dry matter (%)	90	91	90.7	90.6	90.5	90.4	89.4	90.1
Crude protein (%)	17.3	18.5	18.1	17.8	18.2	18.4	27.8	25.16
Ether extract (%)	5	6.8	6.6	6.5	6.9	7	6.4	7.03
Crude fibre (%)	7	7.5	7.3	7.3	7.4	7.6	6.1	7.03
Ash (%)	9.4	10.8	10.4	9.5	9.6	9.7	8.9	3.23
Nitrogen free extract (%)	51.3	60.6	57	58	64	66.6	40.1	47.6
Metabolizable energy (kcal/kg)	2870	3007	2906	2979	3200	3206	2977	3197

* *Guizotia abyssinica*; Vitamin premix per kg of diet: vitamin A: 2.7 mg; vitamin D3: 0.05 mg; vitamin E: 18 mg; vitamin K3: 2 mg; thiamine: 1.8 mg; riboflavin: 6.6 mg; pantothenic acid: 10 mg; pyridoxine: 3 mg; cyanocobalamin: 0.015 mg; niacin: 30 mg; biotin: 0.1 mg; folic acid: 1 mg; choline chloride: 250 mg; antioxidant: 100 mg; Fe: 50 mg; Mn: 100 mg; Zn: 100 mg; Cu: 10 mg; I: 1 mg and Se: 0.2 mg.

2.5. Management of the Experimental Bird

A total of 108 Lohmann Brown layers of 48 weeks of age were used in this study. The layers were divided into 6 groups of 18 birds each. Each group was further subdivided into 3 groups, each with 6 birds. Each group was housed in a separate cage having a dimension of 1m×0.5m×0.5m, (designed to accommodate 6 chickens), equipped with a feeder and watering trough. Before starting the actual experiment, the birds were an adaptation to their experimental diets for two weeks. The levels of moringa oleifera leaf meal and fenugreek seed powder supplemented for experimental layers were based on the suggestion by Tamiru et al. [29] and El-Shafei et al. [30], respectively.

Finally, the six treatment diets shown in Table 2 were randomly assigned to the experimental layers in a completely randomized design with 3 replications. Birds were fed with the experimental ration at 120g/h/d. Weighed quantities of feed were offered twice a day and refusals were always collected and weighed in the morning of the following day before the feed is offered. Fresh and clean water was made available all the time. Daily feed intakes per group were determined by subtracting the feed refused from the feed offered. Daily mean feed consumption, hen-day egg production, and feed conversion ratio were recorded weekly. Egg qualities were measured every week.

Table 2. Treatment allocation to the experimental layers.

Treatments	Birds/ treat	Rep/ treat.	Birds/rep.
Commercial layer's ration+0g/h/d, control treatment (T1)	18	3	6
Commercial layer's ration + 3.6 g/h/d (100% MOLM and 0 % FSP (T2)	18	3	6
Commercial layer's ration + 3.6g/h/d (25%FSP &75% MOLM (T3)	18	3	6
Commercial layer's ration + 3.6g/h/d (50%FSP & 50% MOLM (T4)	18	3	6
Commercial layer's ration + 3.6g/h/d (75% FSP & 25% MOLM (T5)	18	3	6
Commercial layer's ration + 3.6 g/h/d (100% FSP and 0% MLOM (T6)	18	3	6
Total	108		

Treat = treatment, rep = replication, FSP= Fenugreek Seed Powder, MOLM= Moringa oleifera leave Meal, T1, T2, T3, T4, T5 and T6 = Treatment, ME=Metabolizable Energy.

2.6. Data Collection

2.6.1. Production Performance

Eggs were collected daily and subjected to quality evaluation on weekly basis. The collected eggs from each treatment group were individually weighed followed by the determination of total egg production (%) and average egg weight (gram) using a sensitive electronic balance. Daily feed intakes per group were determined by subtracting the feed refused from the feed offered. Total egg mass was computed by multiplying the mean egg weight by the total number of eggs produced. Daily egg mass per hen was computed by dividing the total egg mass by the number of hens that were initially housed and the total number of days in which the hens were in-laid. The feed conversion ratio was determined by dividing the mean weekly feed consumed by the mean weekly egg mass. Feed conversion ratio (feed consumed/egg mass) was then calculated as grams of feed: grams of egg mass output [31].

2.6.2. Measurement of Egg Quality

For external and internal egg qualities evaluation, six eggs were randomly selected from each treatment group on weekly basis (2 eggs per replicate) and a total of 36 eggs were picked every week on a random basis, marked individually, weighed, and broken to determine for determination of external and internal egg quality characteristics. The specific gravity of eggs was determined according to the procedure described by Olsson, [32], where eggs were immersed in salt solutions with densities varying from 1.070 to 1.090 with an interval of 0.005. Egg weight was measured with a sensitive laboratory balance with accuracy of 0.01g. The length and width of each egg were measured using digital calipers. The width was measured as the distance between the two ends of an egg at the wider cross-sectional region using digital calipers. The egg's length was measured as the distance between the broad and narrow ends of the egg. The egg shape index was calculated as the percentage of the egg breadth/broad end (width) to the egg length. Shell weight and shell strength was evaluated by using an egg force reader (EFR-01, Orka Food Technology, West Bountiful, UT, USA), shell thickness was measured on the large end, equatorial region and small end, respectively, by using eggshell thickness gauge (Robotmation Co.Köyliö, Finland) and the average value was measured as the eggshell thickness measurement.

Albumen and yolk index were calculated as a percentage, taking the ratio of their respective heights of the average or mean of breadth and length. An albumen and yolk ratio were calculated taking their weights as the percentage of total egg weight. The diameter of the Yolk was calculated as the average of yolk length and breadth. The yolk albumen ratio was calculated as the weight of yolk /weight of albumen. Egg yolk colour was evaluated by Roche colour fan (Hoffman-La Roche Ltd., Basel, Switzerland). Height of albumin and yolk were measured with an electronic digital caliper (SH14100025; Shenhan, Shanghai, China), albumin weight and yolk weight were evaluated. Haugh unit was calculated according to formula developed by Haugh [33].

2.7. Statistical analysis

All the data collected were subjected to analysis of variance (ANOVA) according to Completely Randomized Designs consisting of six treatments with 3 replications using the General Linear Models (GLM) procedure of Statistical Analysis System (SAS, version 9.3, 2014). When significant differences were observed, treatment means were compared using Tukey multiple comparison test. All statements of statistical differences were based on $p < 0.05$.

The model used for bird's performance and egg quality affected by treatment:

Where; Y_{ij} = the response variables;

μ = overall mean;

t_i = treatment effect and

B_{ij} = random error

3. Results and Discussion

3.1. Proximate Composition of FSP and MOLM

Results obtained from laboratory chemical composition analysis of commercial feed, Fenugreek seed powder (FSP), and Moringa oleifera leaf meal (MOLM) were presented in Table 1. The results of this study indicated that FSP and MOLM contained 25.16 and 27.8% crude protein, respectively. Sulieman et al. [34] and Etalem et al. [35] reported crude protein content of 24-26% and 28% for FSP and MOLM, respectively, which is in agreement with the results of the present study. The crude protein content of FSP falls within the range of 21.7 to 29.0% reported by El Nasri and El Tinay [36] and of MOLM with 22.99 to 29.36% reported by Sultan [37], respectively.

The crude fiber content of FSP and MOLM was 7.03 and 6.14% respectively (Table 1). The crude fiber content of FSP and MOLM in the present study was in line with the report of Birhane [15] and Su and Chen [38] who reported crude fiber content of 7% and 6.13% for FSP and MOLM respectively. In addition, Yassin et al [2] and Sultan [37] noted that FSP and MOLM contained crude fiber of 7.4% and 6-9.6%, respectively. In contrast to the current finding, Wu et al. [39] reported a lower crude fiber of moringa oleifera leaf (5.9%). The low fiber content in MOLM compared to FSP would tend to imply good palatability and nutrient digestibility when added to commercial diets in poultry

The result of the lipid content of 7.03% and 6.44% was recorded for FSP and MOLM respectively as measured by the percentage composition of ether extract. The higher level of ether extract in FSP compared to MOLM indicates that FSP would serve as a good media to dissolve fat-soluble vitamins and hence enhance absorption of them and improve the palatability of the basal diets. The result of the ether extract obtained in the current study agreed with that of Yassin et al [2] and Busani et al [40] who reported ether extract content of 7.03% and 6.5% for FSP and MOLM respectively.

The total ash content recorded for both feed ingredients in the present study was in agreement with that of El Nasri and El Tinay [36] and Busani et al [40] who reported total ash content of 3.3% and 7.64% for FSP and MOLM, respectively. The total ash content of FSP and MOLM were 3.26-3.52% and 7.6-12 % reported by Hadi and Abed [41] and Su and Chen [38], respectively.

The nitrogen-free extract of feed ingredients is assumed to represent the readily digestible components of carbohydrates. The NFE content obtained from FSP was 47.63% which was higher than that of MOLM contained (40.06%), indicating that FSP is higher in carbohydrates than MOLM. The NFE levels identified in two tested diets were concurrent with Sulieman et al. [34] and Samia et al. [42] who reported NFE content of 47.5% for FSP and 36.28 to 43.90% for MOLM. The calculated metabolizable energy (ME) of FSP and MOLM was 3197 Kcal/ kg of DM and 2977 Kcal/kg of DM respectively (Table 1). The result obtained from the current study was comparable with the report of Yassin et al [2] and Garcia et al. [43] who reported ME content of 3435Kcal/Kg of DM for FSP and 3000Kcal/Kg of DM for MOLM, respectively. The difference in proximate composition of FSP and MOLM might be due to the variation in species, genetics, climate, and edaphic and agronomic activities [42,44].

3.2. Production Performance of the Experimental Layers

Throughout the experimental period, no mortalities were recorded, and the birds didn't show any clinical signs. Supplementation of a combination of different levels of the FSP and MOLM on layer hens was a statistically ($p < 0.05$) different among the treatment groups.

3.2.1. Feed Intake

The results of the mean daily feed intake of the experimental layers in the control group and supplemented groups with a combination of different levels of fenugreek seed powder and moringa oleifera leaf meal were presented in Table 3. Treatment groups supplemented with different proportions of FSP and MOLM had significantly improved ($p < 0.05$) feed intake than the control group. However, there was no significant difference ($p > 0.05$) among the supplemented groups. The improvement in feed intake might be explained due to the presence of a wide range of bioactive components in FSP Abdel-Wareth et al [41] and MOLM Moreno-Mendoza et al [46] which can

increase nutrient digestion and utilization. In agreement with this finding, Samani et al. [47] reported that supplementation of fenugreek powder to a high level of 2% in a laying hen's diet resulted in an increased feed intake. Similarly, the addition of 3g/kg Fenugreek seed Alloui et al [44] and up to 5% of MOLM Olugbemi et al [45] improved feed intake in broiler chicken. A synergic effect on the improvement of feed intake in broilers was achieved when incorporating black cumin, turmeric, and fenugreek at the level of 1 and 2 g kg in broiler chicken Yesuf et al [46]. In contrary with our results, N'nanle et al [51] reported that chicken fed on a diet containing 1% MOL shows a significant reduction in feed consumption compared to the control group.

Table 3. Production performance of the experimental layers fed on the combination of different levels of FSP and MOLM.

Parameters	T1	T2	T3	T4	T5	T6	SEM	P-Value
FI (g)	115.35 ^b	119.49 ^a	120.05 ^a	120.75 ^a	119.95 ^a	119.65 ^a	0.56	0.0016
HDEP	70.37 ^d	73.83 ^{bc}	75.31 ^{ab}	77.2 ^a	74.14 ^{bc}	71.85 ^c	0.341	<.0001
EM	45.1 ^c	47.83 ^b	49.1 ^b	52.1 ^a	48.64 ^b	47.91 ^b	0.296	<.0001
FCR	2.56 ^a	2.49 ^{ab}	2.45 ^b	2.32 ^c	2.46 ^b	2.49 ^{ab}	0.54	<.0001

^{a b c d}: Mean within the same row different superscripts are significantly different ($P < 0.05$). FSP= Fenugreek Seed Powder, MOLM= Moringa Oleifera leaf Meal, FI= Feed Intake, HDEP= Hen Day Egg Production, EM= Egg Mass FCR= Feed Conversion Ratio, SEM= Standard Error of Mean, P-Value= Probability Value.

3.2.2. Feed Conversion Ratio

The results of the feed conversion ratio of the experimental layers in the control group and supplemented groups with a combination of different levels of fenugreek seed powder and Moringa oleifera leaf meal were presented in Table 3. Accordingly, all the supplemented groups had an improvement in FCR compared to the control. An equal proportion of fenugreek seed powder and Moringa oleifera leaf meal (T4) showed a significantly ($p < 0.05$) higher feed conversion ratio. The highest improvement in FCR, ascribing to the synergetic effect of bioactive components in both test diets [48,52]. In line with our finding, Abdel-Wareth and Lohakare [53] and Voemesse et al [54] reported that supplementation of Moringa oleifera leaves meal to egg-laying hens diets significantly improved the feed conversion ratio compared to the control group. Additionally, Paneru et al [55] reported that supplementation of FSP to broiler chicken diets significantly ($p < 0.001$) increased FCR. Moreover, N'nanle et al [51] and Ashour et al [56] reported that chicken fed on a diet containing 1% MOL showed a significant reduction in FCR and dietary supplementation of Moringa oleifera seeds did not significantly influence FCR. The discrepancy in the results of these findings might be due to the use of different chicken genotypes, age, sex, production stage, altitudinal variation, herb chemical composition, and agronomic practices. In addition, a mixture of black cumin, moringa, and chicory seeds resulted in a synergetic effect on the improvement of FCR in the broiler compared to the control group [48].

3.2.3. Egg Production Performance

The results of the mean egg production and egg mass of the experimental layers in the control group and supplemented groups with a combination of different levels of fenugreek seed powder and Moringa oleifera leaf meal were presented in Table 3. The results revealed that the supplemented groups had significantly improved egg production than the control group. The mean egg production and egg mass of T4 was significantly higher than other supplemented groups and control, indicating that an equal proportion of Moringa oleifera leaves and fenugreek seeds had a synergetic effect on egg production performance due to an optimum modulation of active ingredients on digestive efficiency of major nutrients [10,58].

This could be supported by the report of previous active ingredient analysis of each tested diet, though only proximate data was generated in this study to claim justification of observed responses.

Although the positive individual effect of Fenugreek seed by Abdalla et al. [59] and Awadein et al. [60] and Moringa oleifera leaf by Abdel-Wareth and Lohakare [53] on a comparable range of egg production and egg mass was previously reported, the current study was corroborated with the profound synergetic effect on egg production and egg mass when 1g/kg fenugreek seeds and 1g/kg flax seeds were added on top of the control diet [61].

3.3. Eggs Quality Characteristics

3.3.1. External Eggs Qualities

The results of external egg qualities of the experimental layers in the control group and supplemented groups with a combination of different levels of fenugreek seed powder and Moringa oleifera leaf meal were presented in Table 4. The results showed that the mean egg-specific gravity, egg weight, and egg shape index of all supplemented groups was significantly ($p < 0.05$) higher compared to the control group. The result of the current study was in line with that of El-Shafei et al [30] who reported that there was a significant increase in egg weight and egg-specific gravity by adding 2% fenugreek powder compared with the control group. The result of the present study was also in agreement with Jean et al. [62] who noted that a significant effect on egg weight was achieved in 5% MOLM diet-fed birds. The significant increase in the egg weight of the present study might be due to the presence of lysine and methionine in the Moringa oleifera leaf. Supplementation of lysine and methionine in the chicken diet will increase egg mass output by 26% [63], significant improvement in egg weight and egg mass [64], and promote higher egg weight eggs [65]. The higher egg-specific gravity observed in this study might be due to the thickness of an eggshell laid by supplemented experimental layers. According to Olugbemi et al. [66], shell thickness is a function of calcium and phosphorus levels in a layer's ration and is also an indication of the specific gravity.

Supplementation of fenugreek seed powder and Moringa oleifera had no significant ($p > 0.05$) effect on mean egg length and egg width (Table 4). Following the result of our study, supplementation of fenugreek seed powder and Moringa oleifera showed a non-significant effect on egg length and egg width [60,67,68]. Similarly, Sharmin et al [69] reported that feeding Moringa oleifera leaf meal up to 1.5% had no significant difference in egg length and egg width. The present study indicated that egg shape index was not affected by FSP and MOLM supplementation, indicating that the proximate and active components of either of the tested diets might not have modulated egg shape index which could have majorly attributed to the genetic basis of chicken. A prior study confirmed the insignificant effect of each tested diet on the egg-shape index of layer chicken and Japanese quail [21,35,47,68].

Significantly ($p < 0.05$) higher eggshell weight was recorded from the chicken feed on diet containing FSP and MOLM compared to the control group. Under our results, the previous finding indicated that improvement in eggshell weight was attained due to the supplementation of a mixture of fenugreek seeds, linseeds, garlic, and copper sulphate [70]. Similarly, MOLM supplementation at levels of 1, 1.5, and 2% was proven to bring a considerable improvement in eggshell weight [23,71]. The increment of eggshell thickness in chicken that received FSP and MOLM could be due to the rich source of calcium and phosphorus in the tested diets. Contrary to our findings, Awadein et al [60] and Wahab et al [72] reported that supplementation of fenugreek up to 0.5% in the ration of laying hens had no significant influence on eggshell weight. The inconsistency of shell weight among these results could be due to the synergetic effect of MOLM and FSP in our result and the sole inclusion of FSP in layer ration in the previous report [60,72].

According to the result obtained in the current study, the mean eggshell thickness and shell strength of chicken-supplemented MOLM and FSP was significantly ($p < 0.05$) higher than the control group. The mean eggshell thickness of the groups assigned to the T4 was significantly ($p < 0.05$) higher compared to the control (T1) and all other supplemented groups. According to the present study result, the supplementation of MOLM (T2), FSP (T6), and their combination in T3, T4, and T5 improved the eggshell thickness and shell strength of Lohmann Brown layers. The results of the current study were in agreement with that of Park et al. [73] who reported that supplementation of fenugreek seed on Hy line-brown laying hens significantly ($p < 0.05$) increased the eggshell breaking

strength and eggshell thickness compared with the control group. Similarly, the result of Sharmin et al [69] indicated that the addition of MOLM to the hens' diet for a longer period improves shell-breaking strength. Supplementation in commercial diets of 4-6% Moringa oleifera leaf powder increased the eggshell strength and shell thickness in laying hens Siti et al [74]. The higher eggshell thickness and strength observed in this study might be due to the quality of the eggshell laid by the supplemented experimental layers. Good shell thickness is an important bio-economic trait in commercial egg production as it may help to reduce the percentage of cracked eggs and decrease the rate of loss for producers [23].

Table 4. Effect of supplementing a combination of FSP and MOLM on external egg qualities.

Parameters	T1	T2	T3	T4	T5	T6	SEM	P.Value
SG	1.076 ^b	1.085 ^a	1.085 ^a	1.09 ^a	1.084 ^a	1.086 ^a	0.95	0.0003
EWt (g)	60.84 ^b	64.71 ^a	66.8 ^a	67.64 ^a	65.84 ^a	65.92 ^a	0.59	0.0004
EW (cm)	4.489	4.43	4.63	4.56	4.478	4.466	0.057	0.4245
EL (cm)	5.84	5.66	5.81	5.94	5.81	5.94	0.095	0.5844
ESI	76.83	78.32	80.03	76.78	77.16	75.26	1.69	0.6973
SW (g)	6.05 ^b	6.46 ^a	6.63 ^a	6.75 ^a	6.56 ^a	6.57 ^a	0.063	0.0007
SS (kg)	3.013 ^b	3.70 ^a	3.65 ^a	3.85 ^a	3.63 ^a	3.71 ^a	0.102	0.0077
ST (mm)	0.275 ^c	0.353 ^b	0.346 ^b	0.379 ^a	0.345 ^b	0.335 ^b	0.044	<.0001

^{a b c d}: Mean within the same row different superscripts are significantly different ($P < 0.05$). FSP= Fenugreek Seed Powder, MOLM= Moringa Oleifera Leaf Meal, SG = Specific Gravity, EWt = Egg Weight, EW = Egg Width, EL = Egg Length, ESI = Egg Shape Index, SW = Shell Weight, SS = Shell Strength, ST = Shell Thickness, SEM= Standard Error of Mean, P- Value= Probability Value.

3.3.2. Internal Eggs Qualities

Table 5 shows the effects of dietary Moringa oleifera leaves and FSP supplementation on internal egg quality. The mean albumen height obtained from the groups fed on the T4 and T5 was significantly ($p < 0.05$) higher compared to that of the control group. However, there was no significant ($p > 0.05$) difference among the chicken-supplemented experimental diet. The results of the current study were in line with Abou-Elezz et al. [75] and Sharmin et al [69] reported that supplementation of the Moringa oleifera leaf meal had increased the value of albumen height. Similarly, Wubalem et al. [23], reported that there was a statistically significant ($p < 0.05$) difference between all the groups placed on the treatment ratio containing different levels of MOLM compared to control groups in mean albumen height. On contrary, Etalem et al. [35] noted that there was no significant difference in albumen height fed on different amounts of the dried Moringa oleifera leaf meal. Moreover, Wahab et al [72] reported that supplementation of fenugreek seed up to 1% in the ration of Rhode Island Red spent layers did not indicate any significant influence on albumin height. The albumen height observed in this study might be due to the content of protein and amino acid found in fenugreek seed powder and Moringa oleifera leaf meal. Gunawardana [76] reported that the albumen quality increases with increasing dietary protein levels.

Supplementation of MOLM and FSP did not significantly ($p > 0.05$) influence the mean yolk height and yolk index. The results of the current study were in agreement with Williams [77] who reported that supplementation of fenugreek feed additive had no significant effect on yolk height. Similarly, Samani et al. [47] reported that the inclusion of fenugreek in laying hens' diets had no significant effect on the egg yolk index compared to the control groups. By our finding Tesfaye et al [35] reported that there was no significant ($P > 0.05$) difference in the egg yolk index in Dominant Cz layers supplemented cassava root chips and MOLM. Additionally, Sharmin et al [69] reported that supplementation of MOLM in egg-laying hens' diet did not significantly influence yolk height and yolk index.

The results indicated that the albumen weight and yolk weight of all supplemented groups were significantly ($p < 0.05$) different compared to the control group. However, there was no significant ($p > 0.05$) difference in mean albumin weight and yolk weight among chicken fed on a diet

supplemented with FSP and MOLM. The result from the current study indicated that supplementation of FSP and MOLM significantly improve the mean albumen weight and yolk weight. The current results were in line with Awadein et al. [60] who reported that supplementation of fenugreek at a level of 0.1 and 0.5% in the ratio of laying hen had significantly increased egg albumen weight. El-Sheikh et al. [71], reported that there was a significant ($p < 0.05$) improvement in mean albumen weight as a result of the inclusion of 1.5-2% of supplementation of Moringa oleifera leaf powder. Moringa oleifera leaf powder increases yolk weight in the layer diet of chickens at 2.5% and 5%, compared to the control diet [68]. In the current study, increasing yolk weight was the main reason for the increment in albumen weight and this might be explained as an increase in albumen weight in the groups fed a diet supplemented with a combination of fenugreek seed powder and Moringa oleifera leaf meal. Increasing yolk weight was the main reason for the increment in albumen weight. In contrary with our result, Omri et al [78] reported that dietary treatment of grounded fenugreek seed (2%) did not show a significant difference in albumen weight and yolk weight. Additionally, Omari et al [70] also reported that the dietary addition of 2.9 kg of fenugreek seed didn't affect the albumen weight and yolk weight of egg-laying hens.

The mean haugh unit of birds that received a diet containing FSP and MOLM (except T6) was significantly higher compared to the control group (T1). The mean haugh unit of birds allocated to the T6 experimental ration was not significantly different with a control group and other treatments. Improved haugh units were recorded from birds fed on treatment ration containing MOLM (T2), fenugreek seed (T6), and a combination of fenugreek seed powder and Moringa oleifera leaf meal (T3, T4, and T5). Haugh unit ranging between 66.562 and 82.012 was attained which might be due to the existence of essential nutrients found in the FSP and MOLM. The result of the current study was in agreement with that of Awadein et al. [60] who reported that supplementation of fenugreek in the ratio of laying hen was significantly increased haugh unit. El-Shafei et al. [30] reported that supplementation of 2% fenugreek powder had increased in the haugh unit compared with the control group. Lu et al. [79] reported that there was a significant improvement in the haugh unit as a result of supplementation of Moringa oleifera leaf into the layer's ration. In addition, Abdel-Wareth and Lohakare [53] suggested that the haugh unit significantly improved as a response to diets supplemented with 3, 6 and 9 g/kg Moringa oleifera leaves on Hy-Line Brown hens. Moreover, Park et al [73] reported that there was no significant difference in Haugh units of laying hens fed different levels of fenugreek seed extract (FSE).

Significantly ($p < 0.05$) higher mean value of yolk color was recorded from layers received T2, T3, T4, and T5 compared to T6 and the control group (T1). Birds that received T6 were significantly ($p < 0.05$) higher mean value of yolk color compared to T1. However, there was no significant ($p > 0.05$) difference among T2, T3, T4, and T5. Accordingly, supplementation of FSP and MOLM to layer ration in the current experiment had a significance influence on yolk color. The mean yolk color score of the eggs collected from the supplemented groups had got yellow color. The result of the current study was in line with El-Shafei et al. [30] who reported that the supplementation of 1% fenugreek powder had increased yolk color compared with the control group. Dietary supplementation of Moringa oleifera at 1.5 and 5% in egg-laying hens improved yolk color [69,79]. The improvement in yolk color observed in this study might be due to the presence of carotenoids and xanthophylls in FSP and MOLM. Berkhoff et al. [80] noted that yolk color is a key factor in any consumer survey relating to egg quality.

Table 5. Effect of supplementing a combination of FSP and MOLM on Internal egg qualities.

Parameters	T1	T2	T3	T4	T5	T6	SEM	P.Value
AH (mm)	3.64 ^b	5.93 ^{ab}	6.21 ^{ab}	6.6 ^a	6.506 ^a	5.26 ^{ab}	0.48	0.0325
YH (mm)	16.32	16.24	16.01	16.87	15.89	15.73	0.33	0.476
AW (g)	36.56 ^b	38.86 ^a	40.13 ^a	40.62 ^a	39.57 ^a	39.61 ^a	0.35	0.0003
YW (g)	18.23 ^b	19.4 ^a	20.01 ^a	20.3 ^a	19.70 ^a	19.73 ^a	0.18	0.0005
YC	3.53 ^c	8.03 ^a	7.81 ^a	8.83 ^a	7.61 ^a	5.61 ^b	0.29	<.0001
YI	41.82	39.87	40.2	39.3	41.3	39.02	1.01	0.5744

HU	53.54 ^b	73.4 ^a	75.3 ^a	82.01 ^a	77.32 ^a	66.6 ^{ab}	3.25	0.0040
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^{a b c d}: Mean within the same row different superscripts are significantly different (P<0.05). FSP= Fenugreek Seed Powder, MOLM= Moringa Oleifera Leaf Meal, AH= Albumen Height, YH= Yolk Height, AW= Albumen Weight, YW= Yolk Weight, YC= Yolk Color, YI= Yolk Index, HU= Haugh Unit, SEM= Standard Error of Mean, P- Value= Probability Value.

4. Conclusions

Supplementation of a combination of equal parts of fenugreek seed powder and moringa oleifera leaf meal improved the overall egg production performance and egg quality of layers hens.

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