



Effect of Low Protein Diets Balanced with Limiting Amino Acids on Production Efficiency and Egg Characteristics in Swarnadhara Layers

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Abstract

The effect of reducing crude protein (CP) with supplementation of limiting amino acids was evaluated in an experimental trial using Swarnadhara layers. Egg production parameters and egg quality were studied in 240 layers in a completely randomized design in 5 treatments with 6 replications of 8 birds in each. ICAR 2013 specifications for improved native chicken breeds were followed for formulating the control diet (T1) which was based on corn soya bean meal with 16% CP and 2700 kcal ME. The diets T2 to T5 were made isocaloric to control and composed with 15.5, 15, 14.5 and 14% CP respectively, ensuring the levels of limiting amino acids namely methionine lysine, threonine as that of control by adding synthetic preparation. The data on egg production parameters are presented on the basis of hen day egg number, feed efficiency on a dozen egg basis, and period-wise egg weight (EW) during 29- 40 weeks remained statistically non-significant among various dietary groups. The values for eggshell thickness recorded in CP reduced groups (T3, T4, and T5) were comparable with control (T1) except T2 and also comparable among themselves. The mean shape index, albumen index, yolk index, and Haugh Unit score did not show any significant difference among various dietary treatments. In summary, the crude protein content of the Swarnadhara layer diet can be reduced to 14% by limiting AA fortification without any adverse effect on egg production and egg quality during 29-40 weeks of age.

Keywords: Dietary crude protein, Egg production, Egg quality, Egg weight, Feed efficiency, Limiting amino acids

Introduction

It is generally known that providing high protein to animals reduces efficiency of protein utilization and increases heat production which is a major cause of heat stress in tropical zone. The recent trend in feed formulation is to limit CP consumption by adding synthetic amino acids so that the production performance is not altered while reducing the metabolic load of the bird along with feed cost. Various studies on dietary crude protein have revealed that the undigested portion of crude protein results in volatilization and wasting as litter and the unionized portion causes pollution in the room environment. Meluzzi *et al.* (2001) observed that fecal nitrogen content in a layer trial decreased with a reduction in dietary protein content and was about 50 % of the intake. These types of research have proved to mitigate the impacts of environmental pollution raised from commercial poultry rearing. According to reports by Bunchasak and Silapasorn (2005), the ammonia levels when elevated will result in increased respiratory stress for both birds as well as attendants. Many studies on animal nutrition reveal that to reduce the metabolic stress in layers, individual amino acid requirements have to be met in the precise form while formulating the diets. Though there are numerous studies regarding reduced CP levels in commercial layer diets, the information regarding the supplementation of amino acids in low CP diets in Indian native breeds is scanty. Swarnadhara is a backyard poultry strain. This was developed and released by Veterinary College, KVAFSU, Bengaluru, and this study was undertaken to assess the performance of Swarnadhara layers fed low CP diet fortified with limiting amino acids.

Materials and Methods

The experiment was conducted in the Department of Poultry Science, Veterinary College, Bangalore. Two hundred and forty Swarnadhara layers of 20 weeks of age were distributed in a completely randomized design into five treatments with six replications of eight birds after weighing. The experimental period was from 29 weeks to 40 weeks divided into three cycles of 28 days each. 17 hours of lighting was provided during the laying period.

Diet Formulation

The experimental diet was formulated as per the nutrient requirements specified by ICAR 2013 recommendations for improved native chicken breeds of India. The feed ingredients were analyzed for the amino acid composition and proximate composition. The treatments included a control diet (T1) formulated on corn soya meal (16% CP and 2700 kcal ME) and was satisfying the ICAR 2013 requirements of the first four limiting amino acids viz. Methionine-0.35%, Lysine-0.75%, Threonine-0.52%, Tryptophan-0.16%. The other treatment diets (T2 to T5) were made by reducing CP contents (15.5, 15, 14.5 and 14 % respectively). These diets met the amino acid levels as that of control by adding synthetic amino acids. The treatment diets and control were isocaloric. Ingredient composition and analyzed nutrient composition of layer diets are presented in Tables 1 & 2, respectively. Throughout the experimental period, all the layers were fed with a fixed amount of feed to avoid excess fat deposition in the body. The eggs were numbered according to replicates before collection and daily production was recorded. Egg production was estimated for three periods with 28 days each (29-40 weeks of birds' age) in various treatments for further statistical analysis. Period-wise feed efficiency values (kg feed/dozen eggs) were calculated and the data were interpreted accordingly. The eggs produced in each replicate group were weighed at the end of each 28-day experimental period on two consequent days and data was analyzed. At the end of 32, 36, and 40 weeks of age, 60 eggs in total (2 eggs/replication) were collected from the experimental birds and were subjected to a study of different egg quality characteristics. The length and breadth of eggs were measured to find out the average shape index of eggs using a Vernier Calipers. The height of egg white and egg yolk was measured using Ames Haugh Unit Spherometer and diameter by Vernier Calipers. The albumin index and yolk index were calculated by applying the standard mathematical formula in different groups at every 28-day interval. The shell pieces at the broad end and narrow end without shell membranes were kept separated and their thickness was measured using Micrometer. Haugh unit (HU) score (Haugh, 1937) was calculated using the formula, $HU = 100 \times \log (H + 7.57 - 1.7 \times W^{0.37})$ Where H= Albumen height (mm) and W= Egg weight (g). The values were arranged according to treatments under each 28-day period.

Statistical Analysis

Data were analyzed in a completely randomized design using the general linear model ANOVA. The procedures described by Snedecor and Cochran (1989) were followed in the analysis. In all cases, significance was set at $P < 0.05$.

Table 1: Ingredient composition of Layer diets

Ingredients	Treatments				
	T1(Kg)	T2(Kg)	T3(Kg)	T4(Kg)	T5(Kg)
Maize	65.73	66.16	66.26	66.25	66.55
DORB	-	1.21	2.5	4.32	5.5
Soya	22.55	20.9	19.5	17.7	16.2
Shell grit	7.0	7.0	7.0	7.0	7.0
Mineral Mixture*	3.0	3.0	3.0	3.0	3.0
DCP	0.5	0.5	0.5	0.5	0.5
Salt	0.3	0.3	0.3	0.3	0.3
Lysine	0.1	0.13	0.13	0.13	0.13
Methionine	0.1	0.12	0.13	0.14	0.16
Threonine	0.31	0.3	0.3	0.28	0.28
Additives **	3.17	3.17	3.17	3.17	3.17
Total	100	100	100	100	100

DORB- Deoiled Rice Bran, DCP-Di calcium Phosphate

*Mineral Mixture: Each 100g Contains Calcium-32g, Phosphorus-6g, Copper-100 ppm, Cobalt-60 ppm, Manganese-2700 ppm, Iodine 100 ppm, Zinc-60ppm, Iron-120ppm

**Additives: Vitamin AB2D3- 25 g (each gram contains-Vit A-82500 IU, B12-50mg, D3-12,000 IU and K-10mg.), Vitamin B Complex with E-25 g (each gram contains Vit B1-4mg, B6-8mg, B12-40mcg, E-40mg. Calcium D pantothenate-40 mg and Niacin-60mg.) Herbal Liver stimulant-100g; Semduramicin- 30.0g, Tetracyclin- 30.0g, a commercial Toxin binder-100g

Table 2: Analyzed nutrient composition of Layer trial diets

Nutrients %	Treatments				
	T1	T2	T3	T4	T5
CP	15.998	15.525	15.056	14.999	14.034
Ca	3.611	3.607	3.604	3.599	3.596
P ava	0.466	0.467	0.468	0.470	0.471
P total	0.589	0.595	0.602	0.612	0.621
Fat	2.839	2.839	2.837	2.828	2.820
Crude Fibre	3.006	3.137	3.277	3.475	3.640
ME(Kcal/Kg)	2712	2711	2709	2704	2699

Results and Discussion

The effect of reducing CP with balanced LAA (Limiting Amino Acids) on egg production, feed efficiency and egg weight in Swarnadhara layers is presented in Table 3.

Table 3: The effect of reducing CP with balanced LAA on Egg production, Feed efficiency and Egg weight

Parameters	T1	T2	T3	T4	T5
HD No	CP-16%	CP-15.5%	CP-15%	CP-14.5%	CP-14%
P1 ^(NS)	19.08±0.57	18.67±0.57	19.25±0.5	17.44±0.95	17.83±1.31
P2 ^(NS)	21.06±0.43	19.73±0.46	20.71±0.59	19.62±1.01	19.67±1.18
P3 ^(NS)	18.77±0.60	19.04±0.53	19.12±0.76	18.98±0.75	18.10±1.09
Total ^(NS)	56.06 ±0.26	53.84±0.31	56.96±0.54	53.98±2.06	53.69± 1.09
FCR/ DOZ					
P1 ^(NS)	2.65± 0.08	2.71± 0.08	2.62± 0.07	2.80± 0.17	2.87± 0.23
P2 ^(NS)	2.4 ±0.05	2.56± 0.06	2.44± 0.07	2.6 ±0.14	2.61 ±0.19
P3 ^(NS)	2.7 ±0.09	2.66 ±0.07	2.66 ±0.1	2.68 ±0.1	2.83 ±0.17
EW (g)					
32 weeks ^(NS)	58.63 ±0.86	58.22± 0.77	57.60± 0.81	57.03± 0.74	57.60± 0.71
36 weeks ^(NS)	60.92± 0.87	60.84± 0.88	59.18± 0.91	60.84 ±0.73	59.76± 0.62
40 weeks ^(NS)	62.58± 0.94	63.73±0.84	61.97±0.72	62.79±0.97	61.52± 0.72

NS-Non Significant

As per the analysis, the differences in egg production and feed efficiency between treatments were found to be statistically non-significant ($P \geq 0.05$) during all the periods. The highest production was recorded in the 1% CP reduced group and numerically lower values were obtained with more CP reduction in the first period. The findings on egg production are in close agreement with studies on layers by Ji *et al.* (2014) and Torki *et al.* (2015). In these trials, LAA balanced low CP diets showed non-significant differences in the egg-laying rate and egg number supporting the theory of meeting the amino acid requirement rather than CP *per se*. Junqueira *et al.* (2006) reported that in an 8 weeks experiment, the performance of laying hens in the second laying cycle was comparable between 16 and 20 % CP diet. Contrary to the present results, Rojas *et al.* (2015) also reported that even though the diets were supplemented with several essential AA, the reduction in dietary protein levels from 15 % to 13 % negatively influenced ($p < 0.05$) egg production, feed conversion, and egg mass. He concluded that deficiencies of other AA besides those supplemented in the diet affected the performance of experimental birds because the individual and combined supplementation of Isoleucine, Tryptophan, and Threonine in the 13 % CP diet restored performance to the same level as the control diet in his experiment. Torki *et al.* (2015) observed when the CP reduction is too high (from 16.5 % to 10.5 %) egg production and egg mass were significantly reduced even though all the EAA were balanced. In the present study with Swarnadhara layers using low CP balanced with LAA, the statistical analysis revealed that the 14 % CP group was not significantly different from the 16 % group in egg number indicating the absence of any imbalance of LAA or deficiency of amino nitrogen in the experimental diets. Feed efficiency under various dietary groups during different periods which was expressed as FCR for every dozen egg produced did not show a significant difference among treatments but proved higher efficiency in 2nd and 3rd period. The feed efficiency declined non significantly as the CP levels decreased. The results of this study indicated that the requirement of LAA will be critical in the initial phase of egg production along with deficiency of protein beyond 1% level even with supplementation. Unlike the commercial layers, feed restriction was followed to prevent obesity. The reports of Ji *et al.* (2014) revealed that varying dietary CP had no effect on egg production and egg mass, expressed as percent and grams per hen per day, respectively. These findings were in accordance with the results of the present study but not on par with Adeyemo *et al.* (2012) who reported 14% CP diet balanced with AA was significantly different from the 16% CP diet group in terms of FCR. The variation in suggested amino acid requirements, including the composition of the basic diet, type of bird, dietary energy content, feed intake level and age of layers might have influenced some of the previous studies and might have caused dissimilarities with the present study.

During the peak production phase, there was no significant difference between treatments in terms of achieving optimal egg size. According to Blair *et al.* (1999), egg size was more susceptible to low dietary CP than egg production efficiency. However, the present study concluded that egg size is less susceptible to low CP when compared to the production of eggs which is contradictory to the above findings. Sohail *et al.* (2002) also found that Essential AA had a substantial impact on egg weight and that eliminating an essential AA resulted in a reduction in egg weight in 2 weeks. Roberts *et al.* (2007) found a comparable percentage unit reduction in CP level, and the reduced-CP diet had no effect on egg weight, but hens given the reduced-CP diet produced fewer eggs and so had lower egg mass than those fed with the control diet. The results of Ramarao *et al.* (2011) revealed that 15 % CP was adequate for layers during the entire laying cycle from 21 to 72 wk of age on the basis of EW, BW, and FE data. However, the results of this investigation support the conclusion that a 2 percent drop in CP with the addition of limiting amino acids had no adverse effect on egg weight. Results of egg quality studies are depicted in Table 4.

Table 4: The effect of reducing CP with balanced LAA on mean egg quality parameters during 29-40 weeks of age

Parameters	T1	T2	T3	T4	T5
Shape index ^(NS)	77.98±1.01	76.77 ±0.65	77.25±0.56	76.04±0.62	75.94±0.72
Shell thickness	0.390±0.0 ^b	0.371±0.0 ^a	0.381±0.0 ^{ab}	0.382±0.0 ^{ab}	0.384±0.0 ^{ab}
Albumen index ^(NS)	0.08± 0.0	0.09± 0.0	0.09± 0.0	0.09± 0.0	0.09 ±0.0
Yolk index ^(NS)	0.46± 0.01	0.45± 0.01	0.45± 0.0	0.46± 0.01	0.46 ±0.0
Haugh Score ^(NS) Unit	74.29± 0.85	73.07± 0.9	70.45 ±3.83	75.59 ±1.09	74.96 ±0.9

means in the column bearing at least one common superscript do not differ significantly ($p \geq 0.05$); NS-Non Significant

There are many dietary variables affecting egg quality, particularly shell thickness, which is one of the many significant quality characteristics in the egg industry. In this study, the mean shell thickness of all three periods was

significant among the treatments. The values recorded in CP reduced groups (T₃, T₄, and T₅) were comparable with control (T₁) and also comparable among themselves but was significant with T₂. Apart from good egg production, 1 % CP reduced group produced eggs with optimum shell thickness which is instrumental in egg marketing. Suma (2008) observed that although shell thickness values were statistically ($P \leq 0.05$) different during the experiment using Swarnadhara layers, there was no discernible trend in any of the dietary treatments which partly agrees with the findings in the present study. However, the mean egg shape index of all dietary groups was statistically similar during the study period. These findings are in agreement with Torki *et al.* (2015). On the contrary, Adeyemo *et al.* (2012) reported that shell thickness was significantly lower in 14 % CP when compared to 16 % even with supplementation of AA. The higher shape index value of indigenous chicken might be due to broad and short size of the eggs (Pathak *et al.*, 2016). The current findings on shape index of Swarnadhara eggs was in accordance with the conclusions of Suma (2008) that the shape index of these eggs is unaffected by the amount of energy/protein and calcium/phosphorus in the diets at any given level. The albumen index values remained non-significant between the treatments during the three periods which clearly agrees with the findings of Adeyemo *et al.* (2012) who reported that 14 % and 16 % of CP diets did not make any influence on the albumin index. A Low CP diet balanced with AA could support equal consistency of dense albumen of the eggs. Higher yolk index values indicate that the yolk is more stable, which is significant for egg shelf life and hatchability. In the present study, the overall mean yolk index values were not significantly different among various treatments. The Haugh unit score is based on egg weight and albumen height and serves as an important measure of egg freshness. The amount of ovomucin, especially β -ovomucin, secreted by the magnum indicates the quality of albumen as the β -ovomucin contributes to the gelatinous traits of the thick albumen gel (Daskiran *et al.*, 2009). However, mean HU during all the periods remained comparable among treatments which are supported by Adeyemo *et al.* (2012).

Conclusion

On analysis of the results of the trial, it is observed that 2 % CP reduction from control (16%) in the layer diet with supplementation of lysine, methionine, and threonine did not affect the production rate, egg weight, and egg characteristics of Swarnadhara layers during 29- 40 weeks of age. It is also found that 1% CP reduced diet supported the production of eggs in more numbers having good shell thickness. More studies are required in Swarnadhara layers with a higher rate of CP reduction to find out the maximum level up to which CP can be reduced using synthetic AA without impairing egg production and quality parameters.

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Contribution of Authors

The authors contributed equally.

Conflict of Interests

There is no conflict of interest.

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