

Original Research Article

Replacement of Soyabean Meal with Rice Distiller's Dried Grains with Soluble in Layer Diet on Chicken Egg Quality

ABSTRACT

The experiment was conducted on 144 layers of BV- 300 strain for the period of 12 weeks (59 to 70th weeks) to study the replacement of soyabean meal with rice distiller's dried grains with soluble (rDDGS) in layer diet on chicken egg quality. The layer birds were randomly distributed into 4 equal treatments T1 (Control), T2, T3 and T4 with 36-layer birds in each group having 6 replicates of 6 layers each. The control group T1 fed basal layer ration as per the recommendation of BV-300-layer strain and treatment groups T2, T3 and T4 offered 5, 10 and 15% rDDGS inclusion levels replacing soybean meal in diet and recorded for egg quality parameters. It was observed that, the inclusion of rDDGS at 0, 5, 10 and 15% in treatment groups T1, T2, T3 and T4 respectively, in layer diets were non-significantly influenced on shape index, egg surface area, albumin index, yolk index, yolk colour score, percent eggshell weight and eggshell thickness. The egg weights were significantly ($P<0.05$) decreased in treatment group T4 receiving diet at 15% rDDGS as compared to control group T1 and treatment group T2. However, there was non-significant differences for egg weight in groups T1, T2 and T3. Thus, it was concluded that the rice distiller's dried grains with soluble (rDDGS) can be included replaced up to 10% in layer diet with replacing soybean meal without affecting the external and internal egg quality parameters like eggshell weight, shape index, egg surface area, albumin index, yolk index, yolk colour score, egg surface area and eggshell thickness.

Key words: Layers, Egg weight, Shape index, Albumin index, Yolk index, Yolk colour, Soybean meal, Rice DDGS

1. INTRODUCTION

India is one of the largest producers of eggs and broiler meat. The total poultry production has increased by 16.81% with total poultry population at 851.85 million in 2019 [1]. In layer production, the major cost input is feed, accounting for 70-75% of total cost of commercial egg production. Soybean meal being the most popular choice as protein source in feed however due to high demand and decrease in soybean production, followed by increased soybean meal exports resulted in increased soybean

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price thereby increasing cost of poultry production. Due to the increasing cost of poultry production, one has to find newer cheap feed alternatives for cost-effective poultry production. But the choice of these alternative ingredients depends on many factors such as their availability, absence of toxic principles, nutrient availability, antinutritional factors, quality, nutritional composition and cost.

Distiller's dried grains with soluble (DDGS) is an important alternate protein supplement, which is available in appreciable quantities in recent years [2]. An increase in ethanol production over the last few years, due to increasing prices of conventional oil, limited underground reserves and an addition of ethanol in fuels, has led to an increased supply of DDGS, available as livestock feed [3]. Different types of cereals are used to produce biofuel from corn, rice, wheat, barley and sorghum are the commonly used cereals used for bio-ethanol production. In the present context, due to relative lower price, higher production and easy availability of rice, it is used as a substrate for bio-ethanol production. India is second largest producers of rice in [the World](#) after China, producing approximately 130.3 million tonnes rice in year 2021-22 [4]. Now a days, clean fuel policy in the country has given the guideline to incorporate 10% ethanol in petrol along with Ministry of Petroleum has given the mandate of using 20% ethanol in petrol by the year 2025. An increase in ethanol production over the last few years has led to an increased supply of DDGS, available as livestock feed [3].

Rice distiller's dried grains with soluble (rDDGS) is the by-product of the processing of rice alcohol industry which is produced from the distillation of fermented rice. On chemical analysis, it was found that rDDGS contained moisture 8.28%, DM 91.72%, CP 45%, EE 4.49%, CF 4.89%, TA 10.22%, NFE 27.12%, AIA 4.28%, ADF 15.20%, NDF 37.75%, Ca 0.73%, P 0.77% and GE 4097 kcal/kg [5]. DDGS is also a rich source of water-soluble vitamins, microelements and other biologically active substances like nucleotides, mannan-oligosaccharides, 1,6-glucan, inositol, glutamine and nucleic acids. The cost of rDDGS is less than that of the soybean meal hence rDDGS is used as alternate and economical protein source in poultry feed. Even though rDDGS is one of the best alternatives to the soybean meal in layer feed, but the scanty information is available in literature exploring the inclusion of rDDGS replacing soybean meal in layer diet. Keeping these facts in view, the present research was planned to study the replacement of soybean meal with rice distiller's dried grains with soluble (rDDGS) in layer diet on chicken egg quality.

2. MATERIALS AND METHODS

The experiment was conducted for twelve weeks period at Layer shed of Department of Poultry Science, College of Veterinary and Animal Sciences, Parbhani-431402, Maharashtra (India). The research Proposal was approved by IAEC (CPCSEA Reg No. 270/GO/Rbi/S/2000/CPCSEA) committee of COVAS, Parbhani, Resolution No. IAEC 98/22 date 07/07/2022.

2.1 Experimental Design

The experiment was conducted on 144 layers of BV- 300 strain for period of 12 weeks (59 to 70th weeks) reared in California cage (III-tier) housing system. The layer birds were randomly distributed into 4 equal treatments T1 (Control), T2, T3 and T4 with 36-layer birds in each group having 6 replicates of 6-layer birds each. The control group T1 fed basal ration as per the recommendation of BV-300-layer strain and treatment groups T2, T3 and T4 offered 5, 10 and 15% rDDGS inclusion levels replacing soybean meal. All the groups were provided with similar environmental and management conditions. Ideal and adequate floor space was allotted to all the birds as per the standards. Feeders and drinker troughs were attached to the cages running throughout the length outside the cage. The total 16 hours of light and eight hours of darkness was provided to the birds throughout the experimental period from 59th to 70th weeks (12 weeks). As per the day length, artificial light was provided to meet the requirement of 16 hours of light including day light. The replicates were distributed in such a manner to nullify the row and tier effect.

2.2 Experimental Diet

The chemical analysis of rDDGS was carried out as per AOAC [6]. The metabolizable energy content in rDDGS was reported 3500 kcal/kg as per NRC [7]. The metabolizable energy 3200 kcal/kg in rDDGS was considered for the feed formulation during the experiment. The experimental diets were prepared as per the BV-300-layer strain recommendation. All the layer diets were balanced *iso-caloric* and *iso-nitrogenous*. The details of the feed formulation of layer mashes used in trial are presented in Table 1.

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Table 1. Feed formulations of layer mash and nutrient composition for different treatment groups

Sr. No.	Ingredient (%)	Groups			
		T1	T2	T3	T4
1	Maize	56.21	53.91	51.61	49.31
2	De oiled rice bran	11.000	14.00	17.00	20.00
3	Soyabean meal	20.000	14.20	8.40	2.60
4	Rice DDGS	-	5.00	10.00	15.00
5	Marble grit	8.30	8.30	8.30	8.30
6	Limestone powder (LSP)	2.50	2.50	2.50	2.50
7	Dicalcium phosphate (DCP)	1.00	1.00	1.00	1.00
8	L-Lysine	-	0.12	0.24	0.36
9	DL-Methionine 98%	0.12	0.10	0.08	0.06
10	Trace mineral premix (Tracemin-CL)*	0.10	0.10	0.10	0.10
11	Vitamin premix (Lay vit)**	0.05	0.05	0.05	0.05
12	Salt pure	0.26	0.26	0.26	0.26
13	Sodium bicarbonate	0.10	0.10	0.10	0.10
14	Choline chloride 60%	0.15	0.15	0.15	0.15
15	Phytase 5000	0.01	0.01	0.01	0.01
16	Liver-tonic	0.05	0.05	0.05	0.05
17	Toxin binder	0.15	0.15	0.15	0.15
	Total	100.00	100.00	100.00	100.00
Nutrients (% Dry matter basis) calculated values					
1	Metabolizable Energy (kcal/kg)	2551.79	2552.18	2552.57	2552.96
2	Crude Protein (%)	15.53	15.54	15.55	15.56
3	Ether Extract (%)	2.45	2.49	2.52	2.56
4	Crude Fibre (%)	4.38	4.53	4.68	4.82
5	Calcium (%)	4.07	4.07	4.06	4.06
6	Available Phosphorus (%)	0.41	0.41	0.40	0.40
7	Dig. Lysine (%)	0.67	0.67	0.67	0.67
8	Dig. Methionine (%)	0.35	0.36	0.36	0.37
9	Dig. Cystine	0.21	0.24	0.26	0.28
10	Dig. Methionine +Cystine	0.56	0.60	0.62	0.65

*Trace mineral premix: Each kg contains: Manganese-80g, Zinc-80g, Iron-60g, Copper-15g, Iodine-1g and Selenium-300mg.

**Vitamin premix: Vitamin A-12.5 MIU, Vitamin D3-3.3 MIU, Vitamin E-40g, Vitamin K-2g, Vitamin B1-4g, Vitamin B2-10g, Vitamin B6-5g, Vitamin B12-0.016g, Niacin-33g, Cal. D. Pantothenate-15g, Folic acid-1g, Biotin-0.1g.

2.3 Parameters Studied

The impact of this research was assessed in terms of internal and external quality of egg, two eggs randomly collected from each replicate i.e. total twelve eggs from each treatment were collected at 4th, 8th and 12th weeks. Daily egg weight was measured by using a 0.0g sensitive digital scale during the experimental period. The measurements of length and width of egg were done by using digital vernier calliper. Then the eggs were cracked on the flat glass surface to measure width and height of albumen and yolk. Height of albumen and yolk was measured using circular spherometer. The shell thickness without shell membrane was measured with a digital micro-meter screw gauge. The egg quality parameters were calculated by using following formulas.

Shape index of egg:

$$\text{Shape index} = \frac{\text{Width of egg}}{\text{Length of egg}} \times 100$$

Egg surface area (cm²):

Egg surface area was calculated by following formula-

$$\text{Egg surface area (S)} = 4\pi r^2$$

Radius (r) was calculated as $\frac{1}{4}$ (length + breath) of egg.

$$\pi = 3.14159$$

Albumen index:

$$\text{Albumen index} = \frac{\text{Albumen Height}}{(\text{Albumen Length} + \text{Albumen width})/2} \times 100$$

Yolk index:

$$\text{Yolk index} = \frac{\text{Yolk height}}{\text{Yolk width}} \times 100$$

The colour of yolk was compared by Roche Yolk Colour Fan (RYCF). The eggshell weight was weighed on a digital scale (0.0 g precision). The shell thickness without shell membrane was measured with a digital micro-meter screw gauge.

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2.4 Statistical Analysis

All the data was analysed with Completely Randomized Design and Two Factor Factorial Experiment using software Web Agri Stat Package-2.0 (WASP 2.0) developed at ICAR research Complex, Goa [8]. The differences among treatments, within treatment groups were determined by analysing the data generated by using the Two Factor Factorial Experiment. Significant level among the treatment groups were determined by ANOVA and treatment means were compared by critical differences (CD).

3. RESULTS AND DISCUSSION

3.1 Proximate Chemical Analysis of rDDGS:

The proximate analysis for the rice DDGS was carried out as per AOAC [6]. The proximate chemical analysis of rice DDGS showed that it contained percent of moisture 6.08, crude protein 44.00, crude fibre 3.10, total ash 5.47, ether extract 3.39, total phosphorus 0.60 and calcium 0.27. Similar, analysis results of rDDGS were observed by Gupta *et al.*, 2016 and reported that rDDGS contained moisture 8.28%, DM 91.72%, CP 45%, EE 4.49%, CF 4.89%, TA 10.22%, NFE 27.12%, AIA 4.28%, ADF 15.20%, NDF 37.75%, Ca 0.73%, P 0.77% and GE 4097 kcal/kg.

3.2 Egg Weight

The data pertaining to the weight of the eggs (g) produced by the birds from different groups during experimental period are presented in Table 2. The egg weights were significantly ($P < 0.05$) decreased in treatment group T4 receiving diet at 15% rDDGS as compared to groups T1 and T2. However, there was non-significant differences for egg weight in groups T1, T2 and T3. The egg weights were statistically non-significant in treatment groups T3 and T4. This might be due to the high fibre content in rDDGS such as non-starch polysaccharides and potential bioavailability of amino acid. Similar, observations were recorded by Shalash *et al.* [9] reported that increasing DDGS to 15 to 20 in laying hen diets significantly ($P < 0.05$) decreased egg weight compared with other level (0, 5, and 10%) levels. Swiatkiwicz and Koreleski [10] observed that the dietary level of 20% maize DDGS negatively affected daily weight of egg in laying hens. Sedmake *et al.* [11] reported that inclusion of 15-20% corn DDGS in layer diet significantly reduced egg weight. El-Sheikh and Salama [12] revealed that egg weight was significantly decreased when inclusion of cDDGS from 20 to 30% in layers.

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Table 2. Average egg weight (g) of layers fed different levels of rDDGS in treatment groups

Weeks	Groups			
	T1 (0% rDDGS)	T2 (5% rDDGS)	T3 (10% rDDGS)	T4 (15% rDDGS)
59	57.59±0.42	57.54±0.50	57.46±0.57	57.92±0.13
60	57.90±0.73	57.85±0.19	57.49±0.57	57.23±0.50
61	58.21±0.84	57.98±0.13	57.28±0.69	56.94±0.38
62	57.96±0.74	57.94±0.22	57.25±0.61	57.96±0.19
63	58.30±0.86	57.65±0.46	57.50±0.49	57.68±0.22
64	57.62±1.07	57.86±0.19	57.66±0.48	57.42±0.50
65	54.71±0.26	58.01±0.26	58.36±0.17	57.62±0.19
66	58.03±0.30	58.19±0.45	58.21±0.09	56.90±0.41
67	59.49±0.65	56.34±0.20	57.28±0.23	58.07±0.24
68	58.68±0.53	57.34±0.19	57.74±0.36	56.89±0.51
69	57.41±0.14	57.48±0.32	57.80±0.28	57.78±0.15
70	57.26±0.15	58.75±0.34	57.64±0.38	57.84±0.21
Mean	57.76 ^a ±0.33	57.75 ^a ±0.17	57.64 ^{ab} ±0.10	57.40 ^b ±0.13

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These means with at least one common superscript do not differ significantly within a row.

Critical difference (CD) for treatments at 5% level = 0.345

3.3 Shape index

The shape index of the eggs produced by the birds at 59th and 70th weeks of age from different groups are presented in Table 3. From the data, it was revealed that the means of shape index from different groups at 59th and 70th weeks of age were statistically non-significant. The birds from group T4 receiving diet with 15% rice DDGS recorded numerically higher shape index amongst all the groups including control. Thus, the shape index was increased in the eggs with increased proportion of rice DDGS above 5% in layer diets as compared with control group. The inclusion of rDDGS at 5, 10 and 15% in diet replacing soybean meal did not affect the shape index in layers during 59 to 70th weeks of experiment. Similarly, Abd El-Hack *et al.* [13] revealed that the inclusion of DDGS in layer diet had no significant effect on shape index. El-Sheikh and Salama [12] observed that use of 20, 25 and 30% level of DDGS had non-significant

effect on shape index. Gupta *et al.* [14] revealed that addition of rDDGS levels and enzyme supplementation were found non-significant effect on shape index.

Table 3. Egg shape index and egg surface area of layers fed different levels of rDDGS in treatment groups

Egg shape index				
Weeks	Groups			
	T1 (0% rDDGS)	T2 (5% rDDGS)	T3 (10% rDDGS)	T4 (15% rDDGS)
4	76.67±0.62	76.44±0.77	76.28±0.42	77.35±0.75
8	77.18±0.48	76.37±0.81	77.57±0.77	77.71±1.83
12	76.59±0.34	76.60±0.56	77.69±0.47	78.04±1.45
Mean	76.81±0.19 ^{NS}	76.47±0.07 ^{NS}	77.18±0.45 ^{NS}	77.70±0.20 ^{NS}
CD	1.402			
Egg surface area (cm ²)				
4	76.70±0.64	76.90±0.65	76.51±0.78	74.88±0.88
8	76.38±0.77	76.69±0.90	76.29±0.84	78.35±1.35
12	77.05±0.89	76.44±0.76	77.12±0.57	76.63±0.85
Mean	76.71±0.19 ^{NS}	76.68±0.13 ^{NS}	76.64±0.25 ^{NS}	76.62±1.00 ^{NS}
CD	1.243			

Those means with at least one common superscript do not differ significantly within a row.

CD-Critical difference

3.4 Egg surface area (cm²)

The data pertaining to surface area (cm²) of egg produced by the birds from different groups during experimental period are presented in Table 3. The statistical analysis of the data revealed that the differences in egg surface area between the groups were statistically non-significant. The egg surface area was numerically higher in control group as compared with treatment groups T2, T3 and T4 receiving diets at 5, 10 and 15% rDDGS, respectively. No literatures are available on the egg surface area by using DDGS, cDDGS and rDDGS in layers. From the above finding it was revealed that inclusion of rDDGS at 5, 10 and 15% in layer diets recorded non-significantly decreased egg surface area as compared to control group.

3.5 Albumin index

The data pertaining to the albumen index of the eggs produced by the birds from different groups during experimental period are presented in Table 4. The statistical analysis of the data revealed that the differences in albumin index between the groups were statistically non-significant. The findings of the present study are in accordance with the work of Zile and Sajjan [15] reported that inclusion of DDGS with or without multienzyme supplementation in layer diet recorded similar albumin index in all treatment groups. Gupta *et al.* [14] revealed that addition of 100g/kg rDDGS levels plus enzyme supplementation were found to have no significant effect on albumin index. From the above findings it was reported that inclusion of rDDGS up to 15% levels in layer diet had no significant effect on the albumin index.

3.6 Yolk index

The data pertaining to the yolk index of the eggs produced by the birds from different groups during experimental period are presented in Table 4. The statistical analysis of the data revealed that the differences in the yolk index of eggs between the groups were statistically non-significant. However, there was a numerical decrease in the values for yolk index of eggs in treatment groups as compared to control group. The findings of the present study are in accordance with the work of Ghazalah *et al.* [16] observed that addition of DDGS had insignificant effect on yolk index. Olofintoye and Bolu [17] revealed that yolk index was not affected by the inclusion of cDDGS at 0, 10, 20, 30 and 40% levels. El-Sheikh and Salama [12] observed that the yolk index was not affected by inclusion level of cDDGS at 20, 25 and 30%. In contrast with Abd El-Hack *et al.* [18] revealed that eggs produced from hens fed the basal diet or diets included 5 and 10% DDGS gave the best yolk index compared with eggs from hens fed 15% DDGS. Gupta *et al.* [14] revealed that the 100g/kg rDDGS level had significantly ($P<0.01$) higher yolk index. From the above findings it was reported that inclusion of rDDGS at 5, 10 and 15% inclusion levels in layer diets did not influence the yolk index of eggs.

Table 4. Albumin index, yolk index and yolk colour score of chicken eggs of layers fed different levels of rDDGS in treatment groups

Albumin index				
Weeks	Groups			
	T1 (0% rDDGS)	T2 (5% rDDGS)	T3 (10% rDDGS)	T4 (15% rDDGS)
4	9.82±0.28	9.80±0.45	9.73±0.46	9.87±0.32
8	9.77±0.18	9.94±0.20	9.89±0.37	9.70±0.26
12	10.27±0.17	10.13±0.38	10.20±0.50	10.30±0.17
Mean	9.95±0.16^{NS}	9.95±0.10^{NS}	9.94±0.14^{NS}	9.96±0.18^{NS}
CD	0.532			
Yolk index				
4	40.05±0.58	40.16±0.63	40.07±0.59	39.97±0.49
8	41.56±0.82	41.14±0.69	40.54±0.63	40.91±0.60
12	42.22±0.80	42.45±0.44	42.00±0.53	41.95±0.98
Mean	41.28±0.64^{NS}	41.25±0.66^{NS}	40.87±0.58^{NS}	40.95±0.57^{NS}
CD	1.012			
Yolk colour score				
4	4.92±0.08	4.75±0.13	4.75±0.13	4.92±0.15
8	4.83±0.11	4.67±0.14	4.83±0.11	4.92±0.08
12	4.83±0.11	4.83±0.11	4.92±0.08	4.83±0.11
Mean	4.86±0.03^{NS}	4.75±0.05^{NS}	4.83±0.05^{NS}	4.89±0.03^{NS}
CD	0.177			

Those means with at least one common superscript do not differ significantly within a row.

CD-Critical difference

3.7 Yolk colour score

The data pertaining to the mean yolk colour score of the eggs produced by the birds from different groups during experimental period are presented in Table 4. The statistical analysis of the data revealed that the differences in yolk colour score of eggs between the groups were statistically non-significant. It is revealed that the treatment group T4 receiving diet of 15% rDDGS recorded numerically increased yolk colour score as compared to control group. No literature available on the yolk colour score of eggs aspects of rice DDGS addition in layer diet. However, many researchers were carried out the research work on cDDGS in layers on the yolk colour parameters. In

contrast of the present study, Masa'deh *et al.* [19] observed that adding up to 25% DDGS in layer diets improved egg yolk colour at the higher inclusion rate. Attia [20] revealed that yolk colour was significantly increased by feeding 20% DDGS diet as compared to control group. Abd El-Hack *et al.* [13] revealed that as the dietary DDGS level increased, yolk colour density increased. Many researchers reported that use of cDDGS in layers significantly improved the yolk colour score, it may be due to the presence of Xanthophylls and Carotenoids pigmentation in corn DDGS. From the above study it was observed that inclusion of rDDGS at 5, 10 and 15% inclusion levels in layer diets did not influence the yolk colour of eggs.

3.8 Percent eggshell weight

The data pertaining to the mean percent eggshell weight produced by the birds from different groups during experimental period are presented in Table 5. From the data it is revealed that, the percent eggshell from all the groups is statistically non-significant. It is revealed that the percent eggshell weight from treatment group T3 receiving diet at 10% rice DDGS was numerically increased as compared to all other groups. The findings of the present study are in accordance with the work of El-Sheikh and Salama [12] used three levels of DDGS (20, 25 and 30 %) and revealed that shell weight present were not affected by DDGS level. Gupta *et al.* [14] observed that the different level of DDGS supplementation had no significant effect on eggshell weight. From the above finding it was revealed that, the use of rDDGS at 5, 10 and 15% inclusion levels were non-significantly affected on percent eggshell weight.

3.9 Eggshell thickness (mm)

The data pertaining to the shell thickness of the eggs produced by the birds from different groups during experimental period are presented in Table 5. From the data it is revealed that, the eggshell thickness from all the groups is statistically non-significant. The eggshell thickness was non-significantly affected by the inclusion of rDDGS at 0, 5, 10 and 15% with replacing soybean meal in layer diet. The findings of the present study are in accordance with the work of Swiatkiewicz and Koreleski [10] reported that the 0, 5, 10, 15 and 20% levels of cDDGS in the diet had no negative effect on eggshell thickness. Rew *et al.* [21] reported that eggshell thickness was not affected by feeding of cDDGS at 0,10 and 20% in laying hens. From the above finding it was revealed that the inclusion of rDDGS in diets had no significant effect on eggshell thickness and rDDGS can be used up to 15% without any effect on eggshell thickness.

Table 5. Percent eggshell weight and eggshell thickness (mm) of layers fed different levels of rDDGS in treatment groups

Percent eggshell weight				
Weeks	Groups			
	T1 (0% rDDGS)	T2 (5% rDDGS)	T3 (10% rDDGS)	T4 (15% rDDGS)
4	12.13±0.30	12.19±0.23	12.98±0.24	11.78±0.21
8	11.81±0.22	12.23±0.34	12.24±0.39	12.19±0.28
12	12.46±0.17	11.91±0.28	12.14±0.26	12.11±0.29
Mean	12.13±0.19^{NS}	12.11±0.10^{NS}	12.45±0.26^{NS}	12.03±0.13^{NS}
CD	0.430			
Eggshell thickness (mm)				
4	0.39±0.01	0.38±0.00	0.39±0.00	0.38±0.01
8	0.36±0.01	0.36±0.01	0.36±0.01	0.37±0.01
12	0.37±0.01	0.37±0.01	0.37±0.01	0.36±0.01
Mean	0.37±0.01^{NS}	0.37±0.01^{NS}	0.37±0.01^{NS}	0.37±0.01^{NS}
CD	0.015			

Those means with at least one common superscript do not differ significantly within a row.

CD-Critical difference

4. CONCLUSION

Thus, it was concluded that the rice distiller's dried grains with soluble (rDDGS) can be included up to 10% in layer diet with replacing soybean meal without affecting the external and internal egg quality parameters like eggshell weight, shape index, egg surface area, albumin index, yolk index, yolk colour score, egg surface area and eggshell thickness.

According to the findings, there was a significant different in egg weight, however others parameters such as egg shell weight, thickness showing a non-significant result. Can you explain what contribute to the different in the weight of the egg?

REFERENCES

1. Livestock census. 20 All India Livestock Census, Department of Animal Husbandry and Dairing Ministry of Agriculture, GOI;2019.
2. Raju MVLN, Rao SV, Prakash B. Performance, serum biochemical profile, slaughter characteristics and nutrient retention of Vanaraja chicks fed rice-based distillers dried grains with solubles in diet. *Indian J. Anim. Res.* 2022;56(2):182-186.
3. Noll SL, Brannon J, Parsons C. Nutritional value of corn distiller dried grains with solubles (DDGs): Influence of solubles addition. *Poult. Sci.* 2007;86(1):68.
4. USDA. Commodity Intelligence Report, India Rice: Production down due to decline in planted area. <https://ipad.fas.usda.gov/highlights/2022/12/India/index.pdf> (Accessed 22 December 2022).
5. Gupta J, Wilson BW, Vadlani PV. Evaluation of green solvents for a sustainable zein extraction from ethanol industry DDGS. *Biomass and Bioenerg.* 2016;85:313-319.
6. AOAC. *Official Methods of Analysis*, 17th ed. Association of Official Analytical Chemists, Arlington, VA, USA;2000.
7. NRC. *Nutrient Requirements of Dairy Cattle*, 6th ed. National Research Council, National academic science, Washington, DC;1989.
8. WASP 2.0. *Web Agri Stat Package*, ICAR Research Complex for Goa, Ela, Old Goa, Goa. 403 402. India;2004.
9. Shalash SMM, Abou el-wafa S, Hassan RA, Ramadan NA, Mohamed MS, El-gabry HE. Evaluation of distillers dried grains with solubles as feed ingredient in laying hen diets. *Int. J. Poult. Sci.* 2010;9(6):537-545.
10. Swiatkiewicz S, Koreleski J. 2006. Effect of maize distillers dried grains with solubles and dietary enzyme supplementation on the performance of laying hens. *J. Anim. Feed Sci.* 2006;15(2):253-260.
11. Sedmake E, Khose K, Manwar S, Gole M, Wade M, Wankhede S. Effect of feeding corn distiller's dried grains with solubles on egg quality traits in commercial layers. *Int. J. Livest. Res.* 2018;8(8):273-284.
12. El-sheikh SEM, Salama AA. Utilization of dry corn grains distillation by-product with soluble (DDGS) as an alternative non-conventional feed stuff in laying hen diets. *Egypt. J. Nutri. feeds.* 2020;23(3):473-483.
13. Abd El-hack ME, Mahrose KM, Attia FA, Swelum AA, Taha AE, Shewita RS, Alowaimer AN. Laying performance, physical, and internal egg quality criteria of hens fed distillers dried grains with solubles and exogenous enzyme mixture. *Anim.* 2019;9(4):150.

14. Gupta SL, Tyagi PK, Mir NA, Dev K, Begum J, Mandal AB, Tyagi PK. Feeding value of rice distiller's dried grains with solubles as protein supplement in diet of laying hens. *Trop. Anim. Health. Prod.* 2020;52(3):1229-1237.
15. Zile S, Sajjan S. Effect of replacement of soybean meal with distillers dried grains with solubles in the diets with or without multienzyme supplementation on the performance of layers. *Indian J. Anim. Nutri.* 2014;31(3):297-301.
16. Ghazalah AA, Abd-Elsamee MO, Moustafa ES. Use of distillers dried grains with solubles (DDGS) as replacement for soybean meal in laying hen diets. *Int. J. Poult. Sci.* 2011;10:505-513.
17. Olofintoye OR, Bolu SA, 2013. Effects of corn distillers dried grains on the performance and egg quality of laying hen. *Anim. Res. Int.* 2013;10(1):1665-1672.
18. Abd El-Hack ME, Alagawany M, Farag MR, Dhama K. 2015. Use of maize distiller's dried grains with solubles (DDGS) in laying hen diets: trends and advances. *Asian J. Anim. Vet. Adv.* 2015;10(11):690-707.
19. Masa'deh MK, Purdum SE, Hanford KJ. Dried distiller's grains with solubles in laying hen diets phosphorus. *Poult. Sci.* 2011;90(9):1960-1966.
20. Attia YAE. Utilization of different levels of distillers dried grains with solubles (DDGS) in local laying hen diets. *Egypt. Poult. Sci. J.* 2017;37(2):545-558.
21. Rew HJ, Shin MH, Lee HR, Jo C, Lee SK, Lee BD. 2009. Effects of corn distiller's dried grains with solubles on production performance and economics in laying hens. *Korean J. Poult. Sci.* 2009; 36(1):15-21.