Biomechanism and Bioenergy Research 1(1), 12-15

Investigating the Effects of Using of Biochar as a Replacement for Minerals Premix in Layers Diet on Physical and Mechanical Properties of Their Egg Shells

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INFO

RESEARCH PAPER

KEYWORDS

Biochar; Eggshell Quality; Laying Hens; Mineral Premix; Vitamin C

Received: *08 February 2022*

Revised: *13 March 2022*

Accepted: *17 March 2022*

Available Online: *30 March 2022*

ABSTRACT

Biochar is a mineral compound with a low cost of production. This compound can also be used as a fertilizer in agriculture to help plants grow faster and is a main source of bioenergy for heating and cooking. The goal of this study was to investigate how replacement of biochar with a mineral supplement, as well as its interaction with vitamin C, would affect eggshell quality of laying hens. In a 5×2 factorial treatment design, 400 laying hens were randomly assigned to 10 treatments, 5 replications, and 8 laying hens for each repetition. Factors tested included biochar levels (0, 25, 50, 75, and 100 percent replacement with mineral supplements of diet) and vitamin C levels (0 and 100 mg/kg of diet). The results showed that different experimental diets had no significant effect on eggshell quality (shell breaking strength, shell weight, eggshell thickness, and eggshell ash) of laying hens. The results revealed that biochar, due to its availability and easy production, can replace mineral supplements in laying hens' diet, with no adverse effects on eggshell quality traits.

INTRODUCTION

Eggshell damage is a serious problem and causes major economic losses for the egg industry (Fathi et al, 2019). Additionally, it has been estimated that losses from damaged eggshells account for 8% to 11% of total egg production (van Mourik *et al.*, 2017). On the other hand, the physical and mechanical properties of the egg are important aspects to ensure high quality for consumers. It has been reported that eggshell quality depends on a wide range of factors, such as diet (Fathi *et al.*, 2019). Preparing a diet for laying hens is expensive. The cost of the ration must be reduced by lowering the cost of the ration's components. Mineral supplements are one of the ration's components.

Biochar is a thermal degradation product of biomass that is created in a pyrolysis-like environment by restricting the supply of oxygen. This compound can also be used as a fertilizer in agriculture to help plants grow faster (Novak *et al.*, 2009). The biochar contains two organic acids (humic and fulvic acid), with humic acid playing a function in promoting chicken performance and increasing the animal's immunity (Kocabağli *et al.*, 2002). Most importantly, biochar is less expensive than mineral supplements and can be made quickly. Because the biochar is made up of organic waste, wood, and other agricultural byproducts that have been burned (Beesley and Dickinson, 2011; Laghari *et al.*, 2016). As a result, substituting a mineral supplement for this combination in the diet has the potential of lower the expense of the diet. However, there are few researches on the use of biochar in the diet of laying hens.

One of the properties of vitamin C (VC) is that it has chelating characteristics, which helps birds absorb minerals better (Saki *et al.*, 2011; Whitehead and Keller, 2003). Furthermore, prior studies have shown that VC improved egg quality of laying hens (Delos Reyes *et al.*, 2021). However, no study has compared the biochar to a mineral supplement, and no study has used the biochar in combination with VC in laying hens, to the best of this researcher's knowledge. In line of the foregoing, the purpose of this study was to determine the effects of replacing biochar with a mineral supplement in the diet, as well as its interaction with VC, on physical and mechanical egg properties.

MATERIALS AND METHODS

Birds, Diets, and Experimental Design

A total of 400 laying hens were used in a 5 × 2 factorial experiment with 10 treatments include five levels of biochar (0, 25, 50, 75 and 100% replacement of mineral premix), and two levels of VC (0 and 100 mg/kg of diet), with 5 replicates and 8 hens per replicate. The experiment was performed with laying hens from the age of 50 to 62 weeks. The room temperature was kept at 20 ± 3°C and the daily lighting schedule was standardized (16 h light, 8 h dark) during the entire experiment. Experimental diets and water were provided *ad libitum* during the entire experiment. The basal diet was formulated according to the requirements suggested in agreement with the Bovans white guide in Table 1. The biochar was supplied from a commercial

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DOI: 10.22103/bbr.2022.18995.1005

biochar company (Kuhpayeh, Kerman, Iran). Using a step-by-step dilution approach, biochar was added to the basal diet. Analytical results of biochar compound are shown in Table 2. These analyzes are based on information provided by the company. The trial was conducted according to the animal welfare guidelines at the Veterinary Control and Research Institute of Kerman, Iran.

Ingredients (%)	(50 to 62 weeks)
Corn	46.69
Soybean meal	28.30
Wheat	10.00
Soybean oil	2.83
Dicalcium phosphate	0.99
Carbonate calcium	6.70
Limestone	3.35
Common salt	0.34
DL-Methionine	0.30
Vitamin premix†	0.25
Mineral premix‡	0.25
Calculated analysis (%)	
Metabolizable energy (Kcal/kg)	2850
Crude protein	17.50
Crude fiber	2.31
Calcium	4.10
Available phosphorus	0.46
Arginine	1.25
Lysine	1.04
DL-Methionine	0.46
Methionine + Cystine	0.75
Tryptophan	0.24
Linoleic acid	1.10

Table 1. Composition and calculated analysis of experimental diets (as-fed basis)

†The vitamin premix supplied the followings per kilogram of diet: vitamin A, 88,000 IU; vitamin E, 165 IU; vitamin D3, 40,000 IU; vitamin K3, 22 mg; thiamine, 15 mg; riboflavin, 48 mg; pantothenate, 350 mg; niacin, 80 mg; vitamin B6, 25 mg; folic acid, 25 mg; vitamin B_{12}, 0.1 mg; Biotin, 1.5 mg; Colin chloride, 4,000 mg.
‡The mineral premix supplied the followings per kilogram of diet: Mn, 800 mg; Fe, 660 mg; Zn, 800 mg; Cu, 105 mg; I, 9 mg; Se, 2 mg

Eggshell Quality Traits

Eggshell quality traits were assessed using randomly-collected samples of 6 eggs per replicate with 2 eggs per day during the last 3 days of experiment. Eggshell quality traits including eggshell weight, eggshell thickness, eggshell breaking strength and egg shell ash were measured. Eggshell weight was measured after the eggshell dried at room temperature for 48 h. The shell ratio was calculated as eggshell weight/egg weight × 100. The eggshell thickness was measured by a digital micrometer after removing the shell membrane, and it was measured from three different regions (top, middle and bottom). For the calculation of eggshell ash, the eggshells were dried in an oven 60°C for 48 h. These dried samples were then ground and put to furnace at 650°C for 5.5 h (Aydin *et al.*, 2010). Eggshell breaking strength was measured by using a texture analyzer (STM-1, Santam Co., Tehran, Iran; Fig. 1), described by Ezazi *et al.* (2021)). For this reason, all eggs were subjected into the instrument in a horizontal position utilizing a 5 mm I diameter probe and operation speed of 100 mm/min maximum force (N), which that was punctured the eggshell was chosen as strength of the shell.

Statistical Analysis

The data were analyzed in a completely randomized design with treatments arranged in a 5 × 2 factorial to evaluate five levels of biochar (0, 25, 50, 75 and 100% replacement of dietary mineral premix), two levels of VC (0 and 100 mg/kg of diet), and the interactions among these factors. All data were analyzed using the GLM procedure of Mini-Tab (Mini-Tab, 2016). The means were compared by the Tukey's tests ($P < 0.05$).

Table 2. Analytical results of biochar compound complex

Chemical composition	Percentage
Energy (kcal/kg)	48.5
Crude fiber	5.18
Crude protein	1.46
Ether extract	<0.1
Ash	87.97
Calcium	0.60
Phosphorus	0.04
Selenium	<0.01
Copper	0.01
Iodin	<0.01
Iron	6.9
Manganese	0.02
Zinc	0.01
Humic acid	1.6
Fulvic acid	0.06
Organic matter	12.03

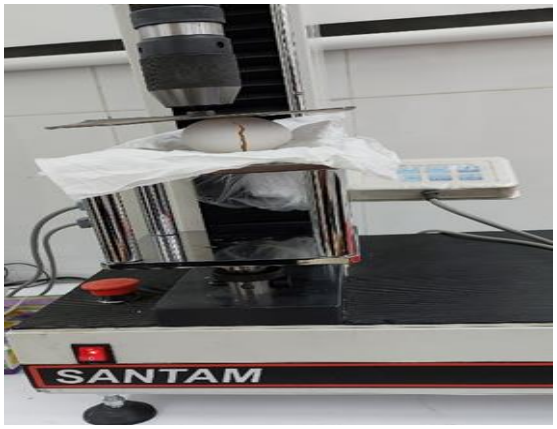

Fig. 1 Measuring eggshell breaking strength by using a texture analyzer.

RESULTS AND DISCUSSION

Table 3 shows the effects of the biochar complex with VC on the eggshell quality of laying hens.

Replacement of biochar levels in the diet from 25% to 100%, with or without VC, showed no influence on eggshell breaking strength, eggshell weight, eggshell thickness, and eggshell ash over the entire experiment period (50 to 62 weeks) ($P > 0.05$). The current study found that biochar at a concentration of 100 percent could be used as a mineral premix alternative in the feed of laying hens with no negative effects on eggshell quality (eggshell breaking strength, eggshell weight, eggshell thickness, and eggshell ash). However, the application of biochar as a mineral premix substitute in poultry is still a relatively new study.

Table 3. Effects of biochar complex and its interaction with vitamin C (VC) on egg shell quality of laying hens at 50-62 weeks of age.

Biochar (% substitution levels with mineral premix)	VC	Shell breaking strength (N/cm^2)	Shell percentage (%)	Eggshell thickness (mm)	Eggshell ash (%)
0	-	35.69	9.531	0.426	86.28
0	+	31.19	9.371	0.420	86.46
25	-	34.71	9.703	0.429	86.74
25	+	35.59	9.470	0.420	86.38
50	-	33.79	9.706	0.431	87.15
50	+	34.26	9.494	0.419	87.23
75	-	31.47	9.663	0.425	86.32
75	+	33.47	9.206	0.407	86.41
100	-	31.90	9.730	0.419	86.14
100	+	31.98	9.681	0.423	86.85
SEM		1.533	0.172	0.007	0.461
Main effect					
0		33.44	9.451	0.423	86.37
25		35.15	9.587	0.424	86.56
50		34.03	9.600	0.425	87.19
75		32.47	9.435	0.416	86.37
100		31.94	9.706	0.421	86.49
SEM		1.086	0.122	0.005	0.326
	-	33.51	9.667^{a}	0.426	86.53
	+	33.30	9.444^{b}	0.418	86.67
SEM		0.687	0.077	0.003	0.206
P-value					
BIO		0.261	0.496	0.666	0.366
VC		0.826	0.048	0.064	0.630
Biochar × VC		0.324	0.824	0.623	0.847

[a-b]Different letters in the same column indicate significant differences (P<0.05), and the same letters mean no significant difference (P>0.05). Vitamin C, without (-) or with (+). SEM, standard error of means.

There have been no reports of the effect of replacing mineral premix with biochar on laying hen egg quality traits. The addition of biochar to the laying hens' feed enhanced shell strength to crushing and thickness, according to Kalus *et al.* (2020). The effect of biochar as a feed supplement on egg quality traits in laying hens was studied by Prasai *et al.* (2018). The results showed that 1%, 2%, and 4% biochar treatments increased eggshell weight, thickness, and breaking strength in 36-week-old hens. There are few reports about the effect of biochar on eggshell in laying hens.Hakan *et al.* (2012) reported that the use of humate supplementation did not affect eggshell thickness in laying hens. However, Abo-Egla *et al.* (2011) showed that humic acid in laying hens diet increased eggshell thickness.

Dietary supplementation of VC has been reported to improve the productive performance of laying hens raised under heat stress conditions (Whitehead and Keller, 2003). It has been proposed that minor beneficial effects of supplemental VC in poultry raised under normal temperature conditions are likely related to the fact that poultry can ynthesize sufficient amounts of endogenous VC to meet their requirements. Therefore, an additional supply of VC is not required (Delos Reyes *et al.*, 2021), which is in accordance with the results of the present research. In line with the present study's findings, Delos Reyes *et al.* (2021) showed that the supplementation of laying hens feed with VC did not affect the eggshell thickness, eggshell strength and eggshell color. The likely reason for the lack of beneficial effects might be related to potential interactions between Ca, vitamin D3, and VC in diets (Delos Reyes *et al.*, 2021; Whitehead and Keller, 2003). It is proposed that the positive effect of VC on eggshell quality may be observable when the dietary supply of Ca and vitamin D3 is inadequate (Delos Reyes *et al.*, 2021; Whitehead and Keller, 2003). Ciftci *et al.* (2005) reported that the use of VC at 200 mg/kg of diet did not affect eggshell percentage compared to the control group. Seven (2008) showed that VC increased eggshell thickness and eggshell weight in laying hens under different environmental temperatures.

CONCLUSIONS

The current study reveals that different levels of biochar (0, 25, 50, 75 and 100% replacement of mineral premix), can substitute mineral supplements in laying hens' diets with no negative impacts on eggshell quality traits (eggshell breaking strength, eggshell weight, eggshell thickness, and eggshell ash), because to its availability and ease of manufacture. In addition, the supplementation of laying hens feed with VC did not affect eggshell quality trait

ACKNOWLEDGEMENTS

The authors wish to express their gratitude to the Shahid Bahonar University of Kerman for equipment help during the experiment and engineer Mohammad Majidi Kohbanani to prepare biochar produced from mine in this study.

CONFLICT OF INTEREST

The authors report there are no competing interests to declare.

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