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EFFECT OF VARIOUS PROPORTIONS OF LARGE AND SMALL PARTICLES OF LIMESTONE ON EGG PRODUCTION, EGGSHELL AND BONE QUALITIES AT THE PEAK OF EGG PRODUCTION IN GOLDEN MONTAZAH CHICKEN

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ABSTRACT : This research examined the inspiration of different proportions of large and small particles of limestone on egg production, eggshell quality and breaking strength of tibia from 32 to 44 weeks of age in Golden Montazah layers diets . Chickens were randomly allocated to four groups - 32 pullets per group were divided into four replicates. Limestone consisting of fine (F< 0.6 mm) and large (L>2.8mm -4.5mm). All chickens obtained a basal diet of the similar formula. Variations between four equal groups depends on the ratio of fine pulverized limestone by larger particle size limestone . Significantly higher eggshell breaking force (31.69N), shell weight (5.65 g), and shell thickness (0.46 mm) were obtained for eggs from the fourth treatment of hens than (29.46 N; 5.33 g; 0.37mm respectively) for eggs from the control group. Tibia breaking strength was significantly higher for the fourth group (350.66 N) vs. (252.82 N) respectively than those in control group. The conclusion of our study present some chances of eggshell quality enhancement by the use of larger limestone particles as a Ca supplement in layer diets. From 60% to 100% replacement of small size by large particle of limestone given helpful results on eggshell and tibia bone qualities.

Key words: Egg production, eggshell quality, limestone, particle sizes, hens, tibia bone



INTRODUCTION

In poultry nutrition calcium is one of the important minerals. Calcium has essential roles as the major element of bone formation and contribution in acid-base balance and enzymatic system, it also the major factor of eggshell. It was assessed that every egg contains 2.2g of calcium present essentially in eggshell.

Animal nutrition required calcium addition, that most of the diets contain of grains and its by-products, which contain very little calcium contents . Adequacy of the recommended quantities of nutritional Ca for layers is still being intended. Castillo et al. (2004), perceiving the valid NRC recommendations (1994) contradictory, carried out a research establishing the effect of 5 levels of Ca (from 2.93% to 4.82%) on production and quality of egg shell in a modern layer hybrid -white eggs, to determine the biologically optimal level to realize maximum production and egg shell quality. Based on research results, the authors reported that under given conditions, biologically optimal level of Ca necessary to realize the maximum production was 4.38%, for maximum egg shell quality 4.64%, and optimal economically level for maximum profit was 4.35% of Ca in diets.

Many researchers in their studies start with the assumption which is becoming scientific proven fact, that the adequate Ca nutrition of laying hens does not include only the adequate level of Ca in diet, but also adequate source and particle size of Ca. It is considered that larger particles of Ca source are retained longer in the gizzard and are slowly being degraded and absorbed during this longer passage through digestive tract, whereas the smaller Ca particles pass faster through the digestive tract and therefore they get degraded/absorbed partially (Svihus, 2011).

Laying hen diets containing limestone as the principle supplemental Ca source due to its abundant natural reserves, low cost and easy incorporation into layer feeds. Different aspects of shell quality were reported to be improved by the partial replacement of limestone with oyster shells in the diet (Roberts, 2004).

There are many factors effect on eggshell quality, involving the age of layers, strain of fowl, nutrition, ambiental conditions and diseases (Woolford, 1994; Vitorovi et al., 1995).

Laying chickens needed a excessive nutritive calcium requirement for shell calcification, because of approximately 95 % of the dried eggshell is calcium carbonate. Pulverized limestone is the major supplier of calcium in chicken diets. However, eggshell development mostly happens at evening when the layers stopped eating, and consummation of requirements is not enough, so it has been indicated that calcium absorption may be extra continued if the pullet is given approach to particulate calcium which might persevere for longer in the gut after consumption ceases (Guinote and Nys, 1991; Roberts and Nolan, 1997 , Roberts ,2010).

Limestone large particle size conducted to excessive amount in gizzard and duodenal soluble Ca at the finale of eggshell calcification in layers (Guinote at al., 1995; Zhang and Coon, 1997).

Zhang and Coon (1997) showed that grander particles of limestone with littler in vitro solubility was retained in the

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gizzard for a longer time , that may enhance Ca preservation.

Roland and Bryant, (1999) concluded that when 50% of pulverized limestone replaced via big particles were obtained to be quite satisfactory so as to optimize eggshell quality

To make sure getting good eggshell quality in the last third of the laying period by replacing two-thirds of the Ca in the diet as large particles which reported by Lichovnikova (2007).

There were some investigations on the replacement of calcium particle size suppliers which have some favorable effects for the goodness of eggshell (Lichovnikova, 2007; Skřivan et al., 2010) besides layer bones when larger grains of calcium sources are presented in the diets (Saunders-Blades et al., 2009; Witt et al., 2009; Araújo et al., 2011; Cufadar et al., 2011; Oliveira et al., 2013; Tunç and Cufadar, 2015).

Various variables described the eggshell strength like thickness, shell stiffness and rupture strength. Eggshells should be strong enough to prohibit broking so as to maintain the embryo until hatching (Altuntas, E. 2010).

The several egg external properties like mass, volume, surface area, shell thickness and weight affect the external properties of hen eggs.

The quasi-static, un destructive compression of an egg between two parallel steel plates is a usual procedure for the criterion of the shell force of a layer egg. The greatest correlation was found between the physical and the mechanical properties of chicken eggs (De Ketelaereet et al., 2002).

Pavlovski et al., (2000) concluded that when hens obtaining a feed intake in which 60 % of pulverized limestone was substituted by two gritty types of limestone had a favorable effects on eggshell quality

Vitorović et al. (2004) pointed out that positive effects on the egg shell quality can be realized by substituting 60 to 80% of lime with large size particles in mixtures for nutrition of older layers. Pavlovski Zlatica et al. (2003)demonstrated some potentials of eggshell quality improvement by substituting 60-80 % of fine limestone by coarse particles had favorable results on eggshell quality.

Skrivan et al., (2010) showed that the slowing solubilisation of Ca sources would make calcium available during the time eggshell calcification and decrease bone Ca and p mobilization

Wang et al., (2014) Was writing a report for higher productive performance, egg quality, and bone qualities when used large particle of limestone than oyster shell for practical applications in laying ducks.

The purpose of the current experiment occur to compare the results of using diets with various amounts of fine limestone substituted by large particles limestone at peak production on egg production, eggshell and bone qualities.

MATERIALS AND METHODS

This investigation was carried out at El-Research Station. Fayoum Animal Production Research Institute, Agricultural Research Center, Ministry of Agriculture, Egypt. Some of the physical and mechanical properties for eggshells and bones were carried out at Engineering Research Agricultural Institute, ARC, Min. of Agri., Egypt.

Chickens and Experimental Design:

One hundred and twenty eight 32-weeks old Golden Montazah chickens were randomly distributed to four groups - 32 pullets per group were divided into four

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replicates - and housed in cages individually from 32 to 44 weeks of age. The substitution part of fine pulverized limestone by larger particle size of granular limestone was the only differences between the groups.

We obtained the two particle types of limestone grit from a commercial provider of limestone which used in poultry industry. Small or Fine (F) and large (L) particles were (F < 0.6 mm) and (L > 2.8-4.5 mm), respectively. Limestone was screened through sieves which used (0.6 mm, 2.8 mm and 4.5 mm) particle sizes of the limestone were measured manually to obtain samples with appropriate diameters.

The firstly treatment (control) obtained the basic diet with merely pulverized limestone being as the calcium addition.

In the other experimental diets fine limestone was replaced by (60% ,. 80% and 100% respectively) of granular limestone

Chickens were supplied with diets and water ad libitum. All treatments obtained a main diet (Table 1) of the similar structure. Experimental birds exposed to 16.30 hours of light at 17 weeks of age.

Experimental diets were balanced and composed to be isocaloric and isonitrogenous with only the sizes of limestone particle inside diets . The diets were analyzed according (AOAC, 1990).

Body weight was carried out by weighing hens at starting and in the ending of investigational period. Egg production logged daily. Feed intake and egg weight were documented biweekly.

All eggs produced during the experimental period weighted to the

nearest gram to calculate the mean egg weight.

To determine egg mass from recording data of egg production and its weight at biweekly as the next form :

EM (egg mass) = EP (egg production) x EW (egg weight) .

To study the quality and characteristics, three eggs were selected per plot.

During the whole experimental period samples were collected at the last two days of each of the four week durations.

The eggshell quality parameters, the geometric mean diameter (Dg), sphericity

 (Φ) , volume (V) and surface area (S) of eggs sampled were determined.

Shell thickness of hen eggs were determined by a dial micrometer with an precision of 0.01 mm (Mitutoyo, 0.01 mm, Japan). For achieving the unit mass of the hens' eggs, they were determined by an electronic balance with an accuracy of 0.001 g. The geometric mean diameter (D) of the hen eggs was calculated thru usage the next relationship which viewed in Eq. (1) (Mohsenin, 1970)

 $Dg=(Lw2)^{1/3}$(1) where L is the length and W is the width (thickness) in mm.

The sphericity (Φ) is expressed by the ratio between the geometric mean diameter of egg sample to the length of the egg (Mohsenin 1970). The surface area of the chicken eggs was found by analogy with a sphere of the same geometric mean diameter, using the term which cited via Altuntas and Sekeroglu (2008).

Sa= π (Dg)².....(2) ϕ ={(LW2)^{1/3}/L}X100.....(3)

Where Sa is the surface area in mm and D is the geometric mean diameter in mm. Egg shape index is expressed as the ratio of the width of a sample of egg to the length of the egg (Anderson et al. 2004).

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The shell thickness was determined approving to Monira et al (2003). Eq. 2 and 3.

The volume of the egg was determined by two methods: (1). Measurement of the volume of eggs with a graduated measuring 100 ml volumetric flask and (2), using the method of Baryeh and Mangope (2003) by measuring the dimensions of length and width and calculated as follows:

Eggs were then cracked, eggshells, and yolks were removed and weighed. Eggshells were dried in air so as to establish the eggshell thickness (containing the membrane). In three points on the eggs (one point on air cell and the randomized two points of equator) employing a digital micrometer .Eggshells weighed using a 0.001g accuracy scale.

To obtain egg characters, yolks removed and weighed on the precision scale AD1000. Eggshells washed, dried for 48 hours, and weighed. Proportions of yolk and shell were achieved as the relation between the weight of each portion and egg weight, the calculation of albumen was determined by difference as following:

% albumen =

100 – (% yolk+% shell).....(5) The examined variables were volume, density, color and Haugh unit, while the egg property variables were shell thickness, percentages of yolk, shell, and albumen %).

Subsequently weight, eggs broken and put onto a glass surface, for determining

the height of thick albumen employing a micrometer. Albumen-height and egg weight were used in the estimation of Haugh units (HU) by the equation

 $HU = 100 \log (H + 7.57 - 1.7W^{0.37})$(6)

which: H is the albumen height (mm); and W is the egg weight (g).

Tibia of the right leg was separated, removal of flesh, and air dried (72 h) for obtaining of mean tibia weight and strength. Bone-breaking force is commonly applied as a response measure for assessing bone mineralization (Merklet, 1983).

Breaking force at the middle of the bone was obtained by a constituents tester (Instron 4411, Instron Corporation, Grove City, PA) using software version 8.09, a standard 50-kg load cell, and a modified shear plate (8 cm in length and 1 mm in width), as explained by Riczu et al., (2004). The digital force gauge (model: FGN-50) was used to measure breaking force and shear strength for eggshells and bones.

Statistical Analysis: Data set were presented by using General Linear Model procedure (GLM) in SPSS16.0. SPSS, (2007). Duncan's multiple range tests Duncan, (1955) was applied to separate means . Statements of statistical significance are based on a probability of p < 0.05. The following model was used to study the effect of treatments on the parameters investigated as follows :

 $Yij = \mu + Ti + eij$

Where Yij = an observation, μ = overall mean , Ti = effect of treatment (i=1,2....4) and eij = experimental random error .

RESULTS AND DISCUSSION

The effects by altering the particle sizes of limestone on egg production and egg

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weight during the investigational period are offered in Table (2).

Although there were significant higher egg production (69.20%, 68.40%) and egg weight (50.57, 50.78g) obtained from the fourth and third groups than in those from the first and second groups, but it was clear that were little effects due to particle size on those traits at those levels of production of Golden Montazah chickens.

These outcomes are in harmony with the findings of Phirinyane etal., (2011) who reported a significant (P<0.05) improve in egg production from 18-21 weeks of age and thereafter the production was remained constantly high .

Contrary to the current findings Cheng and Coon(1990) and Kuleile et al., (2009) finding no effect on egg production by using particle size of limestone.

Non- significant (P<0.05) improve in egg production reported by MC Daniel (1983), for chickens fed oyster shell from 21 to 30 weeks of age.

There was no effect on mortality rate during the experiment. 3-4% of the whole egg production during the investigational period were cracked and shell- less of the total egg production.

Diets containing fine limestone particles recorded a cracked or shell – less eggs, showing that smaller particle sources such as pulverized CaCo₃ pass quickly throughout the digestive tract, consequently the hen does not able to sufficiently extract sufficient calcium to meet its requirements as reported by Phirinyane et al., (2011).

Egg Quality: Limestone Particle size distribution affect on physical characteristics of egg length, Egg diameter (mm), egg volume (cm³), egg weight (g) and density (g/cm^3) are presented in Figs 1-5 and Table (3).

No significant differences appeared in these characteristics (P<0.05), which agreed with Wang et al., (2014) who found that egg shape index or yolk height didn't influenced by using limestone particle sizes.

Variations in shell weight were not significant. A significantly higher shell weight (5.65, 5.55) and shell thickness (0.47, 0.46) occurred in eggs from the fourth and third groups (100%, 80% pulverized limestone substituted by coarse particles) than in those from the first treatment consumed a diet with (100%) fine particles Fig 6.

Krasucki et al., (2002) confirms that calcium carbonate in the diet increased the thickness of eggshells at 33 weeks of age with 60-80% limestone replacement, which agreed with Koreleski and Swiatkiewicz, (2004) during the period of peak egg production in chickens.

It seems from Table (4) and Fig.7 that different ratios of limestone particle size influence the breaking force of the eggshell. A significantly (P<0.05) higher shell breaking force (31.69, 30.19) were observed in eggs from the fourth and third treatment which received diets with (100%, 80% large particles substituted by fine limestone) than others from the first treatment fed a diet with (0%) coarse particles .

The breaking force of the shell (P<0.05) and the appearance of eggs based on improved albumen height and Haugh units (P<0.05) were improved by using large particle of limestone as consistent with (Korelesk and Swiatkiewicz 2004 ; Skrivan et al., 2010 ;Wang et al., 2014).

The grade of egg yolk pigmentation was advanced under the effect of limestone grind in the diet, which attributed to the

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existence of larger particles in the gizzard caused better destruction of feed cells and more effectual release of xanthophyll specially during the phase of high laying.

As shown in Table (4) and Fig.8 the larger particles of limestone improved (P<0.05) the breaking force of fresh tibia at 80% and 100% replacement of fine limestone when contrasted with the control diet . These outcomes are in accordance with the finding of Koreleski and Swiatkiewicz (2004) showed that large particles of limestone improved tibial strength. Wang et al., (2014) concluded that the breakage force of the tibias (P<0.05) , were increased by addition the limestone rather oyster shell but not the other tibial characteristics.

Contrary to the current results, De Witt, et al., (2009) showed that large particles of limestone gave no valuable influence on egg production and egg shell quality characteristics of laying hens at later stage (>54 weeks of age) of production. At 24 weeks of age Phirinyane et al., (2011) reported that the proportion of fine (<1.0 mm) and granular (> 2.0 - 3.8 mm) does not effect on egg production and egg shell quality.

Overall, the findings achieved from the current experiment concluded the possibilities of promoting egg shell quality and the best tibia signs from using limestone in large particle size as a Ca source in diets for chickens.

CONCLUSION

The current study concluded that the usage of large particles limestone as a Ca supplier in layers diet improvement eggshell and tibia bone qualities. Positive effects on eggshell quality by substituting 60 - 100 % of pulverized particles by particles of limestone larger Significantly higher shell weight (5.65 g) , shell thickness (0.46 mm) occurred in eggs from the fourth treatment of chickens than the others obtained of control group (5.33 g , 0.37 mm respectively). Eggshell breaking strength and tibia bone strength were increased at 80% and 100% levels of limestone substitutions.

Ingredients (%)	Exp.			
Corn	63.56			
Soybean meal 44%	24.65			
Wheat bran	2.55			
Limestone	6.97			
Salt	0.3			
Vit. &Min.	0.4			
Di calcium phosphate	1.51			
Dl- Methionine	0.06			
Calculated analysis :				
Protein	16.15			
Met. Energy(kcal/kg)	2714			
Calcium %	3.02			
Av. Phosphorus	0.409			
Lysine %	0.895			
Methionine	0.351			

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Table (1) :percentage and calculated compositions of the experimental diets .

Vitamin and mineral premix at (0.1~% Vit. And 0.3% Min.) of the diet supplies the following per kg of the diet : Vit. A 14000000 IU , Vit. D_3 3000000 IU , Vit. E 8000 mg , Vit K 4000 mg , Vit B_1 3500 mg , Vit B_3 10000 mg , Vit. B_6 3500 mg , Vit. B_{12} 30 mg , Biotin 300mg , Pantothenic acid 20000 mg , Nicotinic acid 5000 mg , Folic acid 20000 mg , & Colene 500000 mg , Mn 100000 mg , Zn 80000 mg , Fe 50000 mg , Cu 12000 mg , I 1000 mg , Se 300 mg , Co 300 mg .

 Table (2): The effect of different particle sizes limestone on productive parameters of layers during the experimental period .

Parameters	Particle size ratios (Large : fine)			
	0%	60%	80%	100%
Egg production %	$60.68^{\circ} \pm 1.161$	62.17 ^b ±1.251	$68.40^{a} \pm 1.137$	$69.20^{a} \pm 1.804$
Egg weight (g)	$50.29^{a} \pm 0.123$	49.49 ^b ±0.156	$50.78^{a} \pm 0.223$	$50.57^{a} \pm 0.103$

a,b. Mean within a row for each item with different superscripts are significantly different (P < 0.05).

Parameters	Particle size ratios (Large : fine)				
i ai aineters	0%	60%	80%	100%	
Egg weight (g)	$50.91^{ab} \pm 0.803$	$49.75^{\rm b} \pm 0.830$	$53.59^{a} \pm 1.144$	$52.42^{ab} \pm 0.919$	
Egg volume (ml)	49.67±0.187	47.67±0.860	49.77±0.810	49.40±0.761	
Egg length (mm)	54.39±0.417	53.69±0.486	54.56±0.635	54.31±0.325	
Egg diameter (mm)	41.49 ^{ab} ±0.212	40.85 ^b ±0.266	42.22 ^a ±0.380	41.73 ^{ab} ±0.294	
Egg density (g/cm ³)	1.04 ± 0.122	1.05 ± 0.008	1.07 ± 0.010	1.08 ± 0.033	
Shell weight (g)	5.33 ^{ab} ±0.077	5.17 ^b ±0.145	$5.55^{a} \pm 0.090$	$5.65^{a}\pm 0.145$	
Shell percentage (%)	10.93 ± 0.076	10.56±0.457	10.70±0.466	10.03±0.336	
Yolk percentage (%)	$30.25^{b} \pm 0.432$	$31.03^{ab}\pm 0.581$	$30.53^{b}\pm0.453$	$32.34^{a}\pm0.733$	
Albumen percentage (%)	58.81±0.490	58.41±0.788	58.76±0.722	57.61±0.619	
Yolk color	$6.80^{b} \pm 0.300$	$6.80^{b} \pm 0.439$	$6.82^{a} \pm 0.349$	$7.21^{a}\pm 0.486$	
Yolk height	17.15±0.172	16.66±0.189	16.73±0.223	17.08 ± 0.332	
Albumen height	$5.42^{ab} \pm 0.198$	$5.40^{\circ} \pm 0.167$	$5.94^b\pm\!0.258$	$7.11^{a}\pm 0.233$	
HU	$74.49^{ab}\pm 1.760$	72.73 ^c ±2.245	$86.70^{a} \pm 1.318$	$79.67^{b}\pm 1.360$	
Shell thickness (mm)	$0.37^{b}\pm 0.0149$	$0.43^{a} \pm 0.0121$	$0.47^{a}\pm 0.005$	$0.46^{a}\pm 0.016$	
Sharp end	0.338 ± 0.005	0.337±0.006	0.361 ± 0.007	0.358±0.012	
Equator	$0.372^{ab}\pm 0.003$	$0.359^{b}\pm 0.005$	$0.383^{a} \pm 0.008$	$0.352^{b}\pm 0.005$	
Blunt end	$0.351^{a} \pm 0.005$	$0.329^{b} \pm 0.009$	$0.353^{a} \pm 0.006$	$0.318^{b} \pm 0.006$	

Egg production, eggshell quality, limestone, particle sizes, hens, tibia bone

Table (3): The influence of limestone particle size on external and internal egg quality characteristics of Golden Montazah chicken at peak production.

a,b,c Mean within a row for each item with different superscripts are significantly different(P<0.05)

Table (4):	The influence of	of limestone par	ticle size on s	hell and tibi	ia breaking	strength
	of Golden Mor	ntazah chicken	during the exp	perimental j	period.	

Parameters	Particle size ratios (Large : fine)			
	0%	60%	80%	100%
Shell breaking force (N)	29.47 ^b ± 1.176	$27.23^{b} \pm 1.011$	30.19 ^{ab} ±1.380	$31.69^{a}\pm 2.009$
Tibia breaking strength (N)	252.83 ^b ±21.986	$276.87^{b} \pm 20.633$	298.5 ^b ±12.701	$350.67^{a} \pm 20.718$

a,b, Mean within a row for each item with different superscripts are significantly different

 $(P\!\!<\!\!0.05)$.

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Fig(1):The effect of Particle size ratios (Large : fine) on Egg length (mm)



Fig(2):The effect of Particle size ratios (Large : fine) on Egg diameter (mm)



Fig(3): The effect of Particle size ratios (Large : fine) on Egg weight (g)



Fig(4):The effect of Particle size ratios (Large : fine) on Egg volume (ml)



Fig(5): The effect of Particle size ratios (Large : fine) on Egg density (g/cm3)



weight (g)



Fig(7):The effect of Particle size ratios (Large : fine) on Shell breaking force (N)



Fig(8):The effect of Particle size ratios (Large : fine) on Tibia breaking strength (N)

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الملخص العربي

دراسة تأثير إستبدال الأحجام الجزيئية المختلفة للحجر الجيري في علائق دجاج المنتزه الذهبي البياض على إنتاج البيض وجودة كلا من القشرة وصلابة العظام خلال ذروة الانتاج

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أجريت هذه الدراسة بمحطة بحوث الدواجن بالفيوم لمعرفة تأثير إستبدال الأحجام الجزيئية المختلفة للحجر الجيري كمصدر للكالسيوم في علائق دجاج المنتزه الذهبي البياض من عمر 32 – 44 اسبوع وتأثير ذلك على إنتاج البيض وجوده القشرة وكذلك درجة صلابة كلا من قشرة البيض وعظمة الساق.

تم إستخدام أقطار مختلفة من الحجر الجيري منها الناعم بحجم جزيئات أقل من 0.6 مم ، وكبير الحجم (أكبر من 2.8 مم وأقل من 4.5 مم) . تم استبدال الحجر الجيري الخشن بنسب صفر % ، 60% ، 80% ، 100% من الحجر الجيري الناعم) . توصلت النتائج الي زيادة معنوية في قوة صلابة قشرة البيض (31.69 نيوتن) ووزن القشرة (5.65 جم) وأيضا سمك القشرة (0.46 مم) للعينات المتحصل عليها من المعاملة الرابعة عن غير ها من مجموعة الكنترول (29.46 نيوتن ، 5.33 جم ، 0.37 مم) علي التوالي . وبالنسبة لقوة صلابة قوتن) . أظهرت النتائج زيادة معنوية (35.66 نيوتن)للمجموعة الرابعة مقارنة بالمجموعة الاولى (252.82 نيوتن) .

وتوصي النتائج المتحصل عليها أن إستخدام الحجر الجيري بجزيئات كبيرة الحجم أدت إلى زيادة في كُل من صفات جودة القشرة (سمك القشرة ووزن القشرة ودرجة الصلابة) هذا بالإضافة إلى زيادة قوة صلابة الساق في المعاملات المقدم لها الحجر الجيري بأحجام جزيئية كبيرة عن مجموعة المقارنة وبالتالي هناك إمكانية لإستبدال الحجر الجيري الناعم كمصدر للكالسيوم بجزيئات كبيرة الحجم بنسب إستبدال من 60% إلى 100%مع تحسين جودة كلا من قشرة البيض وصلابة عظمة الساق.

الكلمات الدالة : إنتاج البيض ،جودة القشرة ، الحجر الجيري ، دجاج ، حجم الجزيئات ، عظم الساق .