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# EFFECT OF EGG SHELL DIET ON PLASMA CALCIUM AND BONE HEALTH OF CHICKS

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## **Abstract**

The primary component of egg shell is calcium carbonate, and eggshell is used as a calcium supplement. The aim of current study designed to calculate the effect of egg shell diet on plasma calcium and bone health of Desi chicks. Desi chicks were divided into three groups, one group is control group, and second group is experimental group for 30% egg shell diet third experimental group for 50% egg shell diet. After completed experiment was conducted for 1 month body weight was calculated. Present study of results was showing Blood sample and calcium was calculated significant difference (P<0.05) increase in the body weight of egg shell treated group. (Fed 30% egg shell diet & fed 50% egg shell diet). The supplementary egg shell diet treated groups showed significant increase (P<0.05) in Plasma calcium. Histopathological studied of tibia and femur bone was conducted. Perichondrium and Osteoblasts spaces occur in treated group. No mortality was observed in chicks and other parameters were normal compared to control group and show positive result in plasma calcium parameter in chick after giving different doses of egg shell diet 30% and 50%.

Key words: Plasma calcium, Tibia Bone, Femur bone, Egg shell, Perichondrium, Osteoblasts

1. Introduction: In Pakistan, poultry goods, in particular eggs and egg products are essential components of human meals and nutritious food items. They are abundant in selenium, phosphorus, and protein, riboflavin, choline, and vitamin B12, pantothenic acid, zinc, folic acid, and vitamins [1] Chick's eggshell is a domestic waste product that comes from places like hatcheries, poultry farms, egg manufacturing

facilities, residences, and dining establishments. The primary component is calcium carbonate. Collagen is a component of the eggshell membranes that may be removed and used for a variety of things, including the biochemical, pharmaceutical, food, and cosmetics sectors [2]. To increase the safety and quality of eggs, there is a tremendous interest in researching the various elements that affect cuticle and

eggshell quality. The cuticle qualities of eggshells, in particular, are very varied due to a variety of influences, such as Age, genetics, nutrition, and living conditions for hens [3]. In the metabolic processes of pullet chicks, calcium is essential. The blood of chickens and other animals contains calcium in three different ways: attached to plasma proteins, both free and attached to inorganic substances ionized or dissociated [4]. Bone is a complicated compound material made up of a matrix material, water, and a nanocrystal carbonated crystals phase. The bones' mechanical characteristics based on its geometrical distribution, mineralization level, porosity, and structural arrangement at various scales, as well as the characteristics of the components that make up bone [5]. Bone characteristics and development are also impacted by several aspects, including genetics, sex, nutrition, and environment [6]. The tibiae bone is one of the most mineralized bones in the skeleton and serves as a reliable marker of skeletal mineralization in general [7]. Egg Shell diet on plasma calcium and bone mineral density [8]. The amount of mobility permitted by various husbandry practices' is further determined that morphometric measurements and the radiography density of a hen's bone's structural constituents are closely related to the bone's cracking strength [9]. This study is to check the

#### 2. Material and Methods

and bone health of chicks.

effect of egg shell diet on plasma calcium

#### **Experimental animals:**

12 Desi chicks were bought from the University of Veterinary and Animal Science they were 28-30 weeks old. The chicks were put in cages that had been cleaned, and the Minhaj University in Lahore's zoology department's animal housing is kept at a temperature of 25 degrees Celsius. One week following the modification, the chickens were separated into three groups. One of the groups is the control group, while the other two groups are used as the experimental groups. All groups' daily feed consumption, egg weight, eggshell weight, thickness, and relative density were monitored throughout the trial.

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# Assembling and preparing eggshells

Eggshells originated from a restaurant that served egg burgers Area Township near Hamdard chock Lahore. Eggshells were properly cleaned with tap water after collecting, and they were then dried outside for a few days. Eggshells were prepared by grinded with a crusher after drying, and the powder was then saved.

#### **Dosing Treatment**

**Group1** (Control group) the control group is receiving Feeds 2, 100g daily basis.

**Group 2 (Experimental Groups)** the experimental group received 70g feed and 30 % egg shell diet daily basis.

**Group 3 (Experimental Group)** 50% of eggshell diets were administered as a therapy mixed with Feed 13 diet Eggshell was provided in order to assess the quantity and caliber of the eggs.

#### **Blood Sampling**

The time of dissection after completing 29 day's experimentation of the mean body weight was measured in each group of chicks. Blood samples will be obtained from the jugular vein of every chick using a non reusable syringe. Blood will be obtained in a (EDTA) test tube for the plasma calcium

#### Statistical analysis

test.

Data analysis by using Graph pad prism 5 Turkey's test and one way analysis of variance (ANOVA) is used to investigate data. When the P - value (p< 0.05) was obtained, the data was declared significant to check the effect of egg shell diet on plasma calcium and bone health of chicks.

#### 3. Results and Discussion

In order to estimate, the effect of egg shell diet on plasma calcium and bones health of chicks. The weight and mass of these chicks gently improved. All of the chickens were gaining weight. The hens' health was becoming better while still receiving a 30% eggshell diet and plain feed. When given ordinary feed and a diet that included 50% eggshells, diet is beneficial which increase body weight.

#### **Body weight**

Identical sized chicks were taken and separated into three groups. There were five chicks in every group. The initial body weight, final body weight and mean body weight increase of control, and egg shell diet group 20 weeks old desi chicks. There was partial weight gain in Experimental group 3 was given 50% egg shell diet vs control group simple feed 2. Equivalent amount of diet was given in every chick. The egg shell

treated group has final weight  $(0.8 \pm 0.1224)$ and initial weight  $(0.65 \pm 0.1)$  as compared to control group  $(1.1 \pm 0.169)$ . The weight and mass of these chicks are gently increased as shown in figure. The egg shell diet play important role to increase weight percentage in chicks [10]. Observed the egg shell diet and different levels of Mg show significance result increase yolk weight, body weight, albumen weight, and improve egg shell quality and increase plasma calcium level in blood, bone health improvement in chickens. Egg shell treated groups 30% and 50% compared to control group show significant increase in weight gain  $(0.25 \pm 0.111)$  [11].

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#### Plasma calcium

Plasma calcium show significant increase (P<0.05) in egg shell treated groups 30% and 50%. Egg shell combination of different dose groups 30%  $12.248 \pm 0.3454$  and 50%  $11.484 \pm 0.4875$  of egg shell diet compared to control group  $16.184 \pm 1.1745$  in 20 weeks old chicks. [12] Observed the effect of egg Shell diet on plasma calcium and bone mineral density. There is no significant improvement in bone quality, egg quality and plasma calcium. The lack of changes in plasma calcium levels during the entire laying cycle. This change in the egg shell diet most likely contributed to the elevated levels in plasma calcium. The [13] observed eggshell flour has an effect on blood calcium levels in pregnant mice, saw major differences in blood calcium levels giving eggshell flour for 7 days at doses of 5.4 mg/dl, 10.8 mg/dl, and 21.6 mg/dl, as well as a standard calcium powder tablet, in raising blood calcium levels. In current

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study plasma calcium show significance in EG2 ( $12.248 \pm 0.3454$ ) as compared to control group  $16.184 \pm 1.1745$  as shown in table 4.2 [14] analyzed the dietary containing at least 50 % large calcium sources had positive effect on mineral contents of tibia. Eggshell die t 75% show positive effect on blood plasma calcium and tibia bone in chickens.

# **Bones Histology**

The administration of the Feed 13 + 30% egg shell diet, the EG1 increased bone weight in ion evaluation the CG1. G2 were feed 13 + 50% egg shell diet, and after that, when compared to the CG and E1G1, they acquired weight in the bones. When bone histology of hens all group (CG, EG1 and EG2) were done then significance were observed. The histology of tibia and femur bones at Magnification 10x Tibia bone of hen Experimental group two show in (figure 9) chondrocytes, and collagen fibers and experimental three show group Perichondrium as compared to control group as shown in (figure 10& 8). In [15] observed that increase in weight and affecting tibia and significantly increased egg weight, bone mineral density, tibia bone endure impact on calcium source like egg shell [16]. Bones (femur, tibia, and metatarsus) were analyzed for bone mineral substance, thickness, and flouting power femur bone. The rate of calcium utilization in eggshell was higher in the cage system than on the floor device. Egg shell had Plasma calcium were significantly increased, Found considerable difference in 12 weeks old chicks treated with egg shell diet at different doses level

30% and 50%. As a results increase tibia bone density, at dose 50% and reduce the bone mineralization density in 30% egg shell diet group [17]. Leg The management of the feed 13 +30 % egg shell diet, the EG1 increased bone weight. In comparison to the CG1 and EG2, The EG2 were feed 13+50% Egg shell diet, and after that, they increase bone weight, however it was less than that what the EG2 and acquired on a 30% egg shell diet and more than what the CG1 had increased. [18] Observed calcium intake had no effect on the characteristics of the tibia, productivity, or reproductive efficiency 4% calcium in their feed and that substituting oyster shell for 40% of the coarse limestone had no effect on shell development, quality, or efficiency.

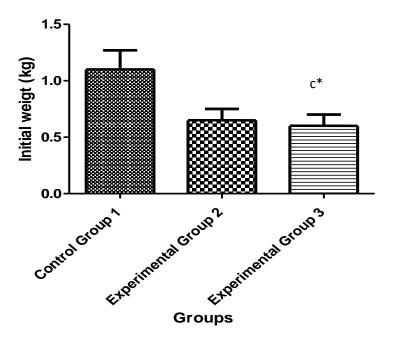
. In comparison to the CG1 and EG2, The EG2 were feed 13+50% Egg shell diet, and after that, they increase femur bone weight, however it was less than that what the EG2 and acquired on a 30% egg shell diet and more than what the CG1 had increased femur bone weight. In control group femur bone shows Osteoblasts evaluation as shown in (figure 5). The femur bone of experimental group 30% egg shell treated group show Perichondrium that is represent (p) as shown in (figure 6) and experimental group 50% egg shell treated group Cf represents collagen fibers and C represents chondrocytes as shown in (figure 7). Tibia and femur bone show Perichondrium and Osteoblasts spaces occur in treated group. compared to control group and show positive result in chick after giving different doses of egg shell diet 30% and 50%.

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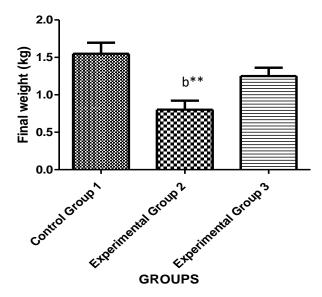
Table 1: Effect of egg shell diet (30% egg shell, 50% egg shell) at initial body weight, final body weight, and weight gain in 20 weeks old desi chicks. Mean  $\pm$  SEM is expressed values.

Groups	Initial weight	Final weight	Weight Gain
Control group 1	1.1 ± 0.169	$1.55 \pm 0.0145$	$0.45 \pm 0.05$
Experimental group 2	$0.65 \pm 0.1$	0.8 ± 0.1224 b**	$0.25 \pm 0.111$
Experimental group 3	$0.625 \pm 0.125$	$1.25 \pm 0.111$	0.75 ± 0.111 c**

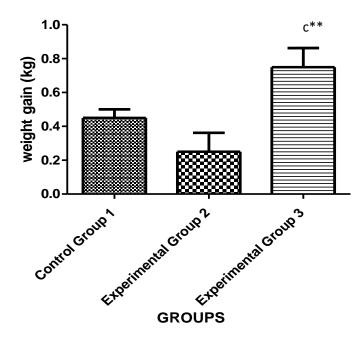
**a**= Control group Vs Experimental group **b**= simple diet + 30% egg shell powder **c**= diet + 50% egg shell powder. P < 0.05\* P < 0.01\*\*



**Figure 01:** Effect of egg shell powder diet 30% and 50% of initial weight in 20 weeks old desi chicks. Mean  $\pm$  SEM is used to precise data. There is a substantial similarity in the final weight of experimental group 2, (simple feed +30% egg shell diet) experimental group 3 (*simple* diet + 50% egg shell diet) compared to control group.



**Figure 02:** Effect of egg shell powder diet 30% and 50% of final weight in 20 weeks old Desi chicks. Mean  $\pm$  SEM is used to precise data. There is a substantial similarity in the final weight of experimental group 2, (simple feed +30% egg shell diet) experimental group 3 (*simple* diet + 50% egg shell diet) compared to control group.



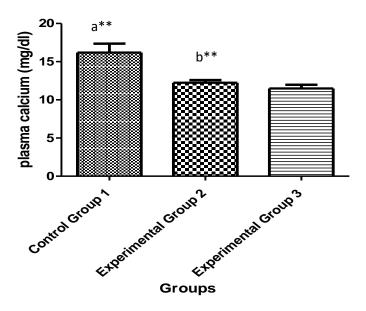
**Figure 03:** Effect of egg shell powder diet 30% and 50% of weight gain in 20 weeks old desi chicks. Mean  $\pm$  SEM is used to precise data. There is a substantial similarity in the final weight of experimental group 2, (simple feed +30% egg shell diet) experimental group 3 (*simple* diet + 50% egg shell diet) compared to control group.

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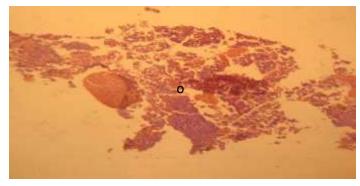
**Table 2:** Effect of egg shell diet (30% egg shell, 50% egg shell) on plasma calcium in 20 weeks old desi chicks.

Control Group 1	Experimental Group 2	Experimental Group 3
16.184 ± 1.1745a**	12.248 ± 0.3454b**	$11.484 \pm 0.4875$

**a**= Control group Vs Experimental group **b**= simple diet + 30% egg shell powder **c**= diet + 50% egg shell powder. P < 0.05\* P < 0.01 Turkeys test



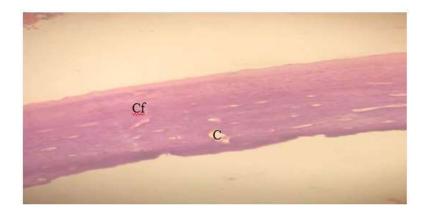
**Figure 04:** Effect of egg shell powder diet 30% and 50% of plasma calcium in 20 weeks old desi chicks. Mean  $\pm$  SEM is used to precise data. There is a substantial similarity in experimental group 2, (simple feed +30% egg shell diet) experimental group 3 (*simple* diet + 50% egg shell diet) compared to control group.



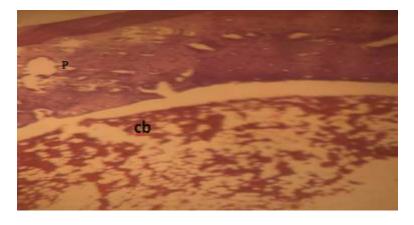
**Figure 05:** photomicrograph of 12 weeks old desi chick of Femur Bone Osteoblasts Evolution in control group.



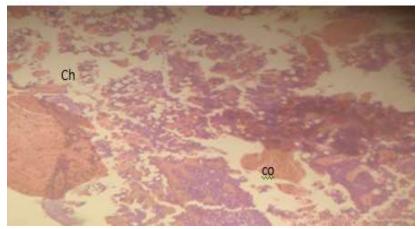
**Figure 6:** photomicrograph of femur bone EIG1Perichondriumof 12 week's old desi chick in egg shell treated group



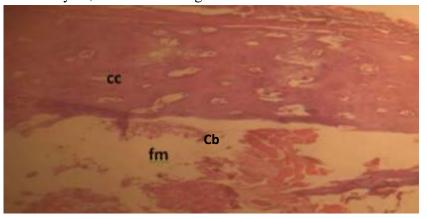
**Figure 7:** photomicrograph of femur bone of E2G2 of 12 week's old desi chick's eggshell treated group. Cf represents collagen fibers and C represents chondrocytes.



**Figure 8:** Photomicrograph of 12 weeks old chicks showing tibia bone control group (CB) chondroblasts, (P) Perichondrium evolution in control group.



**Figure 9:** Magnification 10x Tibia bone E1G1 of 12 weeks old desi chick of in egg shell treated group. Ch show chondrocytes, Co shows collagen fibers.



**Figure 10:** Photomicrograph of 12 weeks old chicks showing tibia bone E2G2 group shows (FM) fibrous matrix, (CB) chondroblasts, (CC chondrogenic cells.

## 4. Conclusion

Poultry farming in Pakistan is a significant and growing sector of the agricultural sector. Poultry needs rising day by day in the form of meats, eggs. The finding of this current study purpose that the effect of egg shell diet on plasma calcium and bone health of chicks in 12 weeks old chicks. The chicks fed a 30% egg shell diet and 50% egg shell diet is beneficial and to get sufficient amount of calcium. Egg shell diet has increase body weight of the chicks as compared to control group. Egg shell diet show significantly increases in Plasma calcium. Egg shell diet

can be good for weight gain, bone health in chicks

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