# Effect of Cage Densities and Betaine Supplementation on Nitrogen Retention in Quails

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## **ABSTRACT**

Application of a high cage density would save the space but has many negative impacts such as causing stress, leads to digestion, absorption, acid-base balance and osmotic disturbances. Betaine is a compatible osmolyte which exerts beneficial functions in assisting the birds encountering the osmotic stress and has been shown to improve nutrient digestion and absorption. The objective of the experiment was to investigate the effect of cage density and betaine supplementation on N (N) retention in Japanese quails (Coturnix coturnix *japonica*). A total of 408 laying quails aged 23 weeks (average body weight of 154.6±5.0 g) was used in this experiment and subsequently 48 quails were used in the measurement of N retention. The experiment was designed to completely randomized design of factorial 3×2 with 4 replicates. The quails were allocated to 24 cages with three cage densities consisted of 40, 45, and 50 birds/m2 equivalent to 250, 222 and 200 cm<sup>2</sup>/birds, respectively, which were represented by 15, 17, and 19 birds per cage (cage area of 0.375 m<sup>2</sup>). The birds were fed a diet without or with supplementation of 0.12% betaine. The N retention was measured by a total collection method using 2 quails from each replicate. The data were subjected to analysis of variance, and when the treatment indicated significant effect, it was continued to Duncan's test. There was no interaction between cage density and betaine supplementation on N retention. Furthermore, increasing cage density from 40 to 50 birds/m<sup>2</sup> did not decrease N consumption, excretion and retention, indicating that N was absorbed according to the birds' requirement. Betaine supplementation did not affect N consumption, excretion and retention. It can be concluded that cage density 50 birds/m<sup>2</sup> can be applied to quails, while betaine supplementation did not affect N retention in quails.

**Keywords:** Quails, cage density, betaine, nitrogen retention

#### **INTRODUCTION**

Cage density is an important environmental factor in livestock production (Esen *et al.*, 2006). High density will result in the decrease feed consumption (Seker *et al.*, 2009), increase the temperature and humidity in the cages due to high evaporation processes (panting) (Baziz *et al.*, 2010; Adebiyi *et al.*, 2011). Panting due to the difficulty of excessive heat expenditure leads to excessive energy use and the birds may subjected to heat stress (Faitarone *et al.*, 2005). This will decrease performance, immunity, increase ammonia, cannibalism and mortality (Heckert *et al.*, 2002; Askar and Assaf, 2004; Dhaliwal *et al.*, 2008).

Heat stress will decrease the efficiency of digestion, absorption and transport of nutrients that result in decrease egg production (Miles, 2001). Along with rising the body temperature, water consumption will increase to maintain body fluid balance. Consequently, the feed consumption and feed efficiency decreases (Ozbey *et al.*, 2004). The high temperatures also decrease the blood supply to the digestive tract because more blood supply

is directed to the respiratory tract, causing the decrease in nutrient absorption (Latipudin and Mushawwir, 2011). Prolonged stresses can lead to dehydration and disruption of digestion, absorption and nutrient metabolism and changes in blood electrolyte content, indicated by changes in acid-base balance and osmotic pressure of body cells (Rahardjo, 2012). Osmotic pressure causes disruption of water balance and intestinal cell volume as well as the structure and function of the small intestine cells that may affect nutrient absorption. The stress also affects the growth of small intestinal villi which became shorter (Sandikci *et al.*, 2004).

The addition of organic osmolite is expected to overcome the osmotic disturbance caused by density stress. Betaine is the most effective organic osmolyte to control osmotic pressure in cells compared with other organic osmolytes such as glycine, proline, glutamine and taurine (Hammer and Baltz, 2002). The accumulation of organic osmolytes in cells and cell organelles during osmotic stress can replace the inorganic ions and protect enzymes and cell membranes from inactivation by inorganic ions, thereby enabling cells to survive (Petronini *et al.*, 1992). Betaine also stimulates cell proliferation in intestinal cells, extending the epithelial cell wall and expanding the villi surface, thus may improve the nutrient absorption (Eklund *et al.*, 2005). The supplementation of betaine is expected to maintain the balance of electrolytes and osmotic pressure in the body cells, especially the digestive tract, thereby optimized the nutrient absorption. Therefore, the objective of the experiment was to investigate the effect of cage density and betaine supplementation on N retention in Japanese quails (*Coturnix coturnix japonica*).

### **MATERIALS AND METHODS**

A total of 408 23-week laying quails with average body weight of  $154.6\pm5.0$  g was used in this experiment and subsequently 48 quails were used in the measurement of N retention. The colony cages were used in this experiment with the size  $75\times50\times35$  cm (p × l × t) representing cage area of 0.375 m<sup>2</sup>. The quails were allocated to 24 cages in  $3\times2$  factorial arrangements with 4 replicates. The three cage densities consisted of 40, 45, and 50 birds/m<sup>2</sup> equivalent to 250, 222 and 200 cm<sup>2</sup>/birds, respectively, which were represented by 15, 17, and 19 birds per cage.

The basal diet, which was devoid of antibiotic and feed enzyme, was formulated to meet the nutrient requirement of laying quail according to the recommendation of the Indonesian National Standard (SNI, 2006). The nutrient composition of the basal diet can be seen in Table 1. The experimental diets were obtained by supplementing 0 (Control) and 0.12% betaine (Betaine) to the basal diet. During the experiment, diet and water were provided *ad libitum*.

**Table 1.** Nutrient composition of the basal diet

Nutrients	Content
Metabolizable energy (kcal/kg)	2800.00
Crude protein (%)	18.01
Crude fiber (%)	4.25
Crude fat (%)	4.86
Calcium (%)	3.40
Available phosphorus (%)	0.50
Lysine (%)	1.04
Methionine (%)	0.45

The experimental diets were applied for two periods of 28 days. At the and of dietary treatments, the determination of N retention was performed by a total excreta collection for 5 days. In total, 48 quails were randomly allocated to 48 individual cages (two quails per

treatment). Three individual cage areas (250, 222, and 200 cm<sup>2</sup>) were applied in the total collection according to the procedure outlined by Ratriyanto *et al.* (2017). The excreta were sun dried and milled through a 0.5 mm mesh screen. The N content was determined using the Kjedhal method and the N retention was calculated according to Grana *et al.* (2013).

The data were subjected to analysis of variance (ANOVA) to determine the effect of treatment and continued with Duncan's test for signifineant effect (Steel and Torrie, 1991).

#### RESULTS AND DISCUSSION

The result indicated no interaction between cage density and betaine supplementation on N consumption, excretion, and retention (Table 2). The cage density of 40, 45 and 50 birds/m² in without or with 0.12% betaine supplementation did not affect N consumption, excretion and retention, because the protein in the diet was adequate to meet the quail requirement. According to Indonesian National Standard (SNI, 2006) the protein requirement for laying quails is 17% while the experimental diets contain 18% protein. The high cage density can cause the increase of temperature inside the cage (Johnson, 1987), but the stress generated when the cage density reaching 50 birds/m² was still acceptable by the quails. Thus, the betaine supplementation did not affect N digestion and absorption, possibly because betaine has not functioned optimally as an organic osmolyte to play a role in maintaining the water balance in the body.

**Table 2.** Effect of cage densities and betaine on nitrogen retention

Treatments		N Intake	N Excretion	N Retention	N Retention
		(g/bird)	(g/bird)	(%)	(g/bird)
Cage density	× betaine				
Density	Suplementation				
40	Control	1.44	0.64	54.72	0.79
	Betaine	1.49	0.76	48.67	0.73
45	Control	1.36	0.63	53.38	0.73
	Betaine	1.52	0.69	54.04	0.82
50	Control	1.52	0.68	55.08	0.84
	Betaine	1.34	0.68	49.87	0.67
P value		0.78	0.21	0.22	0.39
Effect of cag	e density				
40	•	1.48	0.67	54.38	0.81
45		1.51	0.72	51.87	0.79
50		1.35	0.66	51.63	0.70
P value		0.09	0.30	0.56	0.20
Effect of beta	nine				
Control		1.43	0.68	52.25	0.75
Betaine		1.46	0.68	53.00	0.78
P value		0.57	0.94	0.75	0.57

Different cage density did not affect the N consumption, excretion, and retention (Table 2). The cages density of 40, 45 and 50 birds/m² still provide adequate space for movement, access to feed or water and enough access to oxygen. Previous study indicated that the density of 28 and 32 birds/m² did not indicated different feed consumption correlated with the digestible nutrients in quails (Akram *et al.*, 2000). Other observation showed that increasing the cage density in laying hens decreased feed consumption, and increased the mineral contents in excreta (Saki *et al.*, 2012). It is hypothesized that the effects of stress did not increase the levels of glucocorticoid hormones in this study, thus digestibility of protein are relatively constant between treatments. The glucocorticoid hormone especially corticosterone increases the level of amino acid in the blood (Ravindran *et al.*, 2006). The non significant effect of cage density on N retention is associated with the total plasma protein content in this study, which is not affected by the cage density. Azeem and Azeem

(2010) showed that the cages density up to 77 birds/m<sup>2</sup> did not affect the total blood protein content in quails.

Betaine supplementation did not affect N consumption, excretion, and retention (Table 2), indicated that the Control generated similar result compared with the Betaine. The N retention is influenced by N (protein) consumption and N content in excreta. According to Ratriyanto *et al.* (2014) the high content of N in quail excreta indicated the protein inefficiency. Ratriyanto *et al.* (2014) observed that supplementation of 0.14% betaine did not affect protein (N) consumption and N content in excreta. The supplementation of methyl group donors in the form of *DL*-methionine and choline to the basal diet, which was deficient in methionine and low in compatible osmolytes increased N retention in piglets. However, the supplementation of betaine to the diet containing adequate methionine and compatible osmolytes did not affect N excretion and retention (Eklund *et al.*, 2006).

### **CONCLUSIONS**

Increasing the cage density from 40 to 50 birds/m<sup>2</sup> and supplementation of betaine did not affect nitrogen retention in quails. Quails can be raised with the cage density of 50 birds/m<sup>2</sup>.

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