



## Performance and mineral metabolism of broiler replacing commercial diet by rice polish and supplementation of citric acid

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### Abstract

During an experimental period of 28 days citric acid (CA) was tested as a growth promoter instead of antibiotics with replacement of commercial diet by rice polish (RP). Newly 240 hatched broiler chicks (Cobb 500) distributed into eight dietary groups (3 replicate cages having 10 birds in each), 1=Control (commercial diet), 2=Commercial diet+0.5% CA, 3=5.0% RP, 4=5.0% RP+ 0.5% CA, 5=10.0% RP, 6=10.0% RP+0.5% CA, 7=15.0% RP, 8=15.0% RP+0.5% CA. Diets were supplemented by acid insoluble ash (1.0% Celite) as marker. At the end, blood sample was collected from all birds. Total ash, mineral content and density of tibia were determined. Final body weight (g/b) of chicks were 1655, 1733, 1642, 1694, 1618, 1656, 1613 and 1631 g, respectively ( $P>0.05$ ). Feed intake (g/bird) was 2359, 2419, 2432, 2433, 2524, 2494, 2519 and 2424 g, respectively ( $P>0.05$ ). FCR varied ( $P<0.05$ ) among the groups were 1.48, 1.44, 1.54, 1.49, 1.62, 1.55, 1.62 and 1.54, where better FCR was in CA groups comparison to non-CA groups. Retention of Ca, P and Mg increased in CA group's comparison to non-CA groups but replacement of 5.0% commercial diet (with or without CA) caused higher retention level. Higher dressing percentage observed in CA group (65.4, 65.9 for group 2, 4) comparison to non-CA groups (63.8, 63.9 for groups 1, 3). Bone mineral concentration (total ash, Ca, P and Mg) slightly increased in CA groups ( $P>0.05$ ). In general, replacement of a commercial diet by RP up to 15.00% would be possible maintaining growth performance of broiler where further supplementation of 0.5% CA showed more advantages by increasing mineral density of bone.

**Keywords:** : broiler, rice polish, citric acid, performance, mineral metabolism

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### Introduction

In Bangladesh total livestock production is 3931.37 lakh and total poultry production is 3379.98 lakh (DLS, 2018). For a long time sub therapeutic level of antibiotics are being used in broilers for improved for the improvement of body weight and feed efficiency (Giriprasad *et al.*, 1990; Miles *et al.*, 2006), but many of those develops resistant strain of pathogen and causes health hazard for human via food from animal origin (Khachatourians, 1998). Considering health hazard many alternatives like organic acids, probiotics, prebiotics, herbs and immune stimulants are suggesting by different legislation to provide safe, healthy food from animal origin. Citric acid is an organic acid increased the performance when using 0.5% level in mash diet of broiler (Islam *et al.*, 2008). It might be a useful additive instead of antibiotic growth promoters such as avilamycin and flavomycin, considering performance and health status of broilers (Chowdhury *et al.*, 2009a; Haque *et al.*,

2010). Not only the innate immunity (Chowdhury *et al.*, 2009b; Haque *et al.*, 2010) but also it enhances specific immunity by increasing antibody titre against New Castle Disease vaccine when added in broiler diet (Das *et al.*, 2012). Some study conducted in laboratory scale with mash diet showed excellent results for growth performance of broiler when citric acid was supplemented (Chowdhury *et al.*, 2009b; Haque *et al.*, 2010). Keeping in mind further study conducted by reducing protein and energy level and concluded that the addition of citric acid compensate the performance of broiler depressed due to lowering the nutrients in diet at certain level (Das *et al.*, 2012). A recent study observed that addition of 0.75% citric acid in a commercial diet suitable for growth, carcass traits, macro mineral digestibility and bone mineral density of broiler chicks (Islam *et al.*, 2012). But in commercial diet all the nutrients were well balanced to optimize the growth, so citric acid might not show its effect with full potentiality as well as increase the feed cost per unit weight

gain. Considering the points the study designed to replace commercial diet by rice polish considering lower nutrient content and availability but addition of citric acid to know the performance, mineral digestibility and their metabolism.

## Materials and Methods

### Birds, feeding and management

Two hundred and forty unsexed day old Cobb-500 broiler chicks were kept in 24 pens of 16 square fit with 10 birds per pen. Commercial broiler starter diet, but with 1.0 % Celite was added as a tracer considered as control diet (Table 1). Birds were divided into 8 dietary groups like 1=Control (commercial diet), 2=Commercial diet+0.5% CA, 3=5.0% rice polish (RP), 4=5.0% RP+0.5% CA, 5=10.0% RP, 6=10.0% RP+0.5% CA, 7=15.0% RP, 8=15.0% RP+0.5% CA. Feeding trial was conducted for a period of 28 days. The proximate components of the formulated diets were determined (Table 1) according to the method described by AOAC (1990).

The chicks were handled carefully according to the guidelines of the animal welfare committee of the Faculty of Animal Husbandry, Bangladesh Agriculture University. The birds were kept on deep litter. At the beginning, the room temperature was 34°C. This temperature was decreased to 22°C over the first 3 weeks. Feed was supplied and the amount of leftover feed was recorded when the birds were weighed each week.

### Sampling and analysis of feed and excreta

Feed and excreta sample were collected during the last week of the trial to determine dry matter, total ash, and acid insoluble ash, Ca, P and Mg. The amount of Ca, P, and Mg were determined by auto analyzer (Cobas Mira, Roche, Basel, Switzerland) using standard commercial kits. The digestibility of individual minerals and metabolizable energy was calculated using the following formula.

Apparent digestibility (%) =  $100 - [(\% \text{ indicator in feed} / \% \text{ indicator in excreta}) \times (\% \text{ nutrient in excreta} / \% \text{ nutrient in feed}) \times 100]$

### Determination of blood profiles related to mineral metabolism

Blood samples were drawn from birds during age of 28 days to determine mineral concentration (Ca, P and Mg). Immediately after collection, blood samples centrifuged at 1500 rpm for 10 minutes and serum transferred to a test tube for further analysis.

### Determination of mineral density of tibia

The tibias from each side of the sacrificed birds were chosen, excised from the fresh carcass, and flesh was removed. The tibia individually sealed in a plastic bag to minimize moisture loss. Forty (10 from each group) tibias from the right side were kept in the refrigerator at 4°C. Mineral content was measured using the peripheral quantitative computed tomography (QCT) method (Stratec XCT 960A, Pforzheim, Germany), scanning at 10, 50, and 90 percent longitudinal location of the tibia for total, trabecular and cortical bone of mineral density.

**Table 1:** Composition (g/100g) of diet in different experimental groups

Groups	1	2	3	4	5	6	7	8
DM (g/100 g)	88.00	87.56	88.10	87.67	88.20	87.76	88.30	87.87
Crude protein	23.00	22.89	22.50	22.44	22.00	21.91	21.50	21.40
Crude fibre	4.50	4.48	5.00	5.03	5.60	5.67	6.15	6.12
Ether Extract	7.00	6.97	7.05	7.02	7.10	7.06	7.15	7.12
Ash	8.12	8.08	8.34	8.30	8.56	8.52	8.78	8.73
Ca	0.96	1.01	0.88	0.90	0.90	0.89	0.81	0.85
P	0.58	0.60	0.54	0.54	0.65	0.68	0.67	0.68
Mg	0.29	0.30	0.31	0.32	0.34	0.34	0.36	0.37

1, Control (commercial diet); 2, Commercial diet + 0.5% CA; 3, 5.0% rice polish (RP); 4, 5.0% RP + 0.5% CA; 5, 10.0% RP; 6, 10.0% RP+0.5% CA; 7, 15.0% RP; 8, 15.0% RP+0.5% CA.

**Determination of dry matter, ash, Ca, P, and Mg of the tibia**

The tibia dried at 105°C for 48 h and placed in a desiccators and weight recorded. Total ash was determined by placing dried tibia in muffle furnace for 24 hours at 600°C. Percentage of tibia ash calculated according to the procedure of Al-Batshan *et al.* (1994).

**Statistical analysis**

Initially, the raw data was organized using the computer program Excel (Microsoft Corporation, Renton, WA) and then analyzed using the SPSS 11.5 (SPSS Inc., Chicago, IL). All data analyzed by 1-way ANOVA, and Duncan's multiple range test (Duncan, 1955) conducted to determine the differences among the treatment means (Steel and Torrie, 1980).

**Results**

**Growth performance of broiler**

Initial weight per bird was 59.2 g ± 0.6. Live weight gain does not hampered by the reduction

of nutrient content by replacing the commercial diet up to 15.00 % by rice polish (Table 2). Replacement of commercial diet reduces the crude protein and ether extract content of diet but the reduction compensated by the addition of CA reflecting similar weigh in all groups (P>0.05). Crude fiber content increased linearly in rice polish group but its adverse effect not reflected in the growth performance of broiler. It seems that feed intake were numerically highest (2524 g) in group 5 and lowest (2359 g) in control group (P>0.05). Feed conversion ratio was higher in the group where commercial diet replaced by rice polish by 10.0 and 15.0% (P<0.05) in group 5 and 7 respectively but was similar for the groups 6 and 8 where CA was added (P>0.05).

**Mineral content in blood**

Replacement of commercial diet by rice polish caused some variation of Ca, P and Mg concentration in blood serum but, there is a clear indication of increased their concentration in CA groups comparison to non-CA group of similar nutrient containing diet (Table 3).

**Table 2:** Growth performance of birds in different experimental groups

Groups	1	2	3	4	5	6	7	8
Initial weight (g/b)	59.2 ± 0.6	59.2 ±0.4	59.3 ±1.8	59.1 ±0.8	59.4 ±2.6	59.2 ±1.5	59.3 ±2.2	59.1 ±3.1
Weight gain (g/b)	1596 ±58	1674 ±58	1583 ±69	1635 ±80	1558 ±49	1597 ±73	1554 ±38	1571 ±83
Feed intake(g/b)	2359 ±5	2419 ±136	2432 ±159	2433 ±164	2524 ±166	2494 ±145	2519 ±158	2424 ±163
Feed conversion ratio	1.48 <sup>a</sup> ±0.09	1.44 <sup>a</sup> ±0.03	1.54 <sup>abc</sup> ±0.05	1.49 <sup>ab</sup> ±0.04	1.62 <sup>c</sup> ±0.08	1.55 <sup>abc</sup> ±0.02	1.62 <sup>c</sup> ±0.14	1.54 <sup>abc</sup> ±0.02

\*Mean ± standard error (n=30). <sup>a,b,c</sup>Means with different superscript in the same raw differ significantly ((P<0.05). 1, Control (commercial diet); 2, Commercial diet + 0.5% CA; 3, 5.0% rice polish (RP); 4, 5.0% RP + 0.5% CA; 5, 10.0% RP; 6, 10.0% RP+0.5% CA; 7, 15.0% RP; 8, 15.0% RP+0.5% CA.

**Table 3:** Ca, P and Mg content in blood serum of different experimental groups of birds

Groups	1	2	3	4	5	6	7	8
Ca (mmol/l)	1.95 ±0.26 <sup>cd</sup>	2.00 ± 0.28 <sup>d</sup>	1.62 ±0.24 <sup>ab</sup>	1.74 ±0.43 <sup>bc</sup>	1.47 ±0.19 <sup>a</sup>	1.49 ±0.27 <sup>a</sup>	1.84 ±0.42 <sup>bcd</sup>	2.29 ±0.30 <sup>e</sup>
P (mmol/l)	1.97 ±0.26 <sup>ab</sup>	2.00 ±0.26 <sup>a</sup>	1.69 ±0.24 <sup>cd</sup>	1.76 ±0.31 <sup>cd</sup>	1.57 ±0.14 <sup>d</sup>	1.59 ±0.14 <sup>d</sup>	1.59 ±0.22 <sup>d</sup>	1.81 ±0.20 <sup>bc</sup>
Mg (mmol/l)	0.74 ±0.10 <sup>a</sup>	0.79 ±0.08 <sup>a</sup>	0.61 ±0.08 <sup>b</sup>	0.64 ±0.13 <sup>b</sup>	0.58 ±0.08 <sup>bc</sup>	0.61 ±0.08 <sup>c</sup>	0.64 ±0.11 <sup>b</sup>	0.75 ±0.08 <sup>a</sup>

\*Mean ± standard error (n=30). <sup>a,b,c</sup>Means with different superscript in the same raw differ significantly ((P<0.05). 1, Control (commercial diet); 2, Commercial diet + 0.5% CA; 3, 5.0% rice polish (RP); 4, 5.0% RP + 0.5% CA; 5, 10.0% RP; 6, 10.0% RP+0.5% CA; 7, 15.0% RP; 8, 15.0% RP+0.5% CA.

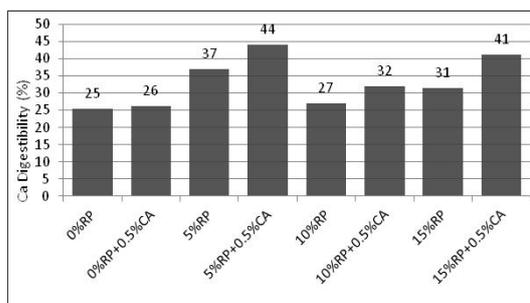
**Table 4:** Mineral density (mg/cm<sup>3</sup>) of bone due to dietary citric acid in different groups

Groups	1	2	3	4
Total density (10%)	218 ± 18 <sup>a</sup>	208 ± 26 <sup>a</sup>	200 ± 23 <sup>a</sup>	223 ± 26 <sup>a</sup>
Trab. density (10%)	135 ± 40 <sup>a</sup>	111 ± 40 <sup>a</sup>	106 ± 50 <sup>a</sup>	121 ± 47 <sup>a</sup>
Cort. density (10%)	295 ± 9 <sup>a</sup>	305 ± 42 <sup>a</sup>	297 ± 15 <sup>a</sup>	313 ± 35 <sup>a</sup>
Total density (50%)	435 ± 46 <sup>ab</sup>	398 ± 83 <sup>bc</sup>	374 ± 28 <sup>c</sup>	456 ± 32 <sup>a</sup>
Cort. density (50%)	618 ± 38 <sup>a</sup>	568 ± 102 <sup>b</sup>	558 ± 11 <sup>b</sup>	632 ± 23 <sup>a</sup>
Total density (90%)	229 ± 15 <sup>ab</sup>	241 ± 13 <sup>a</sup>	204 ± 17 <sup>c</sup>	224 ± 17 <sup>bc</sup>
Trab. density (90%)	153 ± 43 <sup>ab</sup>	177 ± 28 <sup>a</sup>	137 ± 33 <sup>b</sup>	134 ± 44 <sup>b</sup>
Cort. density (90%)	316 ± 30 <sup>a</sup>	320 ± 28 <sup>a</sup>	290 ± 10 <sup>b</sup>	326 ± 31 <sup>a</sup>

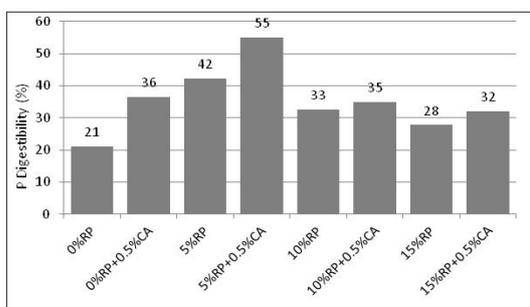
\*Mean ± standard error (n=30). <sup>a,b,c</sup>Means with different superscript in the same row differ significantly ((P<0.05). 1, Control (commercial diet); 2, Commercial diet + 0.5% CA; 3, 5.0% rice polish (RP); 4, 5.0% RP + 0.5% CA; 5, 10.0% RP; 6, 10.0% RP+0.5% CA; 7, 15.0% RP; 8, 15.0% RP+0.5% CA.

**Digestibility of Ca, P and Mg**

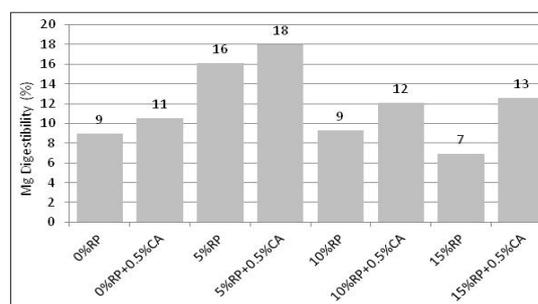
Digestibility of Ca, P and Mg found higher in CA groups in comparison to non-CA groups. Although, in different groups showed variation as the nutrient content slightly varied among the groups (Table 1 and Figures 1a, 1b and 1c)). Digestibility of Ca, P and Mg increased in CA groups comparison to non-CA groups, but was highest in 5.0% rice polish, which is also true for energy digestibility (Figure 1a, 1b, 1c and 2).



**Figure 1a.** Digestibility of Ca in different dietary groups



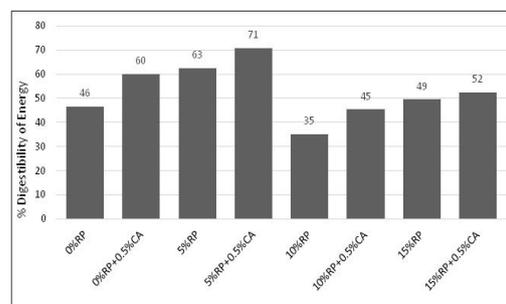
**Figure 1b.** Digestibility of P in different dietary groups



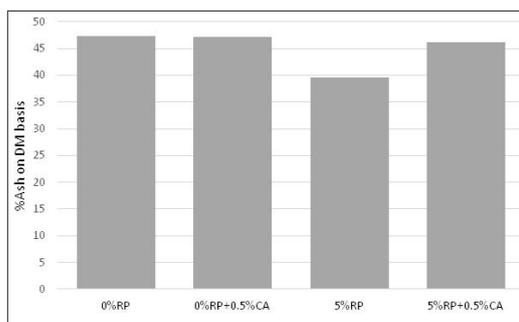
**Figure 1c.** Digestibility of Mg in different dietary groups

**Ash content and mineral density in bone**

Ash content in control group and further addition of 0.5 % CA showed no significant differences but when replaced commercial diet by 5.0 % level of rice polish showed slightly increased level of mineral content as well as mineral density in tibia (Figure 3 and Table 4).



**Figure 2:** Digestible energy in different dietary groups



**Figure 3:** Mineral content of bone (g/100gm DM) in different dietary groups

## Discussion

### Growth performance of broiler

Haque *et al.*, (2010) found that dietary supplementation of 0.5% CA increase weight gain, feed intake, feed conversion efficiency, tibia ash deposition, non-specific immunity and carcass yield of broiler. Chowdhury *et al.*, (2009b) found similar results when CA used as a feed additive instead of the antibiotic growth promoter Avilamycin. The results of this study are in an agreement with also the previous reports (Nezhad *et al.*, 2007; Moghadam *et al.*, 2006). Shahin *et al.*, (2009) found that addition of CA in commercial diet at 0.8% level increased 4.9 % weight in broiler which is also true in this trial that addition of CA increased live weight gain in comparison to non-CA groups. Most of the cases the lower limit of CA support the growth performance of broiler positively. So, compensation of growth due to reduction of nutrients by addition of rice polish was possible due to presence of CA at lower limit.

In this study feed intake was similar in all the groups either replacing by rice polish and addition of CA ( $P > 0.05$ ), which is in agreement with other findings (Shahin *et al.*, 2009; Islam *et al.*, 2010a). But some findings showed that feed intake increased than observed in this trial due to feeding rice bran, but they added 2.0%CA in broiler diet (Atapattu and Nelligaswatta, 2005).

Many researchers reported that addition of CA in broiler diet has positive effect on feed efficiency (Huifang *et al.*, 2005; Nezhad *et al.*, 2007), which also true for this trial. But some other research found numerically improvement of feed efficiency when added in commercial diet (Shahin *et al.*, 2009; Islam *et al.*, 2010a). Huifang *et al.*, (2005) demonstrated the highest feed conversion ratio with addition of 0.3% CA in growing chicks. So, it is clear that lower doses have positive effect on the feed efficiency as found in this study although

level of nutrient decreased due to replacing the diet by rice polish.

In this study CA containing group have higher carcass grade and it is also similar to the findings of Islam *et al.*, (2012). This result is also in agreement with other investigations (Ebrahimnezhad *et al.*, 2008). Compensation of growth performance in rice polish group observed due to the positive effect of CA, because its addition in animal diet may suppress pathogenic growth and improve digestion of nutrients, absorption of digested nutrients through gastro intestinal tract, improvement of mucosal immunity and topical effects on the intestinal brush border (Mroz, 2005). Reducing the bacterial burden due to addition of CA in feed improved gut health parameters significantly, this may be related to the increased availability of the nutrients.

### Mineral content in blood

When 15.0% rice polish added in diet replacing commercial diet but included 0.5% CA, Ca content in blood serum rose highest, nearly similar result was found in case of P and Mg content. From the report of Islam *et al.*, (2012) it observed that addition of CA influenced the availability of macro minerals, but this was not detected in the blood profile, which might be due to rapid turnover of those components, as other researcher found no changes of Ca, P and Mg in blood when added 3.0% CA in diet (Nourmohammadi *et al.*, 2010). But, other researcher found that 2.0% CA in broiler diet increased Mg level in the blood (Brenes *et al.*, 2003).

### Digestibility of Ca, P and Mg

Islam *et al.*, (2010b) reported that the addition of CA in the diet influenced the availability of minerals. But in this study used commercial diets contained standard level of minerals and replaced by rice polish but added CA at lower level, which is also supported by other researcher showing the Ca and P retention was 3.0 and 3.0% in 2.0% CA diet of broiler (Brenes *et al.*, 2003).

### Ash content and mineral density in bone

There are several findings that addition of CA in diet increased bone mineral content and bone strength in different levels (Atapattu and Nelligaswatta 2005; Chowdhury *et al.*, 2009b; Haque *et al.*, 2010; Islam *et al.*, 2010b; Islam *et al.*, 2012;). The increased mineral level in the bone is also related to the availability of the minerals which is responsible for bone formation (Islam, 2012; Boling *et al.*, 2001). In this study both bioavailability and its expression in blood as

well as in bone is clear that the metabolism of minerals related to bone formation is increased due to presence of CA in diet. So, replacement of commercial diet by rice polish would be possible maintaining the performance and increased mineral density in bone due to addition of CA in commercial diet which is replaced at 15.0% level by rice polish.

### Conclusion

From above findings it is clear that replacement of commercial diet containing 23.0% crude protein by 15.0% rice polish would be possible without hampering performance of broiler. But up to 5.0% level of rice bran increased digestibility, blood profile and mineral density of bone. For every case, 0.5% citric acid in diet enhance metabolism of Ca, P and Mg reflected by digestibility, blood profile and bone content of those component. In general, replacement of commercial diet by rice polish up to 15.0% would be possible maintaining growth performance of broiler where further supplementation of 0.5% citric acid caused more advantages for performance and mineral metabolism ( $P > 0.05$ ).

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### Conflict of interest

There is no conflict of interest among the authors.

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