Effects of Citric Acid and Multi Enzyme Supplementation on Growth Performance of Broiler Chicken Fed Diets Containing Rice Bran

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Abstract
Objective of this study was to determine whether adverse effects of diets containing high levels of rice bran (RB) on growth performance of broiler chicken could be overcome by supplemental citric acid (CA) and multi enzyme (ME) alone or in combination. Giving a completely randomized design in 2 x 2 x2 factorial arrangement, 168 broiler chicks in 56 floor pens received one of eight broiler finisher diets *adlibitum* during day 21-42. Treatment factors were two dietary RB; (15 or 30%), CA; (0 or 2%) and ME; (0 or 1000 FTU/kg) levels. RB (30%) did not reduce either the weight gain or live weight. In general, inclusion of CA and ME alone or in combination, did not correct the adverse effects of 30% RB on feed intake. There was a significant ($p<0.05$) RB x ME x CA interaction on feed intake and FCR. At 15% dietary RB level, inclusion of CA to enzyme free diet increased the feed intake significantly. Meanwhile, at 30% RB, inclusion of CA to a ME free diet significantly reduced the feed intake. There were significant two-way interactions on crop and gizzard pH. At 15% RB, birds fed diet with CA and ME, gave significantly better FCR compared to other three diets of that RB levels. At 30% RB, diet without ME but with CA, gave significantly lower FCR compared to other dietary combinations at same RB level. Financial analysis also showed a significant RB x ME X CA interaction. At 15% RB, combination of CA and ME was the best option. Birds fed ME free diet with 30% RB and 2% CA gave the lowest feed cost per kg of live weight gain. It was concluded that Citric acid (2%) in diets having 30% RB does not have adverse effects on growth performance, and minimizes the feed cost per kg of live weight gain.

Key words: Citric acid, Multi enzyme, Performance, Rice bran

Introduction
Rice bran is a locally available and cheaper cereal grain by-product, compared to many feed resource used in the feeds of poultry. The cost of poultry feed formulations can substantially be reduced if higher levels of RB could be used in their diets. However, RB cannot be included at higher levels in poultry diets due to the presence of anti-nutrients such as phytate. Inclusion of more than 20% RB in broiler diets reduced the growth performance (Farrell, 1994). Therefore, means of increasing the maximum inclusion level of RB are important to reduce the feed cost of poultry industry.

Phytate reduces the availability of phosphorus and a range of minerals, amino acids and the energy value. Increase of phosphorus and nitrogen excretion due to phytate is an environmental problems. It has been well established that the hydrolysis of phytate corrected those adverse effects. Supplementation of diets with Phytase (Martin *et al.*, 1998) and CA (Islam *et al.*, 2012) has been reported to correct those adverse effects. Wickramasinghe *et al.*, (2009) have shown that inclusion of CA alone did not mitigate the adverse effects if RB level is high. Objective of this study was to determine the effects of CA and ME (containing phytase, glucanase , amylase, protease , pectinase, lipase and xylanase) alone or in combination on growth performance of broiler chicken fed diets containing either 15 or 30 % RB.

Materials and Methods
A total of 168 broiler birds (Strain Cobb) were brooded up to 10 d under normal incandescent light (40 W, Osram). On 20 d, birds were weighed and allocated into

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56 experimental cages (3'length X 2'width) by balancing weight. Three birds were allocated in each of the experimental unit. The experiment followed a complete randomize design in 2 x 2 x 2 factorial arrangement, with 7 replicate cages per each treatment combination. Treatment factors were two levels of RB; (15% and 30%), 2 levels of CA; (0 and 2%) and 2 levels of exogenous ME; (0 and 1000 FTU/kg). Astro-Zyme was the ME mixture which contained Phytase, Glucanase, Amylase, Protease, Pectinase, Lipase and Xylanase. Except for non phypate phosphorus (NPP), all diets met or exceeded the nutrient requirements as set out by NRC (1994). The dietary NPP level was 0.25%. Ingredient composition and calculated nutrient composition are given in Table 1. Each experimental unit was provided with a feeder and a bell shaped drinker to ensure *ad libitum* feed and water. Paddy husk was used as the litter material. At 41d, one bird from 6 replicate cages each was humanely slaughtered for the carcass evaluation. Parameters measured were dressed weight, weights of the gizzard, liver, pancreases, pH of the crop and gizzard contents were determined. Tibiae were analyzed for fat free tibia ash. Data were analyzed as a completely randomize design in factorial arrangement, using SAS.

Results and Discussion

Table 1. Ingredient composition and the calculated nutrient composition of the experimental diets

Ingredients (%)	Dietary RB Level	
	15	30
Yellow maize	45.3	32.6
Rice bran	15	30
Soya	25.1	23.87
Coconut oil	4.4	4.7
Fish meal	4.9	4.4
DCP	0.95	0.9
Shell grit	0.9	1
Fiber	1	0
DL Methine	0.04	0.03
Salt	0.25	0.25
Vit mix	0.25	0.25
Fiber Degrading Enzyme + Phytase	-/+	-/+
Citric Acid	0/2	0/2

Contrary to Islam *et al.* (2012), CA had no positive effects on either the live weight or the weight gain (Table 2). However, our findings are in line with (Woyengo *et al.*, 2010) who found that CA supplementation had no positive effects on the performance of broilers. Importantly 30% RB had no adverse effects of live weight or weight gain. Contrary to Gallinger *et al.* (2004), 30% RB did not reduce either the weight gain or live weight of the birds. Steyaert *et al.* (1989) have also suggested that RB up to 30% in mash form had no adverse effects on performance of broiler chicken.

Radcliffe *et al.* (1998) have shown a negative interaction between phytase and CA on mineral utilization of poultry and growth performance of pigs, respectively. Results of this experiment showed that there was a significant RB x ME x CA interaction on feed intake and FCR. At 15% dietary RB level, inclusion of CA to enzyme free diet increased the feed intake significantly from 2455 g to 2615 g. Meanwhile, at 30% RB, inclusion of CA to a ME free diet significantly reduced the feed intake from 2321g to 2134 g. This observation suggests that when no ME is added, CA has positive effects on feed intake in diets containing 15% RB whereas inclusion of CA at 30% had adverse effect on the feed intake.

At 15% RB, addition of CA to a diet with ME significantly reduced the feed intake from 2358 g to 2206 g. In contrast, at 30% RB, inclusion of CA to a diet with ME did not change the feed intake. The interactive effects of treatments on feed intake may be related to the significant two-way interactions in crop and gizzard pHs. However, inclusion of CA, ME alone or in combination did not correct the adverse effects of 30% RB on feed intake. Benefits could be achieved if CA is added to a ME free diet having 15% RB.

The feed conversion ratio results revealed that the conversion of nutrients into live weight gain followed a different trend. At 15% RB, birds fed diet with CA and ME, the diet which recorded the lowest feed intake, gave significantly better FCR compared to other three diets of that RB level. At 30% RB, diet without enzyme but with CA, gave significantly lower FCR compared to other dietary combinations at same RB level. Interestingly, that was the diet recorded the lowest feed intake at 30% RB level. Furthermore, FCRs of the birds fed 15% RB with CA and ME (1.98) and those fed 30% RB with CA without ME (1.93) were not statistically different.

Financial analysis showed a significant RB X ME X CA interaction. At 15% RB level, inclusion of CA without ME or ME without CA had no significant beneficial effects on feed cost per kg of live weight gain. At 15% RB, combination of CA and ME was the best option. In contrast, at 30% RB level, inclusion of CA without ME significantly reduced the feed cost per kg of live weight gain, compared to the diet without CA and multi enzyme alone or in combination. The feed cost per kg of live weight gain of the birds fed former diet gave significantly lower FCR compared to all other dietary combinations. Therefore, as far as feed cost is concerned, it can be concluded that the use of 2% CA with 30% RB without ME lower the feed cost per kg of gain.

Relative weight of the gizzard and the tibia ash content were significantly influenced by RB x CA interaction (Table 2).

Table 2. Effects of citric acid and phytase supplementation on growth performance of broiler chicken fed either 15 or 30% dietary rice bran

Parameter	Rice bran (%)								Pooled SEM
	15				30				
	Enzyme								
	-		+		-		+		
	CA (%)								
	0	2	0	2	0	2	0	2	
Weight (42 d)	1824	1841	1756	1820	1824	1840	1830	1808	34
Weight gain (21-42d)	1113	1122	1031	1111	1099	1108	1103	1089	36
Feed intake	2455	2515	2358	2206	2321	2134	2305	2337	16
FCR	2.21	2.25	2.32	1.98	2.14	1.93	2.09	2.17	0.07
Gizzard[1]	4.1	3.6	4.0	3.6	3.6	4.3	3.7	3.8	0.27
Tibia ash (%)	34.1	35.0	34.1	34.3	33.5	36.7	33.3	41.5	1.6
Crop	4.2	4.1	4.3	4.2	4.1	4.3	4.1	4.3	0.10
Gizzard	5.7	5.0	5.0	5.2	5.1	5.1	5.0	5.4	0.15
Cost of feed (Rs)	76.3	78.3	76.7	78.7	71.0	71.0	73.0	71.4	73.4
Feed cost (Rs)	187	196	180	173	164	155	164	171	1.25
Feed cost/ kg live weight gain	169	176	177	156	152	141	149	159	5.8

Parameter	ANOVA						
	Main effects			Interactions			
	RB	E	CA	RB*E	RB*CA	E*CA	RB*E*CA
Live weight 42 d	NS	NS	NS	NS	NS	NS	NS
Weight gain (21-42d)	NS	NS	NS	NS	NS	NS	NS
Feed intake	***	***	***	***	NS	NS	***
FCR	NS	NS	NS	NS	NS	NS	***
Gizzard[1]	NS	NS	NS	NS	*	NS	NS
Tibia ash (%)	NS	NS	*	NS	*	NS	NS
Feed cost (Rs)	***	***	NS	***	NS	NS	**
Feed cost/ kg live weight gain	***	NS	NS	NS	NS	NS	**

It was concluded that Citric acid (2%) could be included with RB (30%) without causing any adverse effect on growth performance, thereby minimizing the feed cost per kg of live weight gain.

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