



Growth performance and carcass yield of broiler chickens in response to carbohydrases and its association with phytase

[*Desempenho e rendimento de carcaça de frangos de corte em resposta à associação de carboidrase com fitase*]

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ABSTRACT

An experiment was conducted to evaluate broiler chicken performance and carcass yield in response to carbohydrase supplementation (Xylanase and Betaglucanase; XB) and association of carbohydrase and phytase (PHY). A total of 1,920 day-old male broiler chicks (Cobb 500) were distributed in a completely randomized design with 8 treatments and 8 replicates each, allocated in 64 pens with 30 chicks each. The treatments were: Positive Control (PC); Negative Control (NC): reduced ME [-120kcal/kg (Starter) and -170kcal/kg (Finisher)]; NC + XB [50g/ton]; NC + XB [100g/ton]; NC + XB [150g/ton]; NC + XB [50g/ton] + PHY [100g/ton]; NC + XB [100g/ton] + PHY [100g/ton]; NC + XB [150g/ton] + PHY [100g/ton]. The inclusion of XB (150) and XB (50, 100, and 150) associated with phytase increased feed intake than positive control when considering the total rearing phase. The feed conversion ratio of all broilers fed diets with energy reduction were worse than positive control, even with the addition of enzymes, and did not differ between them. There was no significant effect of treatments on carcass parameters.

Keywords: additives, corn, metabolizable energy, enzyme, soybean meal

RESUMO

O experimento foi conduzido para avaliar o desempenho e o rendimento de carcaça de frangos de corte em resposta à suplementação de carboidrase (Xilanase e Betaglucanase; XB) e à associação de carboidrase com fitase (PHY). Um total de 1.920 frangos de corte, machos, com 1 dia de idade (Cobb 500), foram distribuídos em um delineamento experimental inteiramente ao acaso, com oito tratamentos e oito repetições, totalizando 64 unidades experimentais (boxes) com 30 aves cada. Os tratamentos foram: controle positivo (PC); controle negativo (NC): redução de EM [-120kcal/kg (fase inicial) e -170kcal/kg (fase final)]; NC + XB [50g/ton]; NC + XB [100g/ton]; NC + XB [150g/ton]; NC + XB [50g/ton] + PHY [100g/ton]; NC + XB [100g/ton] + PHY [100g/ton]; NC + XB [150g/ton] + PHY [100g/ton]. A inclusão de XB (150) e a associação de fitase com XB (50, 100 e 150) aumentaram o consumo de ração em relação ao tratamento controle positivo, considerando-se a fase total de criação. A conversão alimentar de todos os frangos que receberam dietas com redução de energia foi pior que o controle positivo, mesmo com a adição das enzimas, e não diferiu entre elas. Não houve efeito significativo dos tratamentos sobre os parâmetros de carcaça.

Palavras-chave: aditivos, milho, energia metabolizável, enzima, farelo de soja

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INTRODUCTION

Corn and soybean meal are ingredients that have a good digestibility, however depending on the storage, crop and variety, these ingredients may have a change in their nutritional quality. For broilers, these ingredients constitute around 70 and 30% of the diet, respectively. Thus, any small improvement in their quality significantly influences the performance of the broiler chickens and the meat production cost. The carbohydrate fraction of cereal ingredients has non-starch polysaccharides (NSP), which are considered an anti-nutritional factor for broilers, since they produce no endogenous digestive enzymes to degrade this molecule. Thus, exogenous enzymes are needed to improve the energy utilization of the diets for broilers.

Several studies with carbohydrases, have been conducted with corn and soybean meal diets, even though these ingredients are known to cause low viscosity, aiming to improve the digestibility of the fiber fraction of the diet (Kaczmarek *et al.*, 2014; Ferreira Júnior *et al.*, 2016). In addition, studies with phytase supplementation in poultry diets have showed that this enzyme make minerals, amino acids and energy available (Rutherford *et al.*, 2012; Walk *et al.*, 2012; Khodambashi Emami *et al.*, 2013; Ribeiro Júnior *et al.*, 2016). Other researchers (Karimi *et al.*, 2013; Lu *et al.*, 2013; Roofchaei *et al.*, 2017) studied the interaction of carbohydrases and phytase combination in order to evaluate if there is synergistic effects, but the responses are variable, since this effect depends on several factors, as the type of carbohydrase product, the composition and content of dietary NSPs, the concentration of phytate, P, and Ca, and the endogenous phytase activity (Woyengo and Nyachoti, 2011).

In this sense, this study was conducted to evaluate the effect of xylanase and betaglucanase (XB) enzymes on performance and carcass yield of broiler chickens fed corn-soybean meal based diets, and verify if their associations with phytase (PHY) improve broilers' response.

MATERIAL AND METHODS

All experimental procedures were approved by the Ethics Committee on Animal Use (CEUA - 013021/14).

The trial was conducted at the Laboratory of Poultry Sciences of Department of Animal Science, Faculty of Agrarian and Veterinary Sciences-UNESP, Jaboticabal, Brazil. The birds were housed in an experimental facility, with tunnel type system to control the environment. The control of temperature, humidity and air exchange were performed automatically by exhaust fans and climate control system, according to the age of the birds (Broiler..., 2012). During the starter phase incandescent lamps were used as heating source according to the needs of the chicks. Shaving litter was used in all experimental plots. Infant tubular feeders were used until 14 days of age, then substituted by adult tubular feeders. Nipple type drinkers were used during all rearing period. Water and feed were offered *ad libitum* during all the trial period. The lighting program was set at 24hours of light.

All birds were vaccinated according to the challenge of the region: at the hatchery against Marek and Gumboro diseases, at 12 days of age the birds were vaccinated against New Castle disease and Gumboro (strong strain), both via water.

A total of 1,920 Cobb male day-old broiler chicks were weighed (\pm 47g each) and distributed in each treatment with similar body weight means. The birds were distributed in a completely randomized design with eight treatments and eight replicates allocated to 64 pens (1.0 x 3.0m) of 30 chicks each. The experimental treatments were: Positive Control (PC) - diet meeting the nutritional requirements of the birds; Negative Control (NC) - reduction of 120kcal/kg (starter phase) and reduction of 170kcal/kg ME (finisher phase); NC + XB (50g/ton); NC + XB (100g/ton); NC + XB (150g/ton); NC + XB (50g/ton) + PHY (100g/ton); NC + XB (100g/ton) + PHY (100g/ton); NC + XB (150g/ton) + PHY (100g/ton).

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All diets were formulated to meet the nutritional requirements of birds based on the recommendations of the Brazilian Tables for Poultry and Swine (Rostagno *et al.*, 2011), except for energy, which was reduced in negative control diet. Table 1 describes the control diets and their formulated nutritional compositions for starter (1-21days) and finisher (22-42days)

phases. Inert ingredient of the NC diet was partially replaced by the corresponding enzymes in order to obtain the treatments. The enzymes utilized were: XB - Carbohydrase Enzymes (Xylanase 12200U/g + Beta-Glucanase 1520U/g) and PHY - Phytase Enzyme 5000FTU/g (Danisco Animal Nutrition).

Table 1. Composition of positive control (PC) and negative control (NC) diets

Ingredients	1-21days		21-42days	
	PC (%)	NC (%)	PC (%)	NC (%)
Corn	59.069	59.808	63.089	63.079
Soybean Meal 45%	35.165	35.014	29.767	29.769
Dicalcium Phosphate	1.694	1.693	1.227	1.227
Soy Oil	1.615	0.000	3.786	1.854
Limestone	0.898	0.899	0.773	0.773
Salt	0.489	0.489	0.400	0.400
Vit. Min. Premix ¹	0.400	0.400	0.400	0.400
DL-Methionine	0.312	0.311	0.258	0.258
L-Lysine HCl	0.250	0.253	0.224	0.224
L-Threonine	0.108	0.109	0.077	0.077
Inert ingrediente	0.000	1.023	0.000	1.940
Total	100	100	100	100
	Nutritional levels			
ME (kcal/kg)	3000	2880	3200	3030
Crude protein (%)	22.20	22.20	20.00	20.00
Calcium	0.867	0.867	0.715	0.715
Sodium	0.213	0.213	0.198	0.198
Available Phosphorus (avP)	0.424	0.424	0.334	0.334
Lysine (dig)	1.253	1.253	1.099	1.099
Methionine (dig)	0.600	0.599	0.524	0.524
Met+Cys (dig)	0.902	0.902	0.803	0.803
Threonine (dig)	0.814	0.814	0.714	0.714
Tryptophan (dig)	0.235	0.235	0.200	0.200

¹ Folic Acid 200mg; Pantotenic Acid 3120mg; Biotin 10mg. Cloridrohiquinolona 7500mg. Niacin 8400mg; Choline 78.10g; Nicarbazin 27.50g; Selenium 75mg; Vit A 1680000UI. Vit B1 326.50mg; Vit B12 2400MGC; Vit B2 1200mg; Vit B6 624mg; Vit D3 400000UI; Vit E 3500UI; Vit K3 360mg; Mn 18.74g; Zn 17.5g; Fe 11.25g; Cu 1997mg; I 187.47mg.

At 21 and 42 days old, birds and feed leftovers were weighed. The performance parameters measured were: feed intake (FI), body weight gain (BWG) and feed conversion ratio (FCR). At 42 days, three birds per pen were slaughtered by electronarcosis followed by exsanguination to evaluate carcass, breast with bone and abdominal fat yield.

Data were analyzed by general linear models procedures of SAS (2002). The means were tested by Duncan multiple-range test with 5% of significance.

RESULTS AND DISCUSSION

Table 2 presents data of FI, BWG and FCR of broiler chickens from 1 to 21 and 1 to 42 days.

Table 2. Performance of broiler chicks from 1 to 21 and 1 to 42 days old fed diets with metabolizable energy (ME) reduction and supplemented with XB (Xylanase and Betaglucanase) and XB + PHY (Phytase) (mean \pm standard deviation)

Treatment	1 to 21 days		
	Feed Intake (g)	Body Weight Gain (g)	Feed Conversion Ratio (g/g)
Positive Control	1107.3 \pm 20.5 B	781.4 \pm 20.1 AB	1.42 \pm 0.03 A
NC (120kcal/kg reduced ME)	1150.6 \pm 24.3 A	775.3 \pm 25.3 ABC	1.49 \pm 0.03 B
NC + XB (50g/ton)	1138.1 \pm 29.7 AB	767.9 \pm 17.1 BC	1.48 \pm 0.01 B
NC + XB (100g/ton)	1129.1 \pm 14.0 AB	756.8 \pm 12.8 C	1.49 \pm 0.03 B
NC + XB (150g/ton)	1139.9 \pm 18.6 AB	771.0 \pm 13.4 ABC	1.48 \pm 0.02 B
NC + XB (50g/ton) + PHY	1133.4 \pm 27.7 AB	768.6 \pm 9.70 BC	1.47 \pm 0.03 B
NC + XB (100g/ton) + PHY	1165.4 \pm 23.0 A	790.0 \pm 16.7 A	1.48 \pm 0.02 B
NC + XB (150g/ton) + PHY	1165.1 \pm 22.3 A	790.1 \pm 17.0 A	1.48 \pm 0.04 B
Coefficient of variation (%)	1.98	2.23	1.87
p-value	<0.0001	0.0101	0.0001
	1 to 42 days		
Positive Control	4568.7 \pm 55.5 B	2788.8 \pm 31.2 A	1.64 \pm 0.01 A
NC*	4732.7 \pm 128.2 A	2751.6 \pm 60.6 ABC	1.72 \pm 0.02 B
NC + XB (50g/ton)	4647.4 \pm 103.5 AB	2710.5 \pm 59.4 C	1.71 \pm 0.02 B
NC + XB (100g/ton)	4664.7 \pm 75.9 AB	2724.2 \pm 15.7 BC	1.71 \pm 0.02 B
NC + XB (150g/ton)	4730.6 \pm 87.8 A	2786.5 \pm 55.3 A	1.70 \pm 0.04 B
NC + XB (50g/ton) + PHY	4713.6 \pm 64.5 A	2786.4 \pm 53.2 A	1.69 \pm 0.02 B
NC + XB (100g/ton) + PHY	4747.6 \pm 62.2 A	2780.8 \pm 41.5 AB	1.71 \pm 0.03 B
NC + XB (150g/ton) + PHY	4755.1 \pm 104.4 A	2793.4 \pm 65.3 A	1.70 \pm 0.01 B
Coefficient of variation (%)	1.89	1.84	1.39
p-value	0.0027	0.0161	<0.0001

NC= negative control. *Starter (1-21 days) reduction of 120kcal/kg of metabolizable energy, and Finisher (22-42 days) reduction of 170kcal/kg of metabolizable energy. Means with no common letter in row differ significantly ($P < 0.05$) by Duncan test. PHY = Phytase was included at the 100g/ton level.

The ME reduction used in this study (120kcal/kg ME in the initial phase and 170kcal/kg ME in the finisher phase) was significant to increase feed intake in both phases ($P < 0.05$). Usually the nutritional reduction in a diet implies on increase of FI by the broilers to compensate the nutrient deficiency in the diet. As expected, the ME reduction was significant in increasing FI, since NC diets promoted a higher FI when compared to PC. Several studies (Masey *et al.*, 2012; Gopinger *et al.*, 2017) found that the reduction of ME on broilers' diets increases the FI. However, the enzymes inclusions were not able to significantly reduce the FI. It is important to mention that PHY has the effect of increase FI, because phytate acts as an appetite suppressant which, when broken, increases FI in broilers (Cowieson *et al.*, 2011; 2017). The higher FI of these diets reflected in increased BWG, with responses very close to the PC treatment.

The ME reduction of NC diet was not able to reduce the BWG of the broilers. However, for the starter phase (1-21 d) the combination of PHY (100g/ton) with XB over 100g/ton, showed

the best BWG and were higher than the treatments containing up to 100g/ton of XB and also than the lower combination of PHY (100g/ton) with XB (50g/ton). When evaluating the total (1-42 d) rearing phase, diets supplemented with XB (150g/ton) and with the association PHY(100g/ton) + XB (50g/ton) presented higher BWG in comparison to the diets supplemented only with XB (50 and 100g/ton).

Drawing a parallel between the use of XB and its association with PHY, the combination of PHY (100g/ton) with XB (100g/ton) showed higher BWG compared to the diet containing only XB (100g/ton) in the initial phase, and diets supplemented with PHY (110g/ton) + XB (50g/ton) were also superior to the diets containing only XB (50g/ton) from 1 to 42 days, showing that PHY is able to act releasing energy from the treatments with lower XB levels.

The FCR of the broilers fed PC diets was 6.1 and 6.6% better ($P < 0.05$), in initial (1-21) and total (1-42) rearing phases, respectively, when

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compared to the other diets with ME reduction. In both phases, no enzyme supplementation was able to improve FCR in comparison to NC ($P < 0.05$).

Masey *et al.* (2012) reduced ME (100kcal/kg ME) and included xylanase in the diet and observed significantly improvement of FCR by adding the enzyme, which was not observed in this study. This can be explained because of the activity of carbohydrases on the ingredients, what influenced the lack of response in FCR in poultry. The higher BWG was due to increased FI, caused by the reduced ME. The combination of PHY and XB showed also no effect for FCR, since the PHY activity improves FI and BWG and, consequently, does not affect FCR. According to Woyengo and Nyachoti (2011), xylanase associated with phytase may present synergistic effect only if the ingredients used in the diet have high concentration of NSP (more than 10%), and is widely known that corn and soybean meal have low NSP content.

The lack of response on performance variables to carbohydrase enzymes has also been reported in other studies (Vieira *et al.*, 2015; Pasquali *et al.*, 2017). Woyengo and Nyachoti (2011) reported that this effect is because of the type and concentration of NSP in feedstuffs vary because of the type and concentration of NSPs, (i.e. corn is rich in arabinoxylans, whereas barley is rich in beta-glucans). Thus, the difference in amount and type of NSP represents changes on the substrate for the activity of the enzyme in the gastrointestinal tract of broilers, causing a minor response. According to Cowieson *et al.* (2010), diets with a high nutritional value will show less response to NSP degrading enzymes, since the nutrients available in the diet will meet the requirement of the broiler, which will, then, reach or come close to the maximum genetic potential. This occurs especially with the use of genetically improved cereals, which contains less antinutritional factors, such as glucosinolates, tannins, phytic acid and fiber, which reduce the efficiency of the enzymatic utilization.

It is not possible to say that diets containing enzymes showed or not responses to BWG

because PC and NC diets did not differ significantly. However, it was observed that the higher inclusion of XB and its association with PHY presented the best BWG. The enzyme XB (100g/ton) included in the NC diet in initial phase was not able to achieve the same BWG of the broilers of PC, showing the worst BWG. This fact also occurred to the inclusions of XB (50 and 100g/ton) from 1 to 42 days. Yegani and Korver (2013) observed similar results, a negative impact on performance when added different carbohydrases to the diets of broilers. According Olukosi *et al.* (2007), the negative response does not mean that the enzyme had no activity on their respective substrates, but because of the good quality of the ingredient that does not allow the action of the enzyme. As an example, Rios *et al.* (2017) obtained responses on ileal digestibility energy, crude protein and amino acids digestibility from xylanase associated with glucanase, however the performance was not affected. Thus, what could have occurred in the present study is a small effect of the enzymes on NSPs, due to the nature of the ingredients. This does not mean that in a large-scale, at commercial farms, the use of these enzymes is not feasible.

The results of adding different concentrations of XB and its association with PHY, on carcass, breast and abdominal fat yield of broilers slaughtered at 42 days of age are presented in Table 3. There was no significant difference ($P > 0.05$) for any carcass variables evaluated in this study.

Despite the differences found in this study, the carcass, breast and abdominal fat yield were not influenced neither by XB, nor by its association with PHY, meaning that the use of these enzymes in diets with reduced ME does not affect the relationship between carcass and viscera. Similar results were found by Williams *et al.* (2014) and Nunes *et al.* (2015) who did not find differences in carcass yield with the use of carbohydrases and phytase. However, Santos *et al.* (2017) observed a positive effect on carcass yield and breast weight of broilers fed diets supplemented with xylanase in combination with phytase.

Table 3. Carcass, breast and abdominal fat yield of broilers fed diets with metabolizable energy (ME) reduction and supplemented with XB (Xylanase and Betaglucanase) and XB + PHY (Phytase) (mean \pm standard deviation)

Treatment	Carcass Yield (%)	Breast Yield (%)	Abdominal Fat (%)
Positive Control	75.00 \pm 0.94	40.99 \pm 1.31	1.62 \pm 0.38
NC*	75.28 \pm 1.07	40.33 \pm 1.28	1.28 \pm 0.12
NC + XB (50g/ton)	75.15 \pm 0.90	41.30 \pm 1.44	1.49 \pm 0.38
NC + XB (100g/ton)	75.29 \pm 0.54	40.85 \pm 1.19	1.44 \pm 0.13
NC + XB (150g/ton)	75.39 \pm 0.72	40.72 \pm 0.92	1.31 \pm 0.28
NC + XB (50g/ton) + PHY	75.17 \pm 0.47	39.93 \pm 1.39	1.55 \pm 0.23
NC + XB (100g/ton) + PHY	75.28 \pm 0.87	40.96 \pm 0.58	1.35 \pm 0.24
NC + XB (150g/ton) + PHY	75.49 \pm 0.98	40.83 \pm 1.05	1.30 \pm 0.21
Coefficient of variation (%)	1.12	2.81	17.56
p-value	0.9763	0.4096	0.0925

*Starter (1-21 days) reduction of 120kcal/kg of metabolizable energy, and Finisher (22-42 days) reduction of 170kcal/kg of metabolizable energy. Means with no common letter in row differ significantly ($P < 0.05$) by Duncan test. PHY = Phytase was included at the 100g/ton level.

Thus, it is possible to conclude that the XB included to the level of 100g/ton was not very effective for the release of ME from the diet, whereas its use at the highest level associated with PHY showed the best response to BWG. However, none of the enzymes were able to affect FCR, carcass, breast and abdominal fat yields in diets with low ME.

CONCLUSIONS

The metabolizable energy reduction was not able to affect body weight gain, but negatively affected feed intake and feed conversion ratio. The supplementation of carbohydrase and its association with phytase were not able to improve body weight gain and feed conversion ratio of broilers fed diets with metabolizable energy reduction, but increased feed intake, demonstrating a synergistic effect only to this last parameter. The metabolizable energy reduction and the supplementation of carbohydrase and its association with phytase did not influence carcass parameters of broilers at 42 days of age.

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