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## The Effects of Using the Multi Carbohydrase Preparation in Diets Containing Canola Meal on Performance of Broiler Chickens

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**Abstract:** The aim of this study was to investigate the effects the addition of multi enzyme preparation and the replacement value of canola meal (CM) for soybean meal (SBM) on performance broiler chickens. A total of 384 (Ros-308) day-old broiler chicks were allotted randomly to 8 dietary treatments each in 4 replicates of 12 birds per pen. The 2 enzyme levels (0 and 500 gr/ton) and the 4 CM levels (0, 20, 30 and 40) were used in 2 x 4 factorial arrangements in isoenergetic and isonitrogenous diets. The experiment lasted for 42 d and a single mash diet was used throughout the experiment. The body weight (BW) gain and feed consumption were significantly ( $P < 0.01$ ) reduced when levels of CM from 20 to 40% and feed-gain ratio was depressed ( $P < 0.01$ ). A depression in breast weight was observed in birds that were fed the added from CM. Adding levels of CM from 20 to 30% significantly ( $P < 0.01$ ) decreased leg weight. Gizzard weight as percentage on carcass weight increased linearly ( $P < 0.05$ ) with the addition levels of CM from 30 to 40%. The addition of enzyme numerically improved BW gain at 1-21 and 1-42 days and also improved carcass weight and breast weight, but effects were not significant ( $P > 0.05$ ). The interaction between CM and enzyme for total performance parameter at this experiment were not significant. These results showed that CM can be used in place of 20% of the SBM.

**Key words:** Enzyme supplementation, canola meal, growth, carcass response, broiler chickens

### Introduction

A considerable amount of canola meal (CM, the oil free residue of low glucosinolate, low erucic acid rapeseed) a co-product of the canola oil extraction industry, is available for use in animal feeds. The protein content of CM is about 35% and has an excellent balance of amino acids and particularly rich in sulfur-containing amino acids (Zeb *et al.*, 2002; Kocher *et al.*, 2000; Newkirik and Classen, 2002; Ahmad *et al.*, 2007). The introduction of low glucosinolate rapeseed (canola) has made CM a suitable alternative in place of soybean meal (SBM) as a vegetable protein source in broiler diets (Kocher *et al.*, 2001). CM is a commonly used vegetable protein in poultry diets. Leeson *et al.* (1987) who showed that CM could replace up to 100% of dietary SBM without major negative effects on performance, energy or mineral utilization, provided lysine as the limiting amino acid was added. However, the low level of available energy, the reduced level of crude protein as well as lysine and the increased levels of indigestible carbohydrates of CM compared with SBM (Bell, 1993), make CM a less competitive alternative when used at high levels in broiler diets (Kocher *et al.*, 2001). The relatively low level of available energy in CM is associated with high levels of non-starch polysaccharides (NSPs) (Slominski and Campbell, 1990). CM contains on average 2.5%  $\alpha$ -galacto-OS and 18% NSP of which 1.5% is soluble (Bell,

1993) and approximately SBM contains 18-21% NSP, of which 2.5%-3% are soluble (Bach Knudsen, 2001). Furthermore NSPs adversely effecting digestion of other nutrition by attracting water and causing feed to form a bolus that may reduce exposure to digestive enzymes (Buchanan *et al.*, 2007) also this compounds increase stickiness of excreta and increased litter moisture (Francesch and Brufau, 2004). Desirable results of enzyme using in cereal-based diets (Adeola and Bedford, 2005; Gao *et al.*, 2007), has stimulated interest in the application of enzymes to target the vegetable protein components of poultry diets. Marsman *et al.* (1997) and Kocher *et al.* (2002) showed that use of enzyme increased protein and NSP digestibilities and AME content in birds fed SBM-diets.

Addition of multi carbohydrase supplement containing xylanase, amylase, and protease activities to a corn-SBM broiler diets results in a significant improvement in performance (Zanella *et al.*, 1999). A recent study by Vahjen *et al.* (2005) showed that the most effective enzymes for SBM were 1, 4-b-arabinogalactanases, but reported that there were some synergies on the release of sugars when this enzyme was combined with a galactomannanase. There is only limited information available on the anti-nutritive effects of CM NSPs and ability of commercial enzyme products to degrade these components. Meng *et al.*, 2005, who showed that multi

carbohydrase supplement was effective in depolymerizing cell wall polysaccharides of SBM, CM, and peas *in vitro*. The high prices of SBM in Iran limited its consumption in poultry diets. On the other hand, CM is available at a much cheaper price. The current study was designed to determine the effect of multi enzyme (Rovabio Excel) on the different levels CM to evaluate the response of broiler.

### Materials and Methods

**Bird husbandry:** A total of 384, one-day-old broiler chickens (Ras-308) were randomly allotted to floor pen (12 birds in each replicate) with new wood shaving as litter and a floor space was 0.053 m<sup>2</sup> for one bird. Each pen was equipped with one hanging feeder one automatic trough waterer. The house temperature was maintained at 32°C during the first week of age, and then gradually reduced by 2-3°C per week and practiced until a temperature of 22°C was attained. The 24 h of light was provided throughout the experiment. Birds were vaccinated against infectious Influenza and Newcastle disease at 8 old, for infectious bursal disease at 13 old of age and the experiment concluded for 42 day.

**Experimental diets and bird performance:** The analyzed values for CM used in the present study were 88/64, 37/63, 4/47 and 16/48% for DM, CP, EE and CF, respectively (AOAC, 2000). Four levels of CM (0, 20, 30 and 40%) were used with two levels enzyme (0 and 500 gr/ton) in factorial design 4 x 2 in 8 dietary combinations in Isoenergetic and Isonitrogenous diets, following the two stage feeding system, starter (1-21 days) and grower (22-42 days). All diets (Table 1) met National Research Council (NRC, 1994) recommendation. The supplemental enzyme activities reported by the supplier were 2,200 visco Units/g of xylanase and 200 AGL units of  $\beta$ -glucanase and other enzyme such as cellulase, pectinase, protease and mannosidase. Feed and water were provided *ad libitum*. Feed consumption and average bodyweight were monitored weekly of age. At the end of 42 days of age, two birds (male and female) from each replicate were randomly selected for eviscerated to determine the body parameters.

**Statistical analysis:** Data were analyzed according to the GLM procedure for ANOVA using SAS (SAS, 2005). Duncan's multiple-range test was used to separate means when significant effects ( $P < 0.05$ ) were detected by analysis of variance.

### Results

The effects of exogenous enzyme addition to rations and of using CM in place of SBM on broiler performance for 21 and 42 days are shown in Table 2. From 1-21 days, and from 1-42 days, body weight (BW) gain in of broilers fed the diet with enzyme supplementation was

numerically improved over the unsupplemented control, however had no significant in the present study ( $P > 0.05$ ). Feed consumption and feed gain ratio were not significantly affected by enzyme supplementation ( $P > 0.05$ ). Exogenous enzyme didn't significant affect on the carcass weight, breast weight and leg weight ( $P > 0.05$ ). Increasing the level of CM from 1-21 and 1-42 days in diet, resulted in significant reduction body weight gain ( $P < 0.05$ ). feed:gain ratio and feed consumption were depressed in birds fed with increasing CM during all the periods of experiment ( $P < 0.05$ ). At the end of experiment observed that increasing the level of dietary CM in replacement of SBM, the carcass weight, breast weight and leg weight had decreased linearly and the highest decrease in recent organs relevant to 30 and 40% CM levels in replacement SBM. In addition, increasing the levels of CM replacement by in SBM from 20-40% resulted in an increase of relative gizzard weight and relative heart weight (Table 3). The interaction of enzyme and CM was not significant for at over the period of all experiment.

### Discussion

It seems that CM may be viewed as an economically, suitable alternative to SBM as a vegetable protein source in broiler diets. In the present study, when dietary CM was increased from 20-40% in replacing with SBM, it resulted in reduced performance during 1-21 days and 1-42 day. This study indicated that increasing level of CM in diet has adverse effect on the performance of broiler. The results of our study confirmed the result by Newkirk and Classen (2002), Kermanshahi and Abbasi pour (2006) and Saricicek and Serdar (2006).

A significant reduction in performance parameters of broiler chickens may be caused by of high glucosinolate content of the experimental diets. The CM contains substantial concentrations of phenolic compound that cause a bitter taste and decrease its palatability (Shahidi and Naczki, 1992). Therefore, contribution of Glucosinolates (GLs) themselves are biologically inactive molecules, but glucosinolates degradation products are biologically active and known for their diversified biological effects, Thus increasing level CM in diet lead to a reduced in feed intake at 1-21 days and 1-42 days of age. Reduced intake often causes growth depression on diets containing high glucosinolate CM (Hill, 1991; Tripathi *et al.*, 2001; McNeill *et al.*, 2004). This study showed that the above mentioned compounds present in CM are partly responsible for this effect. The reduced intake of GLs containing diets is due to the presence of sinigrin and progoitrin, these both glucosinolates are associated with bitter taste (Fenwick *et al.*, 1982). Progoitrin produces more profound bitter taste compared to sinigrin (Doorn *et al.*, 1998). Although, progoitrin is a non-bitter glucosinolates, but is degraded

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Table 1: Ingredient composition of experimental diets<sup>†</sup>

	Starter diet				Grower diet			
	0 CM%	20 CM%	30 CM%	40 CM%	0 CM%	20 CM%	30 CM%	40 CM%
	----- Ingredients -----							
Corn	57.7	56.55	55.85	55.4	60.43	59.8	58.5	58.3
SBM	32.5	26	22.75	19.5	29.5	23.6	20.65	17.7
CM	-	6.5	9.75	13	-	5.9	8.85	11.8
Fish meal	4.6	5.5	5.9	6.3	4.2	5	5.7	6.1
DCP	0.9	0.6	0.5	0.43	0.8	0.5	0.5	0.4
Grind shale	1.1	1.1	1.1	1.1	1	0.9	0.9	0.7
Salt	0.16	0.16	0.16	0.16	0.17	0.17	0.17	0.17
NaHCO <sub>3</sub>	0.14	0.14	0.14	0.14	0.14	0.14	0.14	0.14
Vegetable oil	1.5	2	2.3	2.5	2.5	2.73	3.3	3.4
Methionine	0.3	0.3	0.4	0.32	0.21	0.21	0.24	0.24
Lysine	0.05	0.1	0.1	0.1	-	-	-	-
Premix †	1	1	1	1	1	1	1	1
Coccidiostats	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
	----- Calculated nutrient composition -----							
CP (%)	22	22	22	22	20.6	20.6	20.6	20.6
ME (kcal/kg)	2990	2990	2990	2990	3020	3020	3020	3020
Av. P (%)	0.52	0.52	0.52	0.52	0.45	0.45	0.45	0.45
Ca (%)	1.01	1.01	1.01	1.01	0.9	0.9	0.9	0.9
Lysine (%)	1.27	1.27	1.27	1.27	1.11	1.11	1.11	1.11
Met+Cys (%)	0.95	0.95	0.95	0.95	0.81	0.81	0.81	0.81

<sup>†</sup>Each diet was divided into 2 parts, with enzyme and without enzyme.

<sup>†</sup>Supplied per kilogram of diet: Vitamin A, 10000 IU; Vitamin D, 9790 IU; Vitamin E, 121 IU; B<sub>12</sub>, 20 µg; Riboflavin, 4.4 mg; Calcium Pantothenate, 40 mg; Niacin, 22 mg; Choline, 840 mg; Biotin, 30 µg; Thiamin, 4 mg; Zinc Sulfate, 60 mg; Manganese Oxide, 60 mg.

Table 2: Effect of supplementation of exogenous multi enzymes preparation on the BW gain, Feed intake and Feed:gain of broiler during 1 to 21 days and 1 to 42 days of age fed different levels CM

Item		BW gain	Feed intake	Feed:gain	BW gain	Feed intake	Feed:gain
		(gr)	(gr)	(gr:gr)	(gr)	(gr)	(gr:gr)
		----- 1 to 21 d -----			----- 1 to 21 d -----		
Enzyme	CM	512.58 <sup>a</sup>	931.11 <sup>a</sup>	1.83 <sup>a</sup>	1877.65 <sup>a</sup>	3720.16 <sup>a</sup>	1.98 <sup>a</sup>
		521.59 <sup>a</sup>	930.36 <sup>a</sup>	1.79 <sup>a</sup>	1904.51 <sup>a</sup>	3797.61 <sup>a</sup>	1.99 <sup>a</sup>
No		8.38	10.27	0.02	28.42	31.85	0.02
Yes	0	548.87 <sup>a</sup>	989.39 <sup>a</sup>	1.8 <sup>ab</sup>	2038.38 <sup>a</sup>	3902.14 <sup>a</sup>	1.91 <sup>b</sup>
<sup>†</sup> SEM	20	525.70 <sup>ab</sup>	905.68 <sup>b</sup>	1.73 <sup>b</sup>	1954.80 <sup>ab</sup>	3748.56 <sup>ab</sup>	1.92 <sup>b</sup>
	30	510.90 <sup>ab</sup>	904.77 <sup>b</sup>	1.78 <sup>ab</sup>	1881.69 <sup>b</sup>	3674.45 <sup>b</sup>	1.95 <sup>b</sup>
	40	482.84 <sup>b</sup>	923.09 <sup>b</sup>	1.92 <sup>a</sup>	1689.38 <sup>c</sup>	3710.62 <sup>b</sup>	2.19 <sup>a</sup>
	SEM	8.38	10.27	0.02	28.42	31.85	0.02
		----- Significance -----					
<sup>†</sup> S.V							
Enzyme		<sup>†</sup> NS	NS	NS	NS	NS	NS
CM		*	**	NS	**	NS	**
Enzyme x CM		NS	NS	NS	NS	NS	NS

<sup>a-c</sup> Values within a column with unlike superscripts differ significantly (P< 0.05).

\*P< 0.05.

\*\*P< 0.01.

<sup>†</sup>Standard Error Means.

<sup>†</sup>Not Significant

<sup>†</sup>Source of variations

by myrosinase or by heat treatment to the extremely bitter compound goitrin (Tripathi and Mishra, 2007). These compounds cause to over work of thyroid gland and enlarged size. Therefore, the phosphorylation coefficient decrease and energy loss in the form of heat from the body. It is, in general, believed that GLs in poultry diets must be less than 2.5 µmol.g<sup>-1</sup> (Mushtaq *et al.*, 2007). Reduced performances were reported by Cowan *et al.* (1999), Szczurek *et al.* (2000) and Ahmad *et al.* (2007) with high levels CM in broilers. Previous studies (Kocher *et al.*, 2000) have also shown similar reductions in body weight of broiler chickens with high levels of CM.

Previous studies (Kocher *et al.*, 2001) demonstrated no adverse effect of low GLs in CM when it was added at 35% of broilers diets. The reduction of body weight gain may also be related to the lysine-arginine imbalance in higher levels of CM in diets (Summers and Leeson, 1978). Also high level of the phytic acid content of CM could partly lead to decrease in growth, comprised of 2.9-3.2% phytic acid (Zhou *et al.*, 1990), whereas SBM contains only 1.4% (De Boland *et al.*, 1975). In addition, non-ruminant animals utilize the protein in CM protein less effectively than that of SBM (Yin *et al.*, 1994). And this may be related to the elevated levels of phytate. The

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Table 3: Effect of supplementation of exogenous multi enzymes preparation and various levels of CM on the Carcass weight, Breast weight, Leg weight, Heart weight, Gizzard weight, Liver weight.

Item		Carcass <sup>1</sup> weight	Breast <sup>1</sup> weight	Leg <sup>1</sup> weight	Heart <sup>1</sup> weight	Gizzard <sup>1</sup> weight	Liver weight
Enzyme	CM						
No		1527.34 <sup>a</sup>	452.34 <sup>a</sup>	400.93 <sup>a</sup>	0.88 <sup>a</sup>	3.03 <sup>a</sup>	2.57 <sup>a</sup>
Yes		1545.94 <sup>a</sup>	460.93 <sup>a</sup>	398.28 <sup>a</sup>	0.91 <sup>a</sup>	3.00 <sup>a</sup>	2.92 <sup>a</sup>
SEM		32.85	13.10	9.01	0.02	0.08	0.06
	0	1724.38 <sup>a</sup>	538.75 <sup>a</sup>	444.68 <sup>a</sup>	0.84 <sup>a</sup>	2.84 <sup>b</sup>	2.90 <sup>ab</sup>
	20	1642.81 <sup>a</sup>	489.06 <sup>b</sup>	428.12 <sup>a</sup>	0.89 <sup>a</sup>	2.74 <sup>b</sup>	2.58 <sup>b</sup>
	30	1485.63 <sup>b</sup>	433.12 <sup>c</sup>	389.06 <sup>b</sup>	0.90 <sup>a</sup>	3.13 <sup>ab</sup>	2.94 <sup>a</sup>
	40	1293.75 <sup>c</sup>	365.62 <sup>d</sup>	336.56 <sup>c</sup>	0.97 <sup>a</sup>	3.36 <sup>a</sup>	2.91 <sup>ab</sup>
	<sup>†</sup> SEM	32.85	13.10	9.01	0.02	0.08	0.06
----- Significance -----							
<sup>‡</sup> S.V							
Enzyme		<sup>†</sup> NS	NS	NS	NS	NS	NS
CM		**	***	**	NS	*	NS
Enzyme x CM		NS	NS	NS	Ns	NS	NS

<sup>a-c</sup> Values within a column with unlike superscripts differ significantly (P< 0.05).

\*P< 0.05.

\*\*P< 0.01.

\*\*\* p < 0.001

<sup>1</sup>Standard Error of Means.

<sup>†</sup>Not Significant

<sup>1</sup>Percentage of carcass weight

<sup>‡</sup>Source of variations

studies showed that the addition of phytase, improve performance broilers in CM-based diets (Newkirk and Classen, 2001; Saricicek and Serdar, 2006).

The effect of enzyme supplementation on growth performance was not significant in the present study. However, many reports have indicated significant improvement of nutritive value of cereal grains such as wheat, barley, ray and triticale by enzyme addition (Cowieson *et al.*, 2006). Mandal *et al.* (2005) reported supplementation of enzyme did not improve energy bio-availability of rapeseed meal. Similarly, carbohydrase did not influence the growth performance, starch digestibility, or content of sunflower meal or CM-based diets (Kocher *et al.*, 2000; Mushtaq *et al.*, 2006). These findings disagree with reports from Rassmussen and Petterson (1997) and Guenter *et al.* (1998) which both showed that the addition of carbohydrase preparations with pectinase activities to CM and canola-soybean meal based diets significantly, improved BW gain. In contrast to the current study, both of those studies used a control diet deficient in energy and protein. This would indicate that the addition of enzymes improved nutrient digestibility and improved the availability of nutrients to the bird in order for them to grow to their full potential. However, all diets in the current experiment were formulated according to the birds' nutrient requirements. In this study, by increasing the CM levels the gizzard weight increased linearly with the CM of the diet (Table 3). The possible reason may be that the CM contains indigestible high fiber and NSPs and the increased bulkiness (physical form) increased the gizzard weight. The CM contains on average 4.5% soluble NSP (Bell, 1993), whereas SBM only contains 2.7% soluble NSP (Irish and Balnave, 1993). Meng and Slominski (2005) indicated that the enzyme effects were not always beneficial in CM-based diet. On the other hand, the heart

weight increased linearly with the addition of levels CM in the diet, but had no significant in the present study. This it seems that the GLs content of the CM causes alters ratio between thyroid hormones concentration (Triiodothyronine and Thyroxinin) in blood and ascend body metabolism. Therefore, increased rate of heart beat and thus heart volume (Olkowski and Classen, 1998). The earliest varieties of rapeseed were associated with heart disease, which was attributed to the high level of erucic acid in the oil (Abdellatif, 1979). In conclusion, the results of this study clearly indicated that 20% CM can be included in broiler diets as the main dietary protein source in place of SBM without loss in bird performance.

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