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Effect of Dietary Phytase and NSP-degrading Enzymes in Diets Containing Rape Seed Meal on Broiler Performance and Carcass Characteristic

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Abstract: This experiment was conducted to investigate the effect of replacement of Soybean Meal (SBM) with Rape seed Meal (RSM) using two type enzymes on broiler performance. Three hundred sixty Ross strain chickens were used in a 2×2×3 factorial arrangement with two levels of Phyzyme (0 and 500 FTU/kg diet), two levels of Grindazyme (0 and 0.17%) and three levels of RSM (0, 25 and 50% replaced to SBM protein) in a completely randomized design three replicates and 10 birds per replicate. Body Weight Gain (BWG), Feed Intake (FI) and Feed Conversion Ratio (FCR) were measured weekly. Carcass weight and carcass components weight recorded at the end of trial (day 42). The results of this experiment indicated that FI, BWG and feed efficiency of broiler were significantly ($p<0.05$) decreased by increasing RSM in all period of experiment. Use of RSM significantly decrease breast and abdominal fat weight of broiler ($p<0.05$). In grower and whole period of experiment the BWG (1353.10 vs 1285.36 g in grower and 1861.30 vs 1798.92 g in whole period) and FI (2638.28 vs 2589.13 g in grower and 4328.44 vs 3368.93 g in whole period) of broiler were significantly ($p<0.05$) increased by addition of Grindazyme, but were not affected by supplementation of Phytase in the diet. Exception abdominal fat weight, there was not significant effect for phytase and Grindazyme on carcass and carcass component weight. Therefore, it was concluded that NSP-degrading enzymes and Pytase may be incorporated in RSM based broiler diet for profitable production.

Key words: Broiler, enzyme, rape seed meal, phytase, carcass

INTRODUCTION

In comparison to about 44% crude protein in Soybean Meal (SBM), the protein content of Rape seed Meal (RSM) is about 35- 40% and has a physiologically suitable amino acid combination in animal nutrition (Hickling, 2001; Kocher *et al*., 2000), but the digestibility of some amino acids is less than that of SBM (Liang, 2002). However, RSM contains nutritionally unfavorable substances such as glucosinolates, sinapin, tannin, phytate (Ciska and Kozlowska, 1998) and non starch polysaccharides (Kocher *et al*., 2000). Among the antinutritional factors, the high fiber level is one of the greatest restrictions to RSM use in poultry diets (Liang, 2002). Dietary fiber accounts for approximately 1/3rd of the RSM and it consists of cellulose (4-6%), non-cellulosic polysaccharides (13- 16%), lignin, polyphenols (5-8%), protein and minerals associated with the fiber fraction (Slominski and Campbell, 1990). Thus, the relatively low AMEn content and less protein and amino acid digestibility that caused by the high level of dietary fiber, are considered as main factors that limited use of RSM in poultry diets. The use of feed enzymes in poultry diets is now commonplace in barley, wheat and oat based diets in many countries (Bedford, 2000; Bedford and Schulze, 1998). Enzymes have the greatest potential use in diets that contain antinutritional factors that hinder nutrient availability. The Non-Starch Polysaccharides (NSP) in feedstuffs have been the main target of commercial feed enzymes. These NSPs which include cellulose, B-glucans, arabinoxylans and pectins may increase viscosity of digesta and resulting in decreased of nutrient digestibility (Liang, 2002). The presence of NSPs may adversely affect the performance of broiler chickens fed high levels of RSM (Bedford, 2000; Annison and Choct, 1991).

Phytate (myoinositol 1-6-hexakis, dihydrogen phosphate) another anti-nutrient factor of RSM. The major portion of Phosphorus (P) in plant feed ingredient is present in form of phytate, whch is largely

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unavailable in monogastric animals (Bozkurt *et al*., 2006). It contains, on average, 70% of the Total P (TP) in the feed ingredients commonly used in poultry diets (Maenz, 2001; Kornegay, 2001a). Phytic acid is present as a mixed salt, phytate, which refers to the phytic acid molecule chelated to mineral cations (such as Ca, Mg, Zn, Fe, Mn and Cu), starch, lipids and also reduce protein availability (Ravindran *et al*., 1999; Bedford and Schulz, 1998; Selle *et al*., 2000; Kornegay, 2001a; Catalá-Gregori *et al*., 2006). The low availability of phosphorus in plant ingredient makes economically and environmentally problems. Phytase activity in the digestive tract of broiler chickens is very low. Thus, broilers haven't adequate levels of phytase activity to effectively hydrolyse the phytate molecule. So inorganic P should be added to broiler diets to meet nutritional requirements and increasing feed costs. Therefore, phytate may be considered an antinutritional factor because it reduces the digestibility of phytate-chelated nutrients. Furthermore, phytate-bound P passed in animal excreta is a source of environmental pollution, contributing to surface water eutrophication (Catalá-Gregori *et al*., 2006).

There are, however, few reports on the addition of enzyme to improve RSM quality. Therefore, the purpose of this study was to investigate the replacement value of SBM with locally grown RSM associated with 2 type enzymes on performance and some organ weights of broiler chickens.

MATERIALS AND METHODS

Birds and diets: The experiment was conducted at the poultry station of Ramin Agricultural and Natural Resources University in Iran. The locally grown Rape Seed Meal (RSM) was purchased from an oil extraction Co. in Neishabour, Iran. For preparing RSM, the oil of the RSM was extracted by hexan. Three levels of (0.0, 25.0 and 50%) RSM protein were replaced with SBM protein and 2 levels of a Phytase enzyme (0 and 500 FTU Phyzyme kg^{-1}, Phyzyme XP is a bacterial phytase from Schizosacchromyces, produced in Danisco Animal nutrition), 2 levels of a dietary NSP degrading enzyme (0, 0.17% Grindazyme produced in Danisco Animal nutrition, with minimum activity of 36000 U g^{-1} xylanase and 15000 U g^{-1} β-glucanase) were added to the diets during starter (7-21 days of age) and grower (21-42 days of age) periods of broiler chickens. All diets were isocaloric and isonitrogenous (2969 kcal kg^{-1} ME, 21.70% crude protein in starter period and 3118.43 kcal kg^{-1} ME, 19.65% crude protein in grower period). The ingredients percentage and chemical composition of diet are shown in Table 1.

Feed and water provided ad libitum. Chickens were used in a 2×2×3 factorial arrangement in a completely randomized design with 360 Ross strain chickens in 12 treatments and 3 replicates and ten birds per replicate.

Sample collection: Feed consumption and body weight gain of chicks were recorded 4 h after the removal of feed and Feed Conversion Ratio (FCR) calculated as the unit weight of feed per unit of body weight gain at end of every week. When the chicks were 42 day of age, 2 chicks (male and female) were selected randomly from each replication (cages) and slaughtered. The liver, heart, gizzard, abdominal fat pad, breast and the femur were immediately weighed. The carcasses without feather, head, feet and internal organs were weighed.

Statistical analysis: All data were analyzed using the GLM procedure of SAS software for analysis of variance. Treatment means when significant ($p<0.05$), were compared using Duncan's multiple range test.

RESULTS AND DISCUSSION

The results of the performance and carcass traits of broiler chickens fed rape seed meal and 2 type enzymes are given in Table 2 and 3. Feed intake, body weight gain and FCR were not significantly affected by dietary phytase in 7-21, 22-42 and 7-42 days of experiment ($p>0.05$). However Broilers fed diets contain phytase have numerically more FI and BWG and had better FCR than broilers fed diet without enzyme in grower and whole period of experiment. Broilers fed diet with phytase weighted 1844.76 g compared with 1815.45g for broilers receiving diet with no phytase in day 42. These results suggested that phytase moderately increased the availability of nutrients and improved feed intake in this period. These results were in agreement with those of Rezaei *et al*. (2007). These scientists reported that broilers receiving 500 FTU kg^{-1} Natuphos phytase have more FI than control treatment. Bozkurt *et al*. (2006), Qian *et al*. (1997), Huff *et al*. (1998), Namkung and Leeson (1999) and Zyla *et al*. (2000) reported that the growth rate and feed conversion ratio of broilers fed the low phosphorus diets containing microbial phytase are comparable with or even better than those obtained for broilers fed the standard phosphorus diets. This matter showed that phytase could compensate the possibility of reduction of the level of Ca and P without any adverse effect. Viverous *et al*. (2002) reported that due to increasing feed intake simultaneously with body weight, effect of phytase supplementation on FCR of broiler chicks was not significant. *In vitro* studies have shown that phytate-protein complexes are insoluble

Table 1: Composition of experimental diets

	Treatments											
Ingredients (%)	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	T_9	T_{10}	T_{11}	T_{12}
Age: 7-21 days												
RSM (%)	0.00	25.00	50.00	0.00	25.00	50.00	0.00	25.00	50.00	0.00	25.00	50.00
Phytase (FTU)	0.00	0.00	0.00	500.00	500.00	500.00	0.00	0.00	0.00	500.00	500.00	500.00
Grindazyme (%)	0.00	0.00	0.00	0.00	0.00	0.00	0.17	0.17	0.17	0.17	0.17	0.17
Corn	58.40	55.82	53.50	58.40	55.82	53.50	58.40	55.82	53.50	58.40	55.82	53.50
SBM	31.20	23.93	16.57	31.20	23.93	16.57	31.20	23.93	16.57	31.20	23.93	16.57
RSM	0.00	9.27	18.55	0.00	9.27	18.55	0.00	9.27	18.55	0.00	9.27	18.55
Fish meal	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Veg. oil	2.00	2.72	3.35	2.00	2.72	3.35	2.00	2.72	3.35	2.00	2.72	3.35
DCP[1]	1.20	1.10	0.98	1.20	1.10	0.98	1.20	1.10	0.98	1.20	1.10	0.98
Dl-met	0.28	0.26	0.24	0.28	0.26	0.24	0.28	0.26	0.24	0.28	0.26	0.24
L-lysine	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18
Salt	0.13	0.13	0.13	0.13	0.13	0.13	0.13	0.13	0.13	0.13	0.13	0.13
Oyster	0.87	0.85	0.81	0.87	0.85	0.81	0.87	0.85	0.81	0.87	0.85	0.81
$NaHCO_3$	0.21	0.21	0.17	0.21	0.21	0.17	0.21	0.21	0.17	0.21	0.21	0.17
Min.Vit. premix[2]	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Calculated analysis												
ME (kcal kg^{-1})	2969.00	2969.00	2969.00	2969.00	2969.00	2969.00	2969.00	2969.00	2969.00	2969.00	2969.00	2969.00
CP (%)	21.70	21.70	21.70	21.70	21.70	21.70	21.70	21.70	21.70	21.70	21.70	21.70
Ca (%)	0.95	0.95	0.95	0.95	0.95	0.95	0.95	0.95	0.95	0.95	0.95	0.95
AP (%)	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Age: 22-42 days												
Corn	62.30	60.30	58.28	62.30	60.30	58.28	62.30	60.30	58.28	62.30	60.30	58.28
SBM	25.80	19.85	13.85	25.80	19.85	13.85	25.80	19.85	13.85	25.80	19.85	13.85
RSM	0.00	7.57	15.14	0.00	7.57	15.14	0.00	7.57	15.14	0.00	7.57	15.14
Fish meal	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Veg. oil	3.58	4.13	4.70	3.58	4.13	4.70	3.58	4.13	4.70	3.58	4.13	4.70
DCP[2]	1.00	0.90	0.80	1.00	0.90	0.80	1.00	0.90	0.80	1.00	0.90	0.80
Dl-met	0.23	0.22	0.20	0.23	0.22	0.20	0.23	0.22	0.20	0.23	0.22	0.20
L-lysine	0.21	0.21	0.21	0.21	0.21	0.21	0.21	0.21	0.21	0.21	0.21	0.21
Salt	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Oyster	0.90	0.88	0.85	0.90	0.88	0.85	0.90	0.88	0.85	0.90	0.88	0.85
$NaHCO_3$	0.20	0.18	0.16	0.20	0.18	0.16	0.20	0.18	0.16	0.20	0.18	0.16
Vit.E	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Min.Vit. premix[1]	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Calculated analysis												
ME (kcal kg^{-1})	3118	3118	3118	3118	3118	3118	3118	3118	3118	3118	3118	3118
CP (%)	19.65	19.65	19.65	19.65	19.65	19.65	19.65	19.65	19.65	19.65	19.65	19.65
Ca (%)	0.90	0.90	0.90	0.90	0.90	0.90	0.90	0.90	0.90	0.90	0.90	0.90
AP (%)	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.451

1: Supplied kg^{-1} of diet: Vitamin A, 10000 IU; Vitamin D_3, 9790 IU; Vitamin E, 121 IU; B_{12}, 20 µg; riboflavin, 4.4 mg; calcium pantothenate, 40 mg; niacin, 22 mg; choline, 840 mg; biotin, 30 ig; thiamin, 4 mg; zinc sulfate, 60 mg; manganese oxide, 60 mg.2: Di calcium phosphate

and less subject to attack by proteolytic enzymes than the same protein alone and this interaction may influence the digestibility of protein (Ravindran *et al*., 1995). Furthermore, phytate is also able to bind endogenous proteins such as trypsin and chymotrypsin in the gastrointestinal tract (Singh and Krikorian, 1982) and these enzymes are released into the gut from the pancreas and if they become bound to the phytate molecule, protein and amino acid digestibility could be reduced. Ravindran *et al*. (1999) and Yi *et al*. (1996) reported that the addition of phytase to corn soybean meal diet released more phytate. Effect of different treatments on carcass characteristics is presented in Table 3. Exception abdominal fat pad, the additions of phytase had no significant effect on carcass weight and carcass components weight of broilers ($p>0.05$). The highest value for carcass weight and carcass components weight was belonged to broilers received phytase. These results were in agreement with those of Rezaei *et al*. (2007). They shown no significant difference for carcass percentage, breast meat, tights and abdominal fat. Ahmad *et al*. (2004) reported that carcass, breast meat, tights and liver weight of chicks were increased in chicks fed with diets supplemented with phytase (1.5 g kg^{-1}). Ahmed *et al*. (2004) reported that carcass weight and carcass component weights (such as breast, thigh and heart weights) were increased by addition of different levels of phytase in broiler diets which are in contrast with the results of the present study. It seems, in this study, amount of Phosphorous and other nutrient is adequate with broiler requirement (NRC, 1994) and the Phytase addition had not more effect.

Table 2: Effect of levels of Phytase enzyme, NSP-degrading enzyme and rape seed meal on broiler performance

	Weigh gain (g)			Feed intake (g)			FCR (Feed: weight gain)		
Variable	7-21	22-42	7-42	7-21	22-42	7-42	7-21	22-42	7-42
No Phytase	505.39	1310.06	1815.45	786.44	2599.58	3386.84	1.56	1.99	1.87
Phytase	516.34	1328.42	1844.76	779.69	2627.84	3410.54	1.51	1.99	1.85
SME	7.03	20.66	21.98	5.04	22.49	23.49	0.02	0.03	0.02
No Grindazyme	513.54	1285.36^{b}	1798.92^{b}	779.80	2589.13^{b}	3368.93^{b}	1.52	2.02	1.87
Grindazyme	508.19	1353.10^{a}	1861.30^{a}	786.33	2638.28^{a}	3428.44^{a}	1.55	1.95	1.85
SME	7.13	19.40	20.58	5.12	21.88	22.50	0.02	0.03	0.02
0 RSM	537.98^{a}	1365.31^{a}	1903.29^{a}	792.13^{a}	2647.78^{a}	3444.42^{a}	1.47^{b}	1.95	1.81^{b}
25% RSM	486.81^{b}	1314.26^{ab}	1801.06^{b}	769.92^{b}	2621.71^{ab}	3391.63^{ab}	1.58^{a}	2.00	1.89^{a}
50% RSM	507.82^{b}	1278.16^{b}	1785.97^{b}	787.17^{ab}	2571.63^{b}	3360.02^{b}	1.55^{a}	2.02	1.89^{a}
SME	6.09	24.80	22.80	5.43	24.80	25.77	0.02	0.03	0.02
T_1	533.33	1366.51	1908.10	791.83	2710.60	3500.60	1.49	1.99	1.83
T_2	495.92	1313.08	1792.24	786.17	2578.33	3349.83	1.59	1.96	1.87
T_3	474.83	1216.67	1724.17	790.50	2528.67	3316.83	1.66	2.08	1.92
T_4	541.83	1289.17	1824.33	799.67	2562.83	3349.33	1.48	1.99	1.83
T_5	497.17	1249.67	1736.83	782.67	2566.17	3316.67	1.58	2.05	1.91
T_6	506.06	1277.17	1807.83	767.17	2588.67	3380.33	1.51	2.05	1.87
T_7	541.95	1393.65	1926.98	790.50	2675.26	3467.09	1.46	1.92	1.88
T_8	507.50	1284.50	1759.33	788.17	2549.09	3339.59	1.55	1.99	1.90
T_9	479.17	1285.96	1781.89	771.50	2556.00	3347.08	1.61	1.99	1.88
T_{10}	535.17	1411.91	1953.74	786.50	2642.93	3460.63	1.47	1.89	1.77
T_{11}	530.67	1409.78	1915.84	791.67	2793.26	3560.43	1.50	1.98	1.86
T_{12}	487.17	1332.83	1830.00	750.50	2613.17	3395.83	1.54	1.96	1.86
SME	12.51	43.53	37.72	12.01	39.82	42.99	0.04	0.07	0.04

^{a,b}Means within a column with no common superscripts differ significantly ($p<0.05$)

The effects of adding NSP-degrading enzyme are given in Table 2. In starter period of experiment, feed intake and body weight gain were not significantly affected by dietary Grindazyme ($p>0.05$). However, in grower and whole period, feed intake and body weight gain were higher in diet contain Grindazyme ($p<0.05$). Enzyme treatment had no effect on FCR in all period. Viverous *et al.* (2002) reported that due to simultaneously increasing of feed intake and body weight, effect of enzyme supplementation on FCR of broiler chicks was not significant that agree with result of current experiment. It seemed supplementation of NSP-degrading enzyme can improve the NSP availability and reduce the negative impact of indigestible component (Annison and Choct, 1991). Studies have indicated that carbohydrase or protease supplementation improve feed conversion ratio (Simbaya *et al.*, 1996), but Alloui *et al.* (1994) and Kermanshahi and Abbasi (2006) reported that the addition of protease and carbohydrase enzyme did not significantly improve bird performance. The results of this experiment indicated that with exception breast weight and abdominal fat pad of broilers, other carcass components were not significantly affected by enzyme treated diet. This result is consistent with Kermanshahi and Abbasi (2006), who found that carcass yield had no significant difference, among enzyme treated and non-treated diet.

The effect of RSM on feed intake, body weight gain and FCR of broiler chickens are shown in Table 2. Feed intake, body weight gain and feed to gain ratio were significantly affected by treatments ($p<0.05$). Replacement of RSM with SBM in 25 and 50% levels had no significant effect on body weight gain, feed intake and feed to gain ratio of broiler chickens. Feed intake of broiler chickens at 7-21, 21-42 and total period was significantly decreased by RSM as its inclusion into the diets increased. In the grower period and in whole period of experiment, lowest feed intake was for 50% replacement. The RSM contains substantial concentrations of phenolic compound that cause a bitter taste and decrease its palatability (Shahidi and Naczk, 1992; Kermanshahi and Abbasi, 2006). It was indicated that the phenolic compounds may contribute to the dark color, bitter taste and astringency of canola meal and may affect feed intake (Campbell and Van der Poel, 1998). The evidence indicates that diet palatability can be adversely affected by the glucosinolates of the RSM (Mawson *et al.*, 1993). But Leeson *et al.* (1987) found that even complete replacement of SBM (100%) with canola meal (<30 μmoles g^{-1} glucosinolate) did not affect the feed intake in broilers and laying hens. The influence of flavor on feed intake is less important for poultry than other livestock animal because the senses of taste and smell among the birds are not developed as well as the other species (Zeb, 1998). However, RSM contains nutritionally unfavorable substances such as glucosinolates, sinapin, tannin, phytate (Ciska and Kozlowska, 1998) and non starch polysaccharides (Kocher *et al.*, 2000) that adversely effected broilers feed intake. Slominski and Campbell (1990) demonstrated that the digestibility of NSP fraction

Table 3: Carcass weight and carcass components weight (g) of broilers on different levels of enzyme and RSM

Variable	Carcass	Breast	Femur	Gizzard	Abdominal fat	Liver	Heart
No Phytase	1411.33	442.33	444.16	47.67	32.37^{b}	43.81	8.74
Phytase	1434.41	455.29	442.05	50.09	37.12^{a}	46.78	8.56
SME	30.90	9.19	12.13	1.23	1.56	1.32	0.28
No Grindazyme	1406.83	443.33	439.83	48.59	32.39^{b}	44.40	8.48
Grindazyme	1438.38	454.41	445.88	49.28	37.24^{a}	46.26	8.79
SME	30.84	9.22	12.08	1.26	1.57	1.26	0.27
0 RSM	1472.50	469.50^{a}	458.25	50.31	37.46^{a}	46.95	8.66
25% RSM	1425.91	449.77^{ab}	449.09	48.50	32.35^{b}	46.70	8.85
50% RSM	1376.82	430.22^{b}	423.18	48.18	35.55^{b}	42.65	8.44
SME	37.54	10.97	14.61	1.47	1.92	1.52	1.34

^{a,b}: Means within a column with no common superscripts differ significantly ($p<0.05$)

of canola meal in laying hens was below 3% and this low digestibility of fiber components was the main factor responsible for depressing protein digestibility, amino acids availability and the ME value in the meal.

The effect of RSM on carcass characteristics is shown in Table 3. The breast weight and abdominal fat pad of broilers were significantly ($p<0.05$) decreased by inclusion of RSM. The more abdominal fat pad of the birds receiving more SBM may be related to higher AME content and availability of nutrient of the SBM. In addition to this possibility, the phytate, tannin and other anti-nutritional factors present in RSM has less digestibility and adversely effect carcass component weight. The reduction in abdominal fat pad content has been observed previously (Janjecic *et al*., 2002) and may be elated to reduction of T_3 hormone in the blood serum.

CONCLUSION

Under the condition of this study, inclusion of RSM had adverse effect on performance and carcass traits. But replacement in 25 and 50% levels had no more effected on performance and carcass traits of broiler chickens. Therefore, it was concluded that NSP-degrading and Pytase enzymes may be incorporated in rape seed meal based broiler diet for profitable production. Further investigations are required to clarify these ideas.

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