



Effects of Enzyme Addition to Broiler Diets Containing Varying Levels of Double Zero Rapeseed Meal

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ABSTRACT : Maize-soybean meal diets with 0, 100, 200 and 300 g/kg double zero rapeseed meal ('00' RSM) with and without an enzyme mixture (xylanase, pectinase, cellulase) at a level of 1.6 g/kg were evaluated with 624 day-old broiler chicks for 5 weeks. The birds were randomly allocated to eight dietary treatments with three replicates of 26 birds each. Average daily gain (ADG) and feed intake (FI) were recorded weekly and ileal viscosity, organ weights, serum enzyme activity, hormonal profile and hematological parameters were measured at the end of week 5. Average daily gain during the weekly periods was significantly influenced by the dietary level of '00' RSM ($p < 0.01$). Inclusion of '00' RSM improved the ADG up to day 28 with the increased level; beyond that time no improvement was recorded when compared to control groups. However, ADG from 1-35 days was significantly different between 300 g/kg inclusion level of '00' RSM and the control diet. Inconsistent decline in feed intake and feed conversion ratio was observed up to day 21 and the trend was reversed thereafter. The proportion of '00' RSM in the diet had a significant ($p \leq 0.05$) influence on thyroid weight but had no effect on the relative weights of liver and heart, serum enzyme activities (γ -glutamyl transferase, alanine amino transferase and aspartate amino transferase), thyroid hormones (T_3 and T_4), hemoglobin level and hematocrit. Significant improvement in ADG was recorded during the 2nd week of age with the addition of enzyme, whereas for all other periods, including the whole period of the trial, higher but non-significant ADG was observed. FI and FCR were not affected by the addition of enzyme but there was a numerical reduction in FCR during the whole period. The addition of enzyme reduced the ileal viscosity at all levels of '00' RSM inclusion. The results suggest that '00' RSM can be included up to 300 g/kg in broiler diets without any adverse effects on health and performance. The addition of commercial enzyme mixture containing xylanase, pectinase, cellulase to broiler diets containing '00' RSM has some effect on growth rate and feed conversion efficiency. (**Key Words :** NSP (Non-starch Polysaccharide), '00' RSM (Double Zero Rapeseed Meal), AST (Aspartate Amino Transferase), ALT (Alanine Amino Transferase), GGT (γ -Glutamyl Transferase), T_3 (Triiodothyronine), T_4 (Thyroxine))

INTRODUCTION

The use of rapeseed meal (RSM) in poultry diets has been highly limited by the presence of glucosinolates and other minor anti-nutritional factors like sinapine and tannin (Fenwick and Curtis, 1980; Leeson and Summers, 2001; Pengbin et al., 2002). With the advent of rapeseed cultivars low in glucosinolates and erucic acid content, the inclusion of double zero rapeseed meal ('00' RSM) in poultry diets is on the rise.

The high levels of non-starch polysaccharides (NSP) in '00' RSM is limiting its unrestricted use in poultry feeding (Slominski and Campbell, 1990; Kocher et al., 2000;

Malathi and Devegowda, 2001; Shim et al., 2003). These NSPs are known to increase the gut viscosity, reduce nutrient absorption in the intestine and affect indirectly the growth and performance of birds (Annison and Choct, 1991; Annison, 1991; Choct and Annison, 1992). Many studies have clearly demonstrated that, the addition of enzymes to diets rich in NSP results in a significant reduction in the intestinal viscosity, enhances energy and protein utilization and in turn improves the performance (Kocher et al., 2000). There is only limited information available on the anti-nutritive effects of '00' RSM NSP and the ability of commercial enzyme products to degrade these compounds. Thus, the NSP content of '00' RSM used in the present trial was quantified and the appropriate enzymes to degrade those NSP were added to different treatment diets.

Despite the availability of '00' RSM, there are still a number of problems, such as the adverse effects of low levels of glucosinolates on the thyroid and the consequent

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Table 1. Composition of the basal diets

Ingredients (%)	Starter diets				Finisher diets			
	Diet I	Diet II	Diet III	Diet IV	Diet I	Diet II	Diet III	Diet IV
Yellow maize	66.1	58.3	56.0	53.0	68.6	66.0	63.3	61.0
Soybean meal	26.0	20.8	15.0	10.0	20.4	14.2	8.5	3.0
Groundnut cake	10.0	7.7	6.0	4.0	8.0	7.0	5.5	3.5
Double zero rapeseed meal	0.0	10.0	20.0	30.0	0.0	10.0	20.0	30.0
Di calcium phosphate ¹	1.6	1.6	1.6	1.6	1.5	1.3	1.2	1.4
Shell grit	1.1	1.0	0.8	0.65	1.5	1.5	1.2	1.1
Salt (g/kg)	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
A B ₂ D ₃ K ² (g/kg)	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Ventrimix-BE ³ (g/kg)	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Monocox ⁴ (g/kg)	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
DL-methionine (g/kg)	2.0	2.0	2.1	2.1	1.6	1.6	1.6	1.7
Lysine HCl ⁵ (g/kg)	0.65	0.77	0.90	0.84	0.51	0.64	0.64	0.77
Choline chloride ⁶ (g/kg)	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Trace mineral ⁷ (g/kg)	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Calculated composition								
Protein (g/kg)	215.7	215.1	214.7	215.9	187.8	190.1	190.3	189.6
ME (MJ/kg)	12.16	12.14	12.18	12.16	12.42	12.46	12.47	12.21
Crude fiber (g/kg)	41.9	48.1	54.6	61.1	39.2	45.9	52.5	58.9
Calcium (g/kg)	10.1	10.3	10.1	10.1	9.7	9.5	9.4	9.5
Available P (g/kg)	4.6	4.6	4.6	4.7	4.3	4.3	4.2	4.2
Lysine (g/kg)	12.0	12.0	12.0	12.0	10.0	10.0	10.0	10.0
Methionine (g/kg)	5.3	5.3	5.3	5.3	4.5	4.5	4.5	4.5

¹ Provides per kg: Calcium, 230 g; phosphorus, 180 g.

² Provides per kg diet: A, 12,375 IU; D₃, 1,800 IU; B₂, 7.5 mg; K, 1.5 mg.

³ Provides per kg diet: B₁, 4 mg; B₆, 8 mg; B₁₂, 40 µg; E, 20 mg; niacin, 60 mg; calcium pantothenate, 12.5 mg.

⁴ Provides per g: Maduramycin, 10 mg.

⁵ Provides per g: Lysine HCl, 780 mg.

⁶ Provides per g: Choline chloride, 600 mg.

⁷ Provides per kg: Mn, 80 mg; Zn, 80 mg; I, 3.29 mg; Fe, 32 mg; Cu, 3.9 mg; Se, 0.1 mg.

interference with metabolism and growth, liver hemorrhages, increased leg problems and appetite depression (Ibrahim et al., 1980; Martland and Butler, 1984; Pusztai, 1989). For these reasons, further investigation is essential to bear out the profitability of this oilseed meal in broiler nutrition. In this perspective, '00' RSM used in this trial was analyzed for the glucosinolate levels, and its effect on the thyroid and liver functioning were tested. The twin objectives of the current study were to evaluate the effects of '00' RSM feeding on the health of broilers and to investigate the effect of enzyme addition to '00' RSM supplemented diets on the growth and performance of broilers.

MATERIALS AND METHODS

Glucosinolate and nonstarch polysaccharide composition of double zero rapeseed meal

Glucosinolate content in double zero rapeseed meal (*Brassica napus*) used in this trial was quantified by HPLC (Agilent Technologies; 1100 series, Singapore) method (ISO, 1992). The total pentosans, pectin, cellulose and total NSP content of '00' RSM were estimated by the procedures of Frazer et al. (1956), Sadasivam and Manickam (1996),

Updegraff (1969) and Englyst and Cummings (1988), respectively.

Experimental design

In a deep litter trial, 624 d-old Cobb broiler chicks were randomly allocated to eight experimental groups, each comprising three replicates of 13 male and 13 female chicks. Four diets containing 0, 100, 200 and 300 g/kg '00' RSM were prepared with (1.6 g/kg) and without a combination of enzyme mixture (xylanase, pectinase and cellulase) to produce eight dietary treatments in a factorial arrangement. Broiler chicks were fed on starter diets from 0 to 3 wk and finisher diets from 4 to 5 wk. The composition of starter and finisher basal diets is shown in Table 1.

The addition of the enzyme mixture at a level of 1.6 g/kg provided following levels of individual enzyme activity; xylanase - 298 U/g, pectinase - 5,000 Pectinase Units/g and cellulase - 788 Carboxy Methyl Cellulose Units/g. The xylanase was derived from *Trichoderma viridae* and *Aspergillus aculeatus*, whereas pectinase and cellulase were derived from *Aspergillus aculeatus*.

Data collection

The weight gain and feed intake were measured once a

Table 2. Chemical, nonstarch polysaccharides and glucosinolate composition of double zero rapeseed meal

Composition	Percent level
Moisture	6.08
CP	37.01
Fat	8.80
Crude fibre	8.63
Acid insoluble ash	0.87
Total pentoson	3.05
Pectin	9.41
Cellulose	9.20
Total NSP	23.50
Glucosinolate content	32.1 µmol/g

wk and summarized after five wks. Two (one male and female) birds from each replicate were sacrificed by cervical dislocation and ileal contents were collected from Meckel's diverticulum to one cm above the ileocecal junction and kept on ice prior to centrifugation (10,000×g, 15 m at 4°C). The supernatant was used for viscosity measurement with a Brookfield Viscometer-LVTD (Brookfield Engineering Laboratories, MA-1031, USA) at ambient temperature with a spindle and chamber (SC14-18). The relative weights of thyroid and liver were expressed as mg/100 g and g/100 g body weight, respectively.

At the end of trial, blood was collected in non-heparinized tubes from two birds (one male and female)

from each replicate by brachial vein puncture. Serum was collected by the procedure of Calne et al. (1992) and stored at -20°C for future analysis. The serum samples were analyzed for triiodothyronine (T₃) and thyroxine (T₄) by radioimmunoassay kits (Solidphase Inc., Portland, Maine 04103 USA) and for activity of γ-glutamyl transferase (GGT), alanine amino transferase (ALT) and aspartate amino transferase (AST) using an automatic analyzer (Boehringer Mannheim, Hitachi, Japan). The hemoglobin and hematocrit were measured using the same auto analyzer.

Statistical analysis

The results were subjected to analysis of variance by the General Linear Model procedure of SAS[®] (SAS Institute, 1996). For all the parameters each replicate was considered as an experimental unit with an exception for average daily gain where records on individual birds were available. The differences between means were compared by Duncan's multiple range test. The level of significance was tested at $p \leq 0.05$.

RESULTS

Chemical, glucosinolate and nonstarch polysaccharides composition in double zero rapeseed meal

The chemical, non-starch polysaccharides and

Table 3. Average daily gain of broiler chickens fed diets containing varying levels double zero rape seed meal ('00'RSM) with and with out enzyme supplementation (Values are means±SEM)

Main effect	Levels	Average daily gain (day)					
		0-7	8-14	15-21	22-28	29-35	1-35
----- g -----							
‘00’ RSM (%)	0	11.5±0.14 ^d	27.1±0.32 ^b	49.3±0.53 ^c	55.5±0.85 ^b	55.5±1.08 ^a	39.3±0.42 ^b
	10	11.9±0.13 ^c	27.5±0.28 ^{ab}	51.1±0.52 ^b	51.3±0.70 ^c	50.1±1.03 ^b	38.4±0.30 ^c
	20	12.9±0.13 ^a	28.0±0.30 ^a	52.8±0.51 ^a	54.9±0.73 ^b	50.9±0.86 ^b	39.9±0.40 ^b
	30	12.5±0.14 ^b	28.1±0.28 ^a	52.9±0.46 ^a	58.9±0.42 ^a	54.6±0.97 ^a	41.4±0.30 ^a
Enz. (g/kg)	0	12.3±0.10 ^a	26.6±0.20 ^b	51.9±0.35 ^a	54.8±0.55 ^a	53.2±0.71 ^a	39.8±0.30 ^a
	1.6	12.2±0.10 ^a	28.9±0.20 ^a	51.2±0.38 ^a	55.6±0.56 ^a	52.5±0.71 ^a	40.3±0.31 ^a
Sex	Male	12.1±0.11 ^a	27.6±0.21 ^a	53.1±0.40 ^a	57.3±0.59 ^a	54.9±0.71 ^a	41.0±0.31 ^a
	Female	12.3±0.09 ^a	27.8±0.19 ^a	50.0±0.31 ^b	53.1±0.49 ^b	50.8±0.69 ^b	38.8±0.29 ^b
Pooled SE		0.071	0.148	0.259	0.390	0.504	0.198
----- Probability -----							
RSM level		0.0001	0.1999	0.0001	0.0001	0.0001	0.0001
Enzyme		0.2911	0.0001	0.0093	0.3416	0.8281	0.5645
Sex		0.0789	0.4965	0.0001	0.0001	0.0001	0.0001
Block		0.0067	0.1719	0.4184	0.1858	0.5108	0.6573
Replication		0.3235	0.3478	0.4629	0.0956	0.1036	0.5792
b (C.G) ¹		0.1745	0.0001	0.0001	0.3820	0.6098	0.0001
RSM×enzyme		0.0001	0.8678	0.0022	0.8804	0.0001	0.0095
RSM×sex		0.1925	0.3170	0.0001	0.0018	0.0009	0.4677
Sex×enzyme		0.8566	0.3698	0.7207	0.1396	0.5632	0.0915
RSM×enzyme×sex		0.8979	0.7470	0.5924	0.5556	0.1911	0.2165

^{a-c} Values within a column with unlike superscripts differ significantly ($p < 0.05$).

¹ Cumulative gain at the commencement of each period has taken as covariate in the model used.

Table 4. Weekly feed consumption and feed conversion ratio of broiler chickens fed diets containing varying levels double zero rape seed meal ('00' RSM) with and with out enzyme supplementation (Values are means±SEM)

'00' RSM (%)	Enzyme (g/kg)	Feed consumption (day)						Feed conversion ratio (day)					
		1-7	8-14	15-21	21-28	29-35	1-35	1-7	8-14	15-21	21-28	29-35	1-35
		g						g/g					
0	0	124 ^a	290 ^a	535 ^a	736 ^c	863 ^b	2,549 ^{cd}	1.60 ^a	1.62 ^a	1.53 ^a	1.90 ^a	2.10 ^c	1.81 ^b
0	1.6	116 ^b	278 ^a	493 ^b	767 ^{bc}	934 ^{ab}	2,589 ^{bcd}	1.38 ^b	1.38 ^b	1.45 ^a	1.96 ^a	2.59 ^b	1.90 ^a
10	0	116 ^b	285 ^a	472 ^{bc}	742 ^c	919 ^{ab}	2,534 ^d	1.36 ^{bc}	1.53 ^{ab}	1.35 ^b	2.08 ^a	2.71 ^{ab}	1.92 ^a
10	1.6	115 ^b	284 ^a	447 ^c	766 ^{bc}	935 ^{ab}	2,546 ^{cd}	1.43 ^b	1.42 ^b	1.22 ^c	2.10 ^a	2.84 ^{ab}	1.90 ^a
20	0	115 ^b	283 ^a	495 ^b	757 ^c	979 ^a	2,629 ^{bc}	1.26 ^c	1.50 ^{ab}	1.31 ^{bc}	1.97 ^a	2.84 ^a	1.91 ^a
20	1.6	116 ^b	282 ^a	484 ^b	775 ^{bc}	968 ^a	2,625 ^{bc}	1.32 ^{bc}	1.39 ^b	1.34 ^{bc}	2.08 ^a	2.53 ^b	1.86 ^{ab}
30	0	117 ^b	280 ^a	491 ^b	806 ^{ab}	952 ^a	2,647 ^{ab}	1.32 ^{bc}	1.47 ^{ab}	1.32 ^{bc}	1.99 ^a	2.57 ^b	1.86 ^{ab}
30	1.6	118 ^b	289 ^a	493 ^b	822 ^a	992 ^a	2,714 ^a	1.37 ^{bc}	1.41 ^b	1.34 ^b	1.96 ^a	2.58 ^b	1.85 ^{ab}
Pooled SE		0.645	1.38	5.73	7.07	10.41	14.16	0.023	0.021	0.020	0.024	0.057	0.010
Probability													
Treatment		0.001	0.350	0.002	0.006	0.029	0.001	0.001	0.033	0.000	0.442	0.001	0.116
b ₁ *		-	-	-	-	-	-	0.232	0.995	0.928	0.518	0.429	0.583

^{a-d} values within a column with unlike superscripts differ significantly (p<0.05).

* Live body weight at the commencement of each period has taken as covariate in the model.

Table 5. Relative weights of thyroid, liver and heart (% of live body weight) of broiler chickens fed diets containing varying levels double zero rape seed meal ('00' RSM) with and with out enzyme supplementation at 28- and 35-days of age

Main effects	Levels	Proportional weight					
		Thyroid (days)		Liver (days)		Heart (days)	
		28	35	28	35	28	35
		----- % -----					
'00'RSM (%)	0	0.0050 ^c	0.0040 ^c	3.38 ^b	3.50 ^{ab}	0.70 ^{ab}	0.56 ^a
	10	0.0080 ^{bc}	0.0070 ^b	3.50 ^b	3.25 ^b	0.63 ^b	0.58 ^a
	20	0.0090 ^{ab}	0.0110 ^a	3.68 ^{ab}	3.59 ^b	0.67 ^{ab}	0.59 ^a
	30	0.0150 ^a	0.0080 ^b	3.60 ^b	3.42 ^{ab}	0.73 ^a	0.60 ^a
Enzyme (g/kg)	0	0.0081 ^a	0.0069 ^b	3.65 ^a	3.34 ^b	0.67 ^{ab}	0.57 ^a
	1.6	0.0084 ^a	0.0094 ^a	3.56 ^b	3.66 ^a	0.69 ^a	0.59 ^a
Sex	Male	0.0086 ^a	0.0082 ^a	3.65 ^a	3.64 ^a	0.68 ^a	0.61 ^a
	Female	0.0079 ^a	0.0082 ^a	3.59 ^a	3.39 ^b	0.69 ^a	0.56 ^a
Pooled SE		0.0006	0.001	0.0882	0.087	0.018	0.015
		----- Probability -----					
RSM level		0.0022	0.0001	0.0924	0.0742	0.1145	0.3432
Enzyme		0.5849	0.0008	0.7321	0.2083	0.8167	0.6229
Sex		0.9205	0.5548	0.5282	0.2351	0.4067	0.8042
Block		0.4419	0.0001	0.2645	0.9147	0.4750	0.5194
$b(x_i - \bar{x})^1$		0.2056	-	0.0601	-	0.6859	-
$b(y_i - \bar{y})^2$		-	0.4677	-	0.0544	-	0.4473
RSM×enzyme		0.5428	0.2027	0.9625	0.9830	0.5644	0.4516
RSM×sex		0.4341	0.2386	0.4865	0.1963	0.6684	0.2832
Sex×enzyme		0.1188	0.2377	0.3566	0.2014	0.3903	0.2717
RSM×enzyme×sex		0.7304	0.998	0.0288	0.0759	0.1611	0.5276

^{a-c} values within a column with unlike superscripts differ significantly (p<0.01).

¹⁻² b₁: Regression co-efficient for the effect of x_i, x_i = live body weight at 28 days of age with an average of X bar b₂: regression co-efficient for the effect of y_i, y_i = live body weight at 35 days of age with an average of Y bar.

glucosinolate composition of double zero rapeseed meal ('00' RSM) is shown in Table 2.

Growth performance

Average daily gain (ADG) during weekly periods significantly influenced by the dietary levels of '00' RSM (Table 3). Inclusion of '00' RSM improved the ADG upto day 28 with the increased levels and thereafter no

improvement was recorded as compared to control groups. However, ADG in 1-35 days was significantly different between 300 g/kg inclusion level and control. Inconsistent decline in feed intake and feed conversion ratio was observed upto day 21 and the trend was reversed after that (Table 4). The general trend showed that inclusion of '00' RSM upto 300 g/kg did not affect the growth performance of the broilers. Significant improvement in ADG was

noticed during the 2nd week of age with the addition of enzyme, whereas for all other periods including whole period of the trial higher but non-significant ADG was observed. Significant interaction between the effects '00' RSM level and enzyme addition on ADG was recorded through out the trial except at 2nd and 4th wk of age. However, FI and FCR were not affected with the addition of enzyme but there was a numerical reduction in FCR for the whole period of trial. ADG significantly differed between sexes in all the periods irrespective of '00' RSM level and enzyme inclusion. ADG of male chick was significantly higher than female chicks by 0.056% during whole period. Incorporation of '00' RSM and enzyme addition had no effect on mortality.

Organ weights

The relative weights of liver and heart did not differ between the treatments, but the relative thyroid weights were higher in the birds fed on diets containing '00' RSM (Table 5). However, triiodothyronine (T_3) and thyroxine (T_4) levels were not influenced by the inclusion of '00' RSM (Data not shown). Addition of enzymes mixture increased the relative weights of liver at 35 days of age.

Intestinal viscosity

The inclusion of varying levels of '00' RSM had no effect on digesta viscosity either at 28 or 35 days of age (Table 6). The addition of enzyme reduced the ileal viscosity at all levels of '00' RSM inclusion and compared to 28 days of age, reduction was significant at 35 days of age.

Haematology and serum enzyme profile

The varying levels of '00' RSM and the addition of enzyme had no effects on serum enzyme activities (AST,

ALT and GGT), hemoglobin and hematocrit (Table 6).

DISCUSSION

The total nonstarch polysaccharides (NSP) content of double zero rape seed meal ('00' RSM) used in this trial was 2.35 g/kg and is in concurrence with the NSP values of canola meal reported by Slominski et al. (1994a) and Slominski et al. (1994b). The glucosinolate content also found to be 32.1 $\mu\text{mol/g}$.

The results of performance parameters indicate that broilers can tolerate high levels of dietary '00' RSM without any detrimental effects on their performance. The results are in agreement with the findings of Leeson et al. (1987) and Cowan (1993) who showed that '00' RSM could be used at high levels (>300 g/kg) in place of soybean meal despite of a considerable increase in indigestible NSP. An inconsistent decline in FI as well as FCR up to 21 days observed in the birds fed on diets without enzyme in the current study were concord with the findings of Leslie and Summers, (1972) and Karunajewa et al. (1990). The contradiction in the performance of broilers fed on '00' RSM may be due to variation in glucosinolate as well as fat content. Higher growth rate in the enzyme-supplemented groups, either significant or not, are on par with the observation of Cowan (1993) but are in contrast to the result of Kocher et al. (2000).

The results for FCR and digesta viscosity suggest that efficiency of energy utilization was better in birds fed on enzyme-supplemented diets compared to those fed on unsupplemented diets. Kocher et al. (2000), Cowan (1993) and Selle et al. (2003) have reported similar results. Comparatively higher viscosity at the fourth week observed in this study may be due to age factor as observed by Smits and Annison (1996). As the birds lack NSP-hydrolyzing

Table 6. Intestinal viscosity, plasma enzymes activity and hematology of broiler chickens fed diets containing double zero rapeseed meal ('00'RSM) with and with out enzyme supplementation (Values are means \pm SEM)

'00 RSM (%)	Enz. (g/kg)	Intestinal viscosity		Serum enzymes			Haematology ⁴	
		28 d	35 d	AST ¹	ALT ²	GGT ³	Hb	PCV
		----- Centipoises -----		----- Unit/liter -----			g/100 ml	%
0	0	1.53±0.14 ^a	1.05±0.02 ^a	139±7.5 ^a	11.3±1.9 ^a	11.7±0.3 ^a	8.60±3.0 ^a	31.3±1.0 ^a
0	1.6	1.37±0.10 ^a	0.63±0.03 ^b	137±9.7 ^a	11.0±0.6 ^a	10±0.01 ^a	8.63±3.5 ^a	32.6±1.4 ^a
10	0	1.47±0.08 ^a	1.13±0.14 ^a	137±10 ^a	13.7±0.9 ^a	11.3±0.9 ^a	8.86±1.8 ^a	33.1± 1.3 ^a
10	1.6	1.40±0.06 ^a	0.80±0.02 ^b	137±10 ^a	10.3±2.2 ^a	13±0.58 ^a	8.40±3.2 ^a	31.7±1.1 ^a
20	0	1.44±0.18 ^a	1.13±0.17 ^a	136±7.2 ^a	9.3±1.4 ^a	11±0.01 ^a	8.87±6.2 ^a	32.5±1.6 ^a
20	1.6	1.40±0.06 ^a	1.13±0.07 ^a	137±11 ^a	10.3±0.9 ^a	11.6±1.8 ^a	8.76±2.3 ^a	31.6±1.0 ^a
30	0	1.50±0.14 ^a	1.13±0.04 ^a	140±11 ^a	10.0±1.0 ^a	11.3±0.9 ^a	8.50±2.0 ^a	32.1±0.9 ^a
30	1.6	1.45±0.20 ^a	1.07±0.02 ^a	133±9.2 ^a	10.3±0.9 ^a	10.7±0.9 ^a	8.57±3.4 ^a	32.5±2.8 ^a
Pooled SE		0.039	0.045	2.83	0.47	0.42	1.15	0.46
		----- Probability -----						
Treatment		0.9854	0.0031	0.9998	0.4795	0.0691	0.9794	0.9887

^{a-b} Values within a column with unlike superscripts differ significantly ($p < 0.05$).

^{1, 2, 3} Aspartate amino transferase, alanine amino transferase and γ -glutamyl transferase respectively.

⁴ Hemoglobin (Hb) and pack cell volume or hematocrit (PCV).

enzymes, the water soluble fractions of NSP increases the digesta viscosity and affect the digestibility and absorption of nutrients (Annison, 1991). The soluble NSP level of '00' RSM varies from 0.15 to 0.32 g/kg (Slominski and Campbell, 1990; Bell, 1993). Although the soluble NSP are degraded by microflora of ileum and ceca (Carre et al., 1990), the end products of microflora degradation like volatile fatty acids are not efficiently utilized by chicken due to inefficient absorption of volatile fatty acids in the intestine (Carre et al., 1995).

The inclusion of high and low glucosinolate RSM in poultry diet causes rapeseed meal hepatosis (Martland and Butler, 1984) which is characterized by increased hemorrhage, reticulolysis and lympho proliferation, and decreased hematocrit (Leeson and Summers, 1976; Smith and Campbell, 1976; Pearson and Butler, 1979; Martland and Butler, 1984). This condition is also characterized by elevated serum levels of aspartate amino transferase (AST), alanine amino transferase (ALT), lactate dehydrogenase, and alkaline phosphatase, suggesting hepatic damage (Pearson and Butler, 1979; Pearson et al., 1983; Martland and Butler, 1984), and γ -glutamyl transferase (GGT) indicating damage to the hepatic biliary system or pancreas (Pearson et al., 1983).

The results of the present trial suggest that the inclusion of '00' RSM having total glucosinolate level of 32.1 μ mol/g has no effect on liver weight and morphology, AST, ALT and GGT levels, hemoglobin concentration and hematocrit. Reports of Szymkiewicz et al. (1988), Wight et al. (1987) and Mandal et al. (1981) support these observations. Relative thyroid weights were higher in the birds fed diets containing '00'RSM (Table 6). However, triiodothyronine (T_3) and thyroxine (T_4) levels were not influenced by the inclusion of '00'RSM. A group of glucosinolates present in the '00' RSM produces nitriles or oxazolidinethiones upon hydrolysis, which are known to block the uptake of iodine by the thyroid gland (Karlson, 1969) and in turn result in altered T_3 to T_4 ratio, hyperplasia and hypertrophy of thyroid gland and reduced growth rate (Goh et al., 1985). The glucosinolate level (32.1 μ mol/g) of the '00' RSM used in the present trial has enough goitrogenic activity to increase thyroid weight, although this does not seem to affect the broiler performance. The data on growth rate, T_3 and T_4 levels corroborate this fact.

It was observed that all the birds fed on diets containing '00' RSM at all the levels showed wet droppings which was limited to only the first week of the trial and subsided subsequently. Reported observations to this effect are lacking. The dark color of the feces in the birds fed on diets containing '00' RSM accompanying with wet dropping in the first week, could be misleading factors in farm level usage of '00'RSM.

The difference between males and females with respect to a particular trait or sexual dimorphism can not be attributed to single clear cut reasoning. However, facts such as greater competition between males, social dominance, different nutritional requirements, impact of hormones for growth and fatness could be involved (Le Bihan-Duval., 1998; Zerehdaran et al., 2004).

It is concluded from this study that i) '00'RSM having total glucosinolate content of 32.1 μ mol/g can be included upto 300 g/kg in the broiler finisher diet without any detrimental effect on growth rate, feed efficiency and health status, ii) The addition of enzyme mixture containing xylanase, pectinase and cellulase to the broiler diet containing '00'RSM (23.5% total NSP) has some effect on growth rate and feed conversion efficiency.

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