



## Growth Performance, Blood Parameters, Immune response and Carcass Traits of Broiler Chicks Fed on Graded Levels of Wheat Instead of Corn Without or With Enzyme Supplementation

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### ABSTRACT:

Four hundred and fifty one-day Cobb broiler chicken were used to investigate the effect of partial replacement of corn with wheat grain without or with different commercial enzyme products supplementation on growth performance, immune response, blood serum parameters and some carcass characteristics of broiler chicken. Chicks were allotted into 9 equal groups (50 chicks of mixed sex per group). Three experimental diet were formulated, basal diet containing 100% corn and 25% or 50% corn of the basal diet was replaced by wheat grain (w/w) and the diet adjust to be iso-nitrogenous and iso-caloric diets, each diet supplemented by Kemzyme plus dry or by Combozyme (enzyme blend commercial products) to be 9 experimental groups. The experimental period continued for six continuous weeks. It was observed that wheat inclusion at 25% instead of corn non significantly reduced body weight and gain of broiler chicken, while 50% replacement improved both parameters when compared with chick group fed on corn-soybean based diet. However, it was observed that wheat inclusion at 25 or 50% instead of corn grain in broiler chicken ration increased feed intake by about 1.3% and 9.8% respectively and consequently deteriorate FCR, PER, EEU and PI by about (4.6% and 4.6%), (8.2% and 6.5%), (4.9% and 4.0%) and (13.7% and (4.2%) respectively when compared with broiler chicken group fed on corn – soybean based diet. Moreover, enzyme supplementation in 25% wheat containing diet increased feed intake but, with 50% wheat inclusion decreased feed intake also, enzyme supplementation in wheat included ration improved FCR, PER, EEU and PI when compared with broiler chick group fed on the same diet without enzyme supplementation. On the other hand, wheat inclusion had no significant effect on blood serum total protein, albumin, globulin and glucose concentrations, while reduced blood serum triglycerides and cholesterol concentrations, also wheat inclusion in the broiler chicken diet had determinately effect on liver function through elevation of some blood serum enzymes when compared with chick group fed on corn-soybean based diet. Regarding immune response, it was observed that wheat inclusion instead of corn reduced phagocytic activity and index, and reduce antibody production against NDV while enzyme supplementation improve the health status of broiler chicken when compared with broiler chicken group fed on the same diet without enzyme supplementation. It was observed that both wheat grain inclusion without or with enzyme supplementation had no significant effect on carcass traits of broiler traits and the most prominent effect was leading to reduction of abdominal fat weight and relative weight and improve thymus gland weight of broiler chicks.

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## 1. INTRODUCTION

Corn is a major ingredient (above 50%) in broiler diets, contributing about 65% of broiler ME requirements. However, increasing corn prices by reason of its preferential diversion toward human consumption and corn ethanol industry have compelled the poultry industry to reduce reliance on corn. Despite numerous scientific investigations no alternative grain to corn has yet been identified. Although yellow corn and wheat continue to be the two dominating cereal grains used worldwide in the broiler industry. Corn is the leading grain in North and South America, the Mideast, Africa and Asia, while wheat prevails in Western Europe, Australia, New Zealand and certain provinces in Canada. Moreover, these two cereals are different, each with unique characteristics and challenges, our knowledge has grown concerning how to formulate, and at times, interchange these grains to achieve essentially equal performance in broilers.

On the other hand, certain markets, such as Egyptian market, prefer white carcasses rather than yellow ones, so the main cereal used for broilers is wheat instead of corn. The protein content of wheat is higher than that of corn (14.1 and 8.5% respectively; National Research Council, 1994), as is NSP content (11.4 and 9.0%, respectively; Choct, 1997). In addition, despite the low protein content in cereals, the inclusion of a high percentage of cereal in diets provides the major part of the protein in the diet (Odetallah *et al.*, 2005). Therefore, the effectiveness of proteases and carbohydrases on broilers fed with wheat-soybean meal based diets could be higher than that with corn-soybean meal based diets. The use of wheat grains in broiler feed is limited by the presence of soluble non-starch polysaccharides, particularly xylans and arabinoxylans components (Pourreza *et al.*, 2007). These compounds reduce the nutritive value of wheat by increasing gut viscosity and thus reducing the availability of nutrients for digestion and absorption (Choct and Annison, 1992). Marquardt *et al.* (1996) reported that wheat-based diets supplemented with exogenous xylanase could deliver identical, or even better, growth and feed conversion rate

than unsupplemented corn-based diets. The improvement in performance is closely associated with improvement in nutrient and energy utilization from the feed. Energy utilization in poultry is usually expressed in terms of AME which accounts for energy loss in the excreta.

Due to the chemical structure of plant cell wall matrix, NSP degrading enzymes has been recommended to enhance poultries performance. Enzyme supplementation is well documented as effective in breaking polymeric chains of NSPs and hence improving the nutritive value of feedstuffs (Giraldo *et al.*, 2008). Therefore adding NSP-degrading enzymes in poultry diets has increased considerably in recent years. Birds do not produce enzymes like cellulase, xylanase, required for the digestion of NSPs. Supplementation of NSPs degrading enzymes may not only reduce the anti nutritive effects of NSPs, but also releases some nutrients from these, which could be utilized by the birds (Balamurugan and Chandrasekaran, 2009). However, the effects of exogenous enzymes can be variable and it depends on a large number of factors such as the age of the bird and the quality and type of diet (Bedford, 2000 and Acamovic, 2001).

Many commercial multienzyme complex are available in the Egyptian market that has the potential to improve the nutritional value of protein-rich vegetable feed ingredients. However, the few studies concerning this multienzyme complex generally show inconsistent results as regards broiler performance and nutrient digestibility (Ebert *et al.*, 2000 and Fischer *et al.*, 2002). Such studies were conducted with broilers fed with corn-soybean based diets and did not consider different inclusion of wheat grain instead of corn in broiler chicken ration under Egyptian environmental conditions. However, very few studies considered the effect of wheat-soybean diet on immune response and blood biochemical changes in broiler. Thus, the objective of this research was to evaluate the effect of partial replacement of corn y wheat grain without or with different commercial multienzyme complex products of broiler chicken ration on growth performance, immune response, some blood serum parameters and carcass quality.

## 2. MATERIALS AND METHODS

This work was conducted at Nutrition and Clinical Nutrition Department, Faculty of Veterinary Medicine, Alexandria University to investigate the possible effect of partial replacement of corn grain by wheat grain without or with different enzyme compound supplementation on broiler chicks growth performance, immune response and carcass quality, as well as some blood biochemical indices.

**Birds used:** A total of 450 one-day-old *Cobb* broiler chicks of mixed sex were used in this experiment. They were obtained from a local Egyptian private hatchery. The broiler chicks were randomly allotted into 9 equal groups (50 chicks/group).

**Accommodation and Management:** The broiler chicks were housed in a clean well-ventilated room previously disinfected by fumigation using formaldehyde gas produced by mixing formalin 40 % with potassium permanganate powder. The room was provided by gas heater in addition to electric lamps of 200 watt over each partition to obtain the suitable temperature needed for broiler chicks. The room floor was partitioned into 9 partitions. Each compartment was bedded by fresh clean wheat straw forming a deep litter of four centimeters depth. Each compartment was provided by suitable feeder and waterer. Prophylactic measures against the most common infectious diseases were carried out by using colistine sulfate (1g/4 liters) for Salmonellosis & E.coli infections during the first three days, Zinc bacitracine (at 0.15% of diet) for clostridia infection and Coxistac (at 0.1% of diet) for coccidiosis. The chicks were vaccinated against Newcastle disease using different types of Newcastle disease vaccines at 7<sup>th</sup>, 12<sup>th</sup>, 28<sup>th</sup> days of age. After vaccination the broiler chicks received AD<sub>3</sub>E vitamins (1 ml/L of drinking water) to improve vitality of chicks.

**Experimental design and feeding program:** The ingredient composition

and chemical analysis of the basal diet used in the experiment are presented in tables 1 and 2 respectively, while the applied experimental design is shown in table 3.

**Measurements:** Body weight development, body weight gain and feed intake of broiler chicks in different groups were. Feed Conversion Ratio (FCR), Protein Efficiency Ratio (PER) and Efficiency of Energy Utilization (EEU) were calculated according to Lambert *et al.* (1936); McDonald *et al.* (1987) and North (1981) respectively.

**Evaluation of immune response:** Immune response of birds was estimated by a group of parameters including Phagocytic activity, phagocytic index and HI tests for Newcastle Disease vaccine (NDV).

**Phagocytic activity and phagocytic index:** These parameters were determined according to Kawahara *et al.* (1991). Fifty micrograms of *Candida albicans* culture was added to 1 ml of citrated blood collected at the end of experiment (42 days). Treated blood samples were put in shaker water bath at 23 – 25° C for 3 – 5 hrs. Smears of blood were made and then stained with Geimsa stain. Phagocytosis was estimated by determining the proportion of macrophages which contain intracellular yeast cells in a random sample of 300 macrophages and expressed as percentage of phagocytic activity (PA). The number of phagocytized candida cells was counted in the phagocytic cells to calculate the phagocytic index according to the following equations : Phagocytic activity (PA) = Macrophages containing yeast/Total number of Macrophages X100. Phagocytic index (PI)= Number of cells phagocytized/Number of phagocytic cells.

**Haemagglutination inhibition tests for Newcastle disease (ND) antibodies :** Blood samples were taken at days 14<sup>th</sup>, 21<sup>th</sup>, 28<sup>th</sup>, 35<sup>th</sup> and 42<sup>th</sup> for Newcastle Disease antibodies and Blood samples were left without anticoagulant to clot. The serum was separated by centrifugation at 3000 rpm for 10 minutes. Microtechnique of haemagglutination inhibition test was done according to Takatasy (1955). Geometric mean titer (GMT) was calculated according to Brugh (1978).

**Blood samples:** At the end of the experimental period, blood samples were taken from 5 birds of different groups. The blood samples were left to drop on the side of the tube to prevent destruction of RBCs. Each blood sample was left to coagulate at room temp. Separation of serum was carried out by centrifugation of coagulated blood at 3000 rpm for 10 min. The clear serum was transferred carefully to clean and dry vials and kept in deep freezer until analysis for determination of serum glucose, total serum protein, albumin, globulin, triglycerides, cholesterol, GOT & GPT, uric acid and creatinine according to Trinder (1969), Doumas et al. (1981), Reinhold (1953), Coles (1974), Sidney and Barnard (1973), Zak et al. (1954), Reitman and Frankel (1957), (Fossatti and Prencipe, 1980) and Giorgio (1974) respectively.

**Carcass characteristic:** At the end of the experimental period, 5 chicks from each replicate of different groups were randomly selected and scarified to calculated the carcass and dressing percentages, also collect the liver, heart, gizzard, spleen, bursa, thymus gland, abdominal fat and Total edible carcass (TEC) and relative weight of each organ was calculated as follows:  $\text{Relative weight} = (\text{organ weight}/\text{Live body weight}) \times 100$ .

**Analytical methods:** Analytical DM contents of diets samples were determined by oven-drying at 105°C for 48h (AOAC, 1985). Ash contents of samples were determined by incineration at 550°C overnight. Crude protein was determined by using Kjeldahl method according to Randhir and Pradhan (1981) and ether extract was determined according to Bligh and Dyer (1959) technique as modified by Hanson and Olly (1963).

**Statistical analysis:** The analysis of variance for the obtained data was performed using Statistical Analysis System (SAS, 1996) to assess significant differences.

### **3. Results and Discussion:**

**3.1. Body Weight Development:** Effect of dietary partial replacement of corn with or with enzyme supplementation on body weight development of broiler chicken is presented in table 4. It was noticed that at the end of the growing period that enzyme supplementation had no significant effect on final body weight of broiler chicken fed on

corn – soybean based diet. These data are in agreement with those obtained by Vahjen et al. (2005) reported that multi enzyme (protease, amylase and cellulase) supplementation to the corn-soy based broiler rations did not affect the final body weight Similarly, Garipoglu et al. (2006) found no effect of multi enzyme supplementation on performance of broilers. Conversely, many researchers (Mathlouthi et al., 2002; Odetallah et al., 2003; Kocher et al., 2003) reported that multi enzyme supplementation improved the live body weight. The lack of effect of multi-enzyme addition on live body weight in this study can be attributed to the insufficiency of enzyme activity and dosage of enzyme used.

Replacement of corn grain by wheat grain at 25% from corn inclusion rate of the broiler chicken ration non significantly ( $P \geq 0.05$ ) reduced final body weight by about 5.5% while higher replacement rate improved ( $P \geq 0.05$ ) broiler weight by about 2.8% when compared with broiler chick group fed on the basal diet based on corn grain only. These data are supported by those obtained by Zarghi and Golian (2009) they revealed that diets containing up to 40% triticale (or 75% of corn replacement) as substitute of corn grain had no negative effect on broiler performance. Moreover, Mateo and Carandang (2006) who indicated that that the feeding value of low tannin sorghum and wheat grains relative to corn were 98% and 96%, respectively. Other studies showed that production of birds, even when fed diets contained of 100% triticale of the grain portion was similar to those fed control diet (Chapman *et al.*, 2005). It was noticed that kemzyme plus (enzyme blend based on xylanase, B.glucanase and amylase enzymes) or combozyme (enzyme blend based on cellulose, hemicellulase, lipase, protease, xylanase, B.glucanase and amylase enzymes) supplementation in broiler diet containing wheat grain at 25% from the corn percent of the ration significantly ( $P \leq 0.05$ ) improved final chick body weight by about 10.6% and 15.89% respectively when fed with broiler chick group fed on the same diet without enzyme supplementation.

**Table 1:** Ingredient composition of the used basal diet.

Ingredients	Feed Type								
	Starter diet (wheat inclusion)			Grower diet (wheat inclusion)			Finisher diet (wheat inclusion)		
	0.0	25%	50%	0.0	25%	50%	0.0	25%	50%
Yellow corn grain	55.3	41.35	27.8	60.34	45.36	30.36	63.13	47.16	31.66
Wheat grain	0.0	13.75	27.5	0.0	15.0	30.0	0.0	15.5	31.0
Soybean meal (44%)	33.59	33.29	33.29	28.95	28.45	28.45	25.78	25.68	25.68
Corn gluten (60%)	4.12	4.12	4.12	3.5	3.5	3.5	3.82	3.82	3.82
Vegetable oil	2.5	2.8	2.8	3.00	3.48	3.48	3.25	3.82	3.82
DCP	2.0	2.0	2.0	1.85	1.85	1.85	1.72	1.72	1.72
Limestone	1.41	1.41	1.41	1.2	1.2	1.2	1.15	1.15	1.15
Lysine	0.23	0.23	0.23	0.29	0.29	0.29	0.28	0.28	0.28
DL-Methionine	0.15	0.15	0.15	0.17	0.17	0.17	0.17	0.17	0.17
Salt	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4
Premix*	0.30	0.30	0.30	0.3	0.3	0.3	0.3	0.3	0.3
Total	100	100	100	100	100	100	100	100	100

\*The premix used was Heromix produced by Heropharm and composed of (per 3 kg) vitamin A 1200000 IU, vitamin D3 250000 IU, vitamin E 10000 mg, vitamin K3 2000 mg, thiamin 1000 mg, riboflavin 5000 mg, pyridoxine 1500 mg, cyanocobalamine 10 mg, niacin 30000 mg, biotin 50 mg, folic acid 1000 mg, pantothenic acid 10000 mg, manganese 60000 mg, zinc 50000 mg, iron 30000 mg, copper 4000 mg, iodine 300 mg, selenium 100 mg and cobalt 100 mg.

**Table 3:** Chemical analysis of the used basal diet.

Items	Feed Type								
	Starter diet (wheat inclusion rate)			Grower diet (wheat inclusion rate)			Finisher diet (wheat inclusion rate)		
	0.0	25%	50%	0.0	25%	50%	0.0	25%	50%
Dry matter%	88.43	88.02	89.11	89.22	89.32	88.65	88.98	88.66	89.27
Crude protein%	21.89	22.03	22.12	20.04	19.99	20.09	18.49	18.91	18.78
Ether extract%	5.16	5.33	5.62	5.37	5.64	6.04	6.26	6.43	6.29
Ash%	6.45	7.08	6.74	6.28	6.89	6.43	6.39	6.55	6.78
Calcium%*	1.08	1.08	1.09	0.96	0.97	0.97	0.90	0.91	0.91
Av. Phosphorus%*	0.51	0.52	0.52	0.48	0.49	0.49	0.45	0.46	0.46
Lysine%*	1.27	1.28	1.29	1.20	1.20	1.20	1.12	1.12	1.12
Methionine%*	0.52	0.52	0.52	0.51	0.51	0.51	0.50	0.50	0.50
ME Kcal/kg diet*	2950	2940	2919	3033	3042	3011	3072	3086	3055
Calorie/protein ratio	134.76	133.45	131.96	151.35	152.18	149.88	166.14	163.19	162.67

\* Calculated according to the feed composition tables given in NRC (1994).

**Table 3: Outline of the experimental design.**

Groups No.	Wheat grain inclusion rate				Enzyme supplementation (g/ton feed)	
	% of corn inclusion rate	% of the total ration			Kemzyme plus	Combozyme
		Starter diet	Grower diet	Finisher diet		
1	0.0	0.0	0.0	0.0	--	--
2	0.0	0.0	0.0	0.0	200 g	--
3	0.0	0.0	0.0	0.0	--	200 g
4	25.0	13.75	15.0	15.5	--	--
5	25.0	13.75	15.0	15.5	200 g	--
6	25.0	13.75	15.0	15.5	--	200 g
7	50.0	27.5	30.0	31.0	--	--
8	50.0	27.5	30.0	31.0	200 g	--
9	50.0	27.5	30.0	31.0	--	200

Kemzyme plus-dry (E 1620): Produced by : KEMIN INDUSTRIES (ASIA) and composed from the following active substances: endo- 1,3(4) betaglucanase(EC 3.2.1.6); endo -1,4-betaglucanase(EC 3.2.1.4); alpha-amylase(EC 3.2.1.1); endo -1,4-beta xylanase (EC 3.2.1.8) and bacillolysins (EC 3.4.24.28). The product used according to the manufacturer recommendation (200g/ton). Combozyme: Produced by American Bio system,inc (ENZYME BLEND) and composed from the following active substances: Cellulase: 75000 CU units. Fungal Amylase : 30,000 SKB units. Fungal Proteases : 1 Million HUT units. Neutral Proteases :100,000 PC units. Alkaline Proteases : 1.2 Anson units. Xylanase:20,000 XU units. B.Glucanase:20,000 BG units. Hemicellulase :20,000 HC units .Lipase:75,000 FIB units. The product used according to the manufacturer recommendation (200g/ton)

**Table 4: Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on body weight development (g/chicks) of broiler chickens.**

Age/ Weeks	Wheat grain level (% from corn content)	Enzyme supplementation		
		Without enzyme	Kemzyme plus Dry	Combozyme
0 day	0.0	40.40±0.31 <sup>ax</sup>	38.78±0.32 <sup>ax</sup>	38.58±0.26 <sup>ax</sup>
	25.0	37.98±0.23 <sup>ax</sup>	39.22±0.26 <sup>ax</sup>	38.76±0.27 <sup>ax</sup>
	50.0	39.40±0.31 <sup>ax</sup>	38.82±0.27 <sup>ax</sup>	39.92±0.31 <sup>ax</sup>
Week1	0.0	157.78±4.9 <sup>bx</sup>	156.85±4 <sup>bx</sup>	160.57±4.6 <sup>bx</sup>
	25.0	174.21±4 <sup>ay</sup>	188.88±3.9 <sup>ax</sup>	174.22±4.8 <sup>ay</sup>
	50.0	173.18±4.19 <sup>ay</sup>	166.72±4.2 <sup>by</sup>	185.16±3.4 <sup>ax</sup>
Week2	0.0	329.06±11.52 <sup>by</sup>	327.78±9.53 <sup>by</sup>	363.27±7.2 <sup>ax</sup>
	25.0	352.81±9.11 <sup>by</sup>	385.21±8.41 <sup>ax</sup>	363.46±11.07 <sup>ax</sup>
	50.0	368.50±9.67 <sup>ax</sup>	346.60±9.44 <sup>by</sup>	384.98±7.78 <sup>ax</sup>
Week3	0.0	547.78±14.13 <sup>bx</sup>	527.13±14.3 <sup>bx</sup>	535.71±13.6 <sup>bx</sup>
	25.0	554.48±12.93 <sup>by</sup>	625.74±12.93 <sup>ax</sup>	577.29±16.9 <sup>by</sup>
	50.0	615.50±15.48 <sup>ax</sup>	520.10±11.52 <sup>by</sup>	638.20±12.71 <sup>ax</sup>
Week4	0.0	904.33±20.52 <sup>abx</sup>	824.22±25.47 <sup>by</sup>	868.85±18.84 <sup>x<sup>by</sup></sup>
	25.0	845.73±19.02 <sup>by</sup>	991.43±20.85 <sup>ax</sup>	1001.38±26.49 <sup>ax</sup>
	50.0	961.46±26.74 <sup>ax</sup>	889.46±20.63 <sup>by</sup>	1013.65±20.94 <sup>ax</sup>
Week5	0.0	1368.11±28.93 <sup>bx</sup>	1448.55±29.33 <sup>ax</sup>	1422.29±20.82 <sup>ax</sup>
	25.0	1281.22±25.67 <sup>bz</sup>	1435.44±29.29 <sup>ay</sup>	1502.23±32.05 <sup>ax</sup>
	50.0	1497.88±35.78 <sup>ax</sup>	1483.57±37.75 <sup>ax</sup>	1516.60±33.76 <sup>ax</sup>
Week6	0.0	1913.97±25.68 <sup>aby</sup>	1889.33±37.05 <sup>by</sup>	2084.62±19.08 <sup>ax</sup>
	25.0	1808.02±32.69 <sup>by</sup>	2000.45±43.28 <sup>ax</sup>	2095.33±40.8 <sup>ax</sup>
	50.0	1966.76±27.36 <sup>ax</sup>	1935.47±40.66 <sup>ax</sup>	2030.26±29.08 <sup>ax</sup>

Values are means ± standard error. Mean values with different letters at the same column (a - c letters) or row (x - z letters) and period differ significantly at (P≤0.05).

However, enzyme supplementation in broiler diet containing higher inclusion rate of wheat grain had no significant effect on body weight of broiler chicken. The lower response with higher inclusion rate of wheat grain may be related to lower enzyme

dosage. The obtained data supported by Madrid et al. (2010) they reported that multienzyme complex containing protease and carbohydrase enzymes supplementation in wheat-soybean meal based diets formulated to exceed nutrient

recommendations did not significantly improve performance when birds were kept in ideal conditions.

On the other hand, it was noticed that broiler chicken fed on different levels (25 and 50%) of wheat grain (substitute to corn grain) higher respond with different enzyme compound (Kemzyme plus or combozyme) supplementation by about (5.9% and 2.4%) and (2.96% and -2.6%) respectively when compared with those fed on corn-soybean based diet. The obtained data are in harmony with (Pourreza *et al.*, 2007 and Zarghi *et al.* 2010) they concluded that the addition of enzyme significantly improved the performance of broiler chickens fed triticale- soy diet as compared to corn-soy diet. This effect could suggest that the effect of multienzyme complex is more noticeable in diets based on ingredients with a high content of the targeted molecules.

### 3.2. Growth Performance Parameters:

Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on final body weight, body gain, feed intake, feed conversion ratio (FCR), protein efficiency ratio (PER), efficiency of energy utilization (EEU) and performance index of broiler chickens are presented in table 5. Statistical analysis of the obtained data indicated that kemzyme plus dry supplementation in corn – soybean based diet non significantly reduced body gain throughout the whole experimental period by about 1.2%, while combozyme supplementation significantly ( $P \leq 0.05$ ) improved total gain by about 9.2% when compared with broiler chicken fed on the same diet without enzyme supplementation. It was observed that wheat inclusion at 25% as corn substitute decreased ( $P \geq 0.05$ ) body gain by about 5.5% while, higher inclusion level of wheat grain improved ( $P \leq 0.05$ ) total gain by about (2.9%) when compared with broiler chick group fed on corn soybean based diet without enzyme supplementation. Moreover, kemzyme plus dry or combozyme enzyme blend products supplementation significantly improved body gain of broiler chicken fed on 25% wheat grain instead of corn by about 10.8% and 16.1% respectively, while enzyme

supplementation in broiler diet containing 50% instead of corn non significantly improved body gain when compared with broiler chicken group fed on the same diet without enzyme supplementation.

Both kemzyme plus dry and combozyme enzyme products supplementation to corn – soybean based diet had no significant effect on feed intake however, improved ( $P \leq 0.05$ ) feed conversion ratio (FCR), protein efficiency ratio (PER) and efficiency of energy utilization (EEU) by about (7.4% and 7.4%), (7.1% and 6.5%) and (7.5% and 7.5%) respectively, on the other hand enzyme supplementation non significantly improved performance index (PI) of broiler chicks when compared with broiler chicken group fed on the same diet without enzyme supplementation. The obtained data are in agreement with (Gutierrez del Alamo *et al.*, 2008; and Hajati, 2010) who observed that enzyme inclusion of broiler chicken diet based on corn and soybean meal increased energy and protein efficiency and decreased feed intake, body weight gain and feed to gain ratio from 29-44 days of age ( $p < 0.05$ ). Relative growth, energy efficiency and protein efficiency was increased and feed intake and feed to gain ratio was decreased by enzyme supplementation from 1-44 days ( $p < 0.05$ ). It was observed that wheat inclusion at 25 or 50% instead of corn grain in broiler chicken ration increased feed intake by about 1.3% and 9.8% respectively and consequently deteriorate FCR, PER, EEU and PI by about (4.6% and 4.6%), (8.2% and 6.5%), (4.9% and 4.0%) and (13.7% and (4.2%) respectively when compared with broiler chicken group fed on corn – soybean based diet. Moreover, enzyme supplementation in 25% wheat containing diet increased feed intake but, with 50% wheat inclusion decreased feed intake also, enzyme supplementation in wheat included ration improved FCR, PER, EEU and PI when compared with broiler chick group fed on the same diet without enzyme supplementation.

Table 5: Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on growth performance parameters of broiler chickens.

Items	Wheat grain level (% from corn content)	Enzyme supplementation		
		Without enzyme	Kemzyme Plus Dry	Combozyme
Initial body weight	0.0	40.40±0.31 <sup>ax</sup>	38.78±0.32 <sup>ax</sup>	38.58±0.26 <sup>ax</sup>
	25.0	37.98±0.23 <sup>ax</sup>	39.22±0.26 <sup>ax</sup>	38.76±0.27 <sup>ax</sup>
	50.0	39.40±0.31 <sup>ax</sup>	38.82±0.27 <sup>ax</sup>	39.92±0.31 <sup>ax</sup>
Final weight (g/bird)	0.0	1913.97±39.9 <sup>aby</sup>	1889.33±37.04 <sup>by</sup>	2084.62±19.08 <sup>ax</sup>
	25.0	1808.02±32.7 <sup>by</sup>	2000.45±43.3 <sup>ax</sup>	2095.33±40.8 <sup>ax</sup>
	50.0	1966.76±27.4 <sup>ax</sup>	1935.47±40.65 <sup>ax</sup>	2030.26±39.08 <sup>ax</sup>
Total gain (g/bird)	0.0	1873.95±39.62 <sup>by</sup>	1851.21±36.79 <sup>by</sup>	2046.72±18.9 <sup>ax</sup>
	25.0	1770.44±32.5 <sup>by</sup>	1961.70±43.1 <sup>ax</sup>	2055.89±36.6 <sup>ax</sup>
	50.0	1927.88±27.11 <sup>ax</sup>	1897.19±40.45 <sup>ax</sup>	1990.63±38.8 <sup>ax</sup>
Daily gain (g/bird)	0.0	44.62	44.08	48.73
	25.0	42.15	46.71	48.95
	50.0	45.90	45.17	47.40
Daily feed intake (g/bird)	0.0	64.95	59.59	66.79
	25.0	65.77	70.69	70.50
	50.0	70.85	58.54	68.27
AFCR	0.0	1.49±0.033 <sup>ax</sup>	1.38±0.028 <sup>by</sup>	1.38±0.012 <sup>by</sup>
	25.0	1.56±0.024 <sup>ax</sup>	1.55±0.035 <sup>ax</sup>	1.47±0.031 <sup>ax</sup>
	50.0	1.56±0.022 <sup>ax</sup>	1.32±0.026 <sup>by</sup>	1.47±0.031 <sup>ax</sup>
APER	0.0	3.53±0.07 <sup>ay</sup>	3.78±0.074 <sup>ax</sup>	3.76±0.035 <sup>ax</sup>
	25.0	3.24±0.07 <sup>bx</sup>	3.36±0.073 <sup>bx</sup>	3.53±0.07 <sup>bx</sup>
	50.0	3.30±0.046 <sup>bz</sup>	3.92±0.08 <sup>ax</sup>	3.53±0.07 <sup>by</sup>
AEEU	0.0	4.53±0.1 <sup>ax</sup>	4.19±0.09 <sup>by</sup>	4.19±0.036 <sup>by</sup>
	25.0	4.75±0.07 <sup>ax</sup>	4.74±0.1 <sup>ax</sup>	4.49±0.09 <sup>ax</sup>
	50.0	4.71±0.07 <sup>ax</sup>	3.99±0.08 <sup>by</sup>	4.44±0.09 <sup>ax</sup>
API	0.0	134.32±5.55 <sup>ax</sup>	142.39±5.49 <sup>ax</sup>	152.73±2.89 <sup>ax</sup>
	25.0	115.89±5.20 <sup>by</sup>	135.25±5.82 <sup>ax</sup>	148.21±5.68 <sup>ax</sup>
	50.0	128.63±3.6 <sup>aby</sup>	152.62±6.72 <sup>ax</sup>	143.53±5.31 <sup>axy</sup>

Values are means± standard error. Mean values with different letters at the same column (a - c letters) or row (x - z letters) and period differ significantly at (P≤0.05).

The poor FCR of birds fed wheat containing diets may be related to lower nutrients, lower nutrient digestibility or higher anti-nutrient factors in wheat grain as compared to corn. The anti-nutritional factors in wheat includes: soluble pentosans, trypsin inhibitor, alkyl-resorcinols and pectins (Korver *et al.*, 2004). The present are supported y those obtained by Madrid *et al.*, (2010) they reported that that the multienzyme complex of protease and carbohydrase enzymes might be effective for improving broiler chickens growth performance parameters and nutrient digestibility in broilers fed with a wheat-soybean meal based diet under commercial farm conditions. poorer FCR was observed in birds fed triticale-based as compared to

those fed corn-based diet (Hermes and Johnson, 2004). Similarly, Vieira *et al.* (1995) reported that the inclusion level of triticale up to 40% in corn-soy diet did not have any negative effect on weight gain or final weight of broiler chickens. The different results obtained in our experiment and other reports may be due to the type of enzyme cocktail and/or variety of wheat used in this experiment as compared to other experiments. Specially, the new varieties of wheat may contain less anti-nutrients such as pentosans and other non-starch polysaccharides.



**Table 6:** Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on some blood serum parameters of broiler chickens.

Items	Wheat grain level (% from corn content)	Enzyme supplementation		
		Without enzyme	Kemzyme plus	Combozyme
Total protein	0.0	4.75±0.49 <sup>ax</sup>	5.43±0.42 <sup>ax</sup>	4.50±0.26 <sup>ax</sup>
	25.0	4.45±0.47 <sup>ax</sup>	5.60±0.43 <sup>ax</sup>	5.40±0.46 <sup>ax</sup>
	50.0	5.00±0.34 <sup>ax</sup>	4.65±0.46 <sup>ax</sup>	5.13±0.76 <sup>ax</sup>
Albumin	0.0	3.50±0.1 <sup>ax</sup>	3.41±0.38 <sup>ax</sup>	3.05±0.36 <sup>ax</sup>
	25.0	3.60±0.26 <sup>ax</sup>	3.95±0.41 <sup>ax</sup>	3.68±0.3 <sup>ax</sup>
	50.0	3.70±0.05 <sup>ax</sup>	3.63±0.07 <sup>ax</sup>	3.65±0.61 <sup>ax</sup>
Globulin	0.0	1.25±0.39 <sup>ax</sup>	2.01±0.53 <sup>ax</sup>	1.35±0.14 <sup>ax</sup>
	25.0	0.85±0.26 <sup>ax</sup>	1.65±0.41 <sup>ax</sup>	1.73±0.37 <sup>ax</sup>
	50.0	1.28±0.36 <sup>ax</sup>	1.03±0.50 <sup>ax</sup>	1.48±0.45 <sup>ax</sup>
A/G ratio	0.0	3.82±0.97 <sup>ax</sup>	1.84±0.72 <sup>ax</sup>	2.25±0.47 <sup>ax</sup>
	25.0	4.48±1.4 <sup>ax</sup>	2.89±0.59 <sup>ax</sup>	2.24±0.64 <sup>ax</sup>
	50.0	2.87±0.54 <sup>ax</sup>	4.05±2.21 <sup>ax</sup>	3.48±1.09 <sup>ax</sup>
Glucose	0.0	71.06±16.88 <sup>ax</sup>	71.14±14.46 <sup>ax</sup>	76.84±16.05 <sup>ax</sup>
	25.0	78.68±15.61 <sup>ax</sup>	77.54±16.44 <sup>ax</sup>	78.80±16.04 <sup>ax</sup>
	50.0	69.32±13.53 <sup>ax</sup>	86.80±17.40 <sup>ax</sup>	72.26±14.21 <sup>ax</sup>

Values are means ± standard error. Mean values with different letters at the same column (a - c letters) or row (x - z letters) and period differ significantly at ( $P \leq 0.05$ ).

**3.3. Blood serum Units:** Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on blood serum total protein, albumin, globulin and glucose concentrations of broiler chickens are presented in table 6. Analysis of variance of the obtained data indicated that enzyme supplementation in corn – soybean based broiler diet had no significant on blood serum total protein or albumin concentrations when compared with broiler chicken group fed on the same diet without enzyme supplementation. Corn substitution by wheat grain at 25% or 50% non significantly reduced total protein concentration and increased ( $P \geq 0.05$ ) blood serum albumin and globulin concentrations when compared with broiler chicken fed on corn – soybean based diet. Moreover, enzyme supplementation in wheat grain containing ration had no significant on blood serum total protein, albumin and globulin concentrations when compared with broiler chicken group fed on the same diet without enzyme supplementation. Regarding blood serum glucose concentration, it was noticed that corn substitution by wheat grain and enzyme supplementation in broiler diet based on corn – soybean or containing different levels of wheat grain had no significant effect on blood serum glucose

concentration when compared with broiler chicken group fed on the same diet without enzyme supplementation. The obtained results are in agreement with Lee et al. (2010) they reported that the concentrations of albumin in serum were not influenced by the wheat inclusion and enzyme treatment in broiler ration.

### 3.4. Blood serum lipid concentrations:

Effect of partial replacement of corn by wheat grain without or with enzyme supplementation on blood serum lipid concentrations are presented in table 7. the obtained data revealed that wheat inclusion as substitute of corn grain at 25 or 50% non significantly reduced blood serum triglyceride, total cholesterol and high density lipoprotein (HDL) concentrations by about (13.3% and 16.8%), (3.4% and 6.9%) and (6.8% and 36.8%) respectively when compared with chicken group fed on corn-soybean based diet without enzyme supplementation. On contrast, wheat inclusion at 25 or 50% as corn substitutes non significantly ( $P \geq 0.05$ ) increased blood serum low density lipoprotein (LDL) by about 3.3% and 18.4% respectively. The obtained data are in harmony with those reported by Zarghi et al. (2010) they noticed that there were not significant differences in TG, cholesterol and LDL between birds fed

diet contained different levels of triticale but the blood serum HDL significantly ( $p < 0.01$ ) decreased in birds fed diet with 75% triticale than those fed corn-soy diet. The inclusion of highly viscous grain in diet induced bird plasma cholesterol and HDL levels as compared to control one. Soluble dietary fibers such as mixed linked  $\beta$ -

Glucans, may reduce the absorption of fat and cholesterol and are known to have cholesterol lowering properties. These effects are all associated with the viscosity forming properties of soluble dietary fibers (Pettersson and Aman 1993).

**Table 7:** Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on some blood serum lipids concentrations of broiler chickens.

Items	Wheat grain level (% from corn content)	Enzyme supplementation		
		Without enzyme	Kemzyme plus	Combozyme
Triglycerides	0.0	173.55±13.39 <sup>ax</sup>	150.28±6.28 <sup>ax</sup>	154.83±22.48 <sup>ax</sup>
	25.0	150.46±5.12 <sup>ax</sup>	181.88±12.57 <sup>ax</sup>	155.45±10.94 <sup>ax</sup>
	50.0	144.38±21.33 <sup>ax</sup>	137.90±27.75 <sup>ax</sup>	183.20±5.29 <sup>ax</sup>
Total cholesterol	0.0	174.80±13.25 <sup>ax</sup>	151.23±4.43 <sup>ax</sup>	171.00±16.28 <sup>ax</sup>
	25.0	168.80±14.14 <sup>ax</sup>	169.86±7.55 <sup>ax</sup>	170.85±6.00 <sup>ax</sup>
	50.0	162.80±4.89 <sup>ax</sup>	156.78±7.63 <sup>ax</sup>	170.50±10.78 <sup>ax</sup>
HDL	0.0	58.50±3.87.0 <sup>ax</sup>	40.50±3.89 <sup>ax</sup>	41.00±9.28 <sup>ax</sup>
	25.0	54.50±4.00 <sup>ax</sup>	51.00±5.69.0 <sup>ax</sup>	52.25±3.35 <sup>ax</sup>
	50.0	37.00±6.38.0 <sup>ax</sup>	42.00±9.74 <sup>ax</sup>	38.00±6.61 <sup>ax</sup>
LDL	0.0	81.59±10.85 <sup>ax</sup>	80.67±6.27 <sup>ax</sup>	99.03±10.86 <sup>ax</sup>
	25.0	84.21±10.45 <sup>ax</sup>	82.48±1.97 <sup>ax</sup>	87.51±10.11 <sup>ax</sup>
	50.0	96.93±3.43 <sup>ax</sup>	87.20±10.30 <sup>ax</sup>	95.86±17.80 <sup>ax</sup>

Values are means  $\pm$  standard error. Mean values with different letters at the same column (a - c letters) or row (x - z letters) and period differ significantly at ( $P \leq 0.05$ ).

Enzyme supplementation in broiler diet containing wheat grain as corn substitutes increased ( $P \geq 0.05$ ) blood serum triglyceride and total cholesterol when compared with chicken group fed on the same diet without enzyme supplementation. However, enzyme supplementation non significantly reduced blood serum triglyceride and cholesterol concentration in broiler chicken fed on corn-soybean based diet when compared with broiler chick group fed on the same diet without enzyme supplementation. The present data are supported by those obtained by Lee et al. (2010) they stated that enzyme supplementation had no significant effect on blood serum cholesterol concentration of broiler chickens. On the other hand these results are in contrast with those obtained by Zarghi and Golian (2009) they indicated that the chickens fed diet supplemented with 500 ppm of enzyme cocktail contained min of 1200 unit  $g^{-1}$  xylanase and 440 units  $g^{-1}$   $\beta$ -glucanase, had higher ( $p < 0.05$ ) cholesterol concentration than birds fed diet

without enzyme addition (143 vs. 134 mg  $mL^{-1}$ ).

**3.5. Liver and kidney function parameters:** Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on some blood serum parameters related to liver and kidney functions of broiler chickens are presented in table 8. Statistical analysis of the obtained data revealed that 25% and 50% of corn replacement by wheat grain without enzyme supplementation significantly increased blood serum GOT concentration of broiler chicken (table 7) by about 94.8% and 272.9% respectively when compared with broiler chick group fed on corn-soybean based diet without enzyme supplementation. However, wheat grain inclusion at 25 or 50% from corn content of broiler chicken ration non significantly ( $P \geq 0.05$ ) increased blood serum GPT concentration by about 8.0% and 17.5% respectively. These data indicated that wheat inclusion in broiler chicken ration had adverse effect on hepatic cell and broiler health condition.

**Table 8:** Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on some blood serum parameters related to liver functions of broiler chickens.

Items	Wheat grain level (% from corn content)	Enzyme supplementation		
		Without enzyme	Kemzyme plus	Combozyme
GOT	0.0	38.75±0.92 <sup>cx</sup>	42.75±5.16 <sup>bx</sup>	51.75±3.76 <sup>bx</sup>
	25.0	75.50±8.10 <sup>bx</sup>	81.25±4.02 <sup>ax</sup>	84.00±2.22 <sup>ax</sup>
	50.0	105.75±14.4 <sup>ax</sup>	101.25±4.02 <sup>ax</sup>	96.50±14.13 <sup>ax</sup>
GPT	0.0	65.75±2.08 <sup>ax</sup>	68.25±3.69 <sup>ax</sup>	71.75±4.34 <sup>ax</sup>
	25.0	71.00±5.48 <sup>ax</sup>	72.25±3.52 <sup>ax</sup>	69.50±4.09 <sup>ax</sup>
	50.0	77.25±6.82 <sup>ax</sup>	71.25±4.62 <sup>ax</sup>	71.75±2.56 <sup>ax</sup>
Uric acid	0.0	5.97±0.43 <sup>ax</sup>	5.91±0.14 <sup>ax</sup>	6.18±0.15 <sup>ax</sup>
	25.0	6.02±0.31 <sup>ax</sup>	6.51±0.22 <sup>ax</sup>	6.29±0.21 <sup>ax</sup>
	50.0	6.50±0.18 <sup>ax</sup>	6.41±0.20 <sup>ax</sup>	5.87±0.19 <sup>ax</sup>
Creatinine	0.0	1.80±0.34 <sup>ax</sup>	2.80±0.62 <sup>ax</sup>	2.60±0.66 <sup>bx</sup>
	25.0	3.00±0.37 <sup>ax</sup>	3.10±0.59 <sup>ax</sup>	0.85±0.16 <sup>bx</sup>
	50.0	2.00±0.56 <sup>ax</sup>	2.10±0.66 <sup>ax</sup>	3.25±1.1 <sup>ax</sup>

Values are means ± standard error. Mean values with different letters at the same column (a - c letters) or row (x - z letters) and period differ significantly at ( $P \leq 0.05$ ).

Enzyme supplementation (both kemzyme plus or combozyme products) had no significant effect on blood serum GOT and GPT concentrations when compared with broiler chicken groups fed on the same diet without enzyme supplementation. The present data are supported by those obtained by Lee et al. (2010) who indicated that there were no significant differences in the activities of serum GOT and GPT among control and enzyme treated groups of broiler chickens. Measurement of GOT and GPT activities are indicative of liver damage in broiler chicks and is therefore a valuable tool for determination of a safe inclusion rate for feed additives. Based on these findings, enzyme blend administered at the levels evaluated in this study may not exert adverse effects on broiler chickens.

Regarding blood serum uric acid and creatinine concentration (Table ), it was noticed that enzyme supplementation in broiler chicken diet based on corn - soybean had no significant effect on blood serum uric acid when compared with broiler chicken fed on the same diet without enzyme supplementation. On the other hand, it was observed that inclusion of wheat grain at 25% or 50% instead of corn grain non significantly increased blood serum uric acid concentration by about 0.8% and 8.9% respectively when compared with broiler chicken group fed on corn - soybean based diet. Moreover,

enzyme supplementation non significantly decreased blood serum uric acid concentration of broiler chicken fed on wheat containing ration. These results are in harmony with those obtained by Hajati (2010) who concluded that enzyme supplementation was reduced the concentration of blood uric acid at 28 and 44 days ( $p < 0.05$ ). This suggests that the enzyme preparation increased nutrient metabolism, particularly protein anabolism of birds, therefore, promoting the growth of chickens. In birds, purine bases are degraded to uric acid. In addition, purines are formed from excess amino-N, which are subsequently degraded to uric acid and excreted in urine (Buyse et al., 2002). Correlation analysis showed a significant negative relationship between plasma uric acid levels and efficiency of protein retention (Swennen et al., 2005). So we can conclude that replacement of corn by wheat grain reduced efficiency of protein utilization and enzyme supplementation improve that effect. Moreover, it was observed that enzyme supplementation in corn - soybean based diet or in broiler diet containing wheat grain instead of corn had no significant effect on blood serum creatinine concentration when compared with broiler chicken groups fed on the same diet without enzyme supplementation. Also, corn substitution by wheat grain had no effect of creatinine blood serum

concentration. All creatinine concentration values are within the normal range and indicated that both wheat grain inclusion or enzyme supplementation had no adverse effect on broiler kidney function.

### 3.6. Immune response:

Phagocytosis: Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on phagocytosis of broiler chickens is presented in table 9. statistical analysis of the obtained data revealed that inclusion of wheat grain instead of corn at 25% non significantly reduced ( $P \geq 0.05$ ) phagocytic activity by about 2.4%, while higher inclusion level of wheat grain significantly ( $P \leq 0.05$ ) reduced phagocytic activity by about 14.5% when compared with broiler chick group fed on corn – soybean based diet without enzyme supplementation. However, 25% or 50% of corn content in broiler chick diet replacement by wheat grain non significantly reduced phagocytic

index by about 11.9% and 9.4% respectively. It was observed that supplementation of both kemzyme plus or combozyme in broiler chicken ration had no significant effect on phagocytic activity or index when compared with broiler chick group fed on the same diet without enzyme supplementation. The obtained data are supported by Soltan (2009) who indicated that enzyme supplementation had no significant effect on both phagocytic activity and index of broiler chickens when compared with the groups fed on the same diet without enzyme supplementation. Moreover, Abeer H. Abdel Razek and Tony (2013) Phagocytic percent and index of broiler chickens significant ( $p < 0.05$ ) increase in treated groups with preiotic and some digestive enzyme compared with control one at 5<sup>th</sup> day post 1<sup>st</sup> and 2<sup>nd</sup> vaccination. Also at 12<sup>th</sup> day post 2<sup>nd</sup> vaccination significantly increased as well.

**Table (9):** Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on phagocytosis of broiler chickens.

Items	Wheat grain level (% from corn content)	Enzyme supplementation		
		Without enzyme	Kemzyme plus Dry	Combozyme
Phagocytic activity	0.0	20.63±1.01 <sup>ax</sup>	20.75±1.11 <sup>ax</sup>	20.50±0.88 <sup>ax</sup>
	25.0	20.13±0.97 <sup>ax</sup>	20.75±1.09 <sup>ax</sup>	19.25±0.95 <sup>ax</sup>
	50.0	17.63±0.89 <sup>by</sup>	18.63±0.96 <sup>axy</sup>	20.63±0.79 <sup>ax</sup>
Phagocytic index	0.0	1.60±0.09 <sup>ax</sup>	1.36±0.11 <sup>ax</sup>	1.36±0.06 <sup>ax</sup>
	25.0	1.41±0.07 <sup>ax</sup>	1.45±0.06 <sup>ax</sup>	1.30±0.09 <sup>ax</sup>
	50.0	1.45±0.11 <sup>ax</sup>	1.50±0.08 <sup>ax</sup>	1.39±0.08 <sup>ax</sup>

Values are means ± standard error. Mean values with different letters at the same column (a - c letters) or row (x - z letters) and period differ significantly at ( $P \leq 0.05$ ).

Antibody titer against NDV: Inclusion of wheat grain in broiler chicken ration at 25% instead of corn grain significantly ( $P \leq 0.05$ ) reduced HI titer against NDV (table, 10) at 21th and 42th of broiler age by about 42.3% and 20% respectively when compared with broiler chicken group fed on corn – soybean based ration. Moreover, enzyme supplementation had no significant effect on HI titer against NDV when compared with broiler chicken group fed on the same diet without enzyme supplementation during different experimental period except during the last

week (6<sup>th</sup> week) of broiler age it was noticed that enzyme supplementation significantly improved HI titer against NDV of broiler chicken. Moreover, it was noticed that kemzyme plus supplementation more effective as immune stimulant and HI titer production against NDV when compared with chicken group fed on the same diet with combozyme supplementation. Antibody titer responses have been used as measures of humoral immune status of birds (Sklan *et al.*, 1994). The immune

system guards the body against foreign substances and protects from invasion by pathogenic organisms. It can be divided into the innate (nonspecific) immune system and the acquired (specific) immune system. The present data are in harmony with those obtained Soltan (2009) who concluded that enzyme had no effect ( $P>0.05$ ) HI titer of broiler chicks when compared with the chicks fed on the same diet containing different levels of palm kernel cakes without enzyme supplementation. Also, the obtained data are supported by those reported by Algedawy et al. (2011) they concluded that both probiotic plus enzyme containing product (Biogen) and enzyme mixture product (Natuzyne) supplementation non-significantly boosted the hemagglutination-inhibition antibody titer of broiler chicken against Newcastle disease and avian influenza vaccines.

To our knowledge, limited studies have been conducted to determine the effects of enzyme supplementation to wheat grain

containing diets on the immunity of poultry. There is much evidence demonstrating that inappropriate diet intake negatively influences the development of immune organs and normal immune responses and nutrients can modulate immune function in human and animals. Energy, proteins, lipids, vitamins, minerals and nucleic acids play an important role in the regulation of cellular and humoral important role in the regulation of cellular and humoral of the enzyme enhanced the digestion of feed and the absorption of nutrients, which in turn could have an effect on body immunity (Kidd, 2004). Results of present study show on body immunity (Kidd, 2004). Results of present study show that, NSP-hydrolyzing enzyme preparations (xylanase and B-glucanase and others enzymes) when added to wheat-corn-soy based diets, can improve the performance in broiler chickens. This improvement is achieved through the enzyme's influence on digestion, absorption, metabolism and immunity of broiler chicken.

**Table 10:** Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on blood serum antibody production against Newcastle disease vaccine of broiler chickens.

Period	Wheat grain level (% from corn content)	Enzyme supplementation		
		Without enzyme	Kemzyme plus	Combozyme
14 <sup>th</sup> day	0.0	3.40±0.51 <sup>ax</sup>	2.20±0.44 <sup>ax</sup>	1.80±0.37 <sup>ax</sup>
	25.0	2.20±0.73 <sup>ax</sup>	2.40±0.25 <sup>ax</sup>	3.00±0.44 <sup>ax</sup>
	50.0	2.80±0.73 <sup>ax</sup>	2.80±0.37 <sup>ax</sup>	2.80±0.86 <sup>ax</sup>
21 <sup>th</sup> day	0.0	5.20±0.37 <sup>axy</sup>	5.80±0.20 <sup>ax</sup>	4.60±0.24 <sup>ay</sup>
	25.0	3.00±0.32 <sup>bx</sup>	3.20±0.58 <sup>by</sup>	3.40±0.24 <sup>bx</sup>
	50.0		4.80±0.58 <sup>ax</sup>	3.60±0.24 <sup>ay</sup>
28 <sup>th</sup> day	0.0	5.00±0.31 <sup>abx</sup>	5.20±0.20 <sup>bx</sup>	4.60±0.24 <sup>bx</sup>
	25.0	4.60±0.24 <sup>by</sup>	6.60±0.24 <sup>ax</sup>	5.40±0.24 <sup>aby</sup>
	50.0	5.60±0.24 <sup>ay</sup>	6.40±0.24 <sup>ax</sup>	6.00±0.31 <sup>axy</sup>
35 <sup>th</sup> day	0.0	4.80±0.37 <sup>ax</sup>	5.00±0.31 <sup>ax</sup>	4.80±0.37 <sup>ax</sup>
	25.0	5.00±0.44 <sup>ax</sup>	6.00±0.44 <sup>ax</sup>	5.00±0.44 <sup>ax</sup>
	50.0	5.00±0.31 <sup>ax</sup>	5.80±0.58 <sup>ax</sup>	4.80±0.37 <sup>ax</sup>
42 <sup>th</sup> day	0.0	6.00±0.44 <sup>ax</sup>	6.00±0.44 <sup>bx</sup>	4.80±0.20 <sup>ay</sup>
	25.0	4.80±0.37 <sup>by</sup>	6.60±0.24 <sup>bx</sup>	5.80±0.37 <sup>axy</sup>
	50.0	6.00±0.44 <sup>ay</sup>	7.400±0.24 <sup>ax</sup>	5.80±0.37 <sup>ay</sup>

Values are means± standard error. Mean values with different letters at the same column (a - c letters) or row (x – z letters) and period differ significantly at ( $P\leq 0.05$ ).

**3.7. Some carcass traits:** Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on some immune organs weights and relative weights (weight as a percentage of live body weight) of broiler chickens are presented in table 11. Regarding of bursa weight and relative weight, it was observed that kemzyme plus dry or combozyme supplementation in corn – soybean based diet significantly reduced bursa weight and relative weight by about (35.2% and 35.2%) and (42.9% and 28.5%) respectively when compared with broiler chicken group fed on

the same diet without enzyme supplementation. On the other hand corn substitution by wheat grain at 25% or 50% non significantly reduced bursa weight and relative weight by about (29.7% and 15.6%) and (28.5% and 28.5%) respectively when compared with broiler chicken group fed on corn – soybean based diet. However, enzyme supplementation with wheat containing diet non significantly improved both bursa weight and relative weight when compared with broiler chicken group fed on the same diet without enzyme supplementation.

**Table 11:** Effect of dietary partial replacement of corn by wheat grain without or with enzyme supplementation on some immune organs weights and relative weights of broiler chickens.

Items	Wheat grain level (% from corn content)	Enzyme supplementation		
		Without enzyme	Kemzyme plus	Combozyme
Bursa weight	0.0	1.28±0.13 <sup>ax</sup>	0.83±0.11 <sup>ay</sup>	0.83±0.044 <sup>ay</sup>
	25.0	0.90±0.062 <sup>ax</sup>	0.98±0.17 <sup>ax</sup>	1.08±0.044 <sup>ax</sup>
	50.0	1.08±0.076 <sup>ax</sup>	1.13±0.09 <sup>ax</sup>	0.95±0.15 <sup>ax</sup>
Bursa relative weight	0.0	0.07±0.008 <sup>ax</sup>	0.04±0.0044 <sup>by</sup>	0.05±0.001 <sup>ay</sup>
	25.0	0.05±0.002 <sup>ax</sup>	0.05±0.008 <sup>ax</sup>	0.05±0.001 <sup>ax</sup>
	50.0	0.05±0.002 <sup>ax</sup>	0.07±0.008 <sup>ax</sup>	0.05±0.008 <sup>ay</sup>
Thymus gland weight	0.0	4.48±0.58 <sup>ax</sup>	6.10±2.08 <sup>ax</sup>	5.25±1.44 <sup>bx</sup>
	25.0	3.00±1.22 <sup>ax</sup>	7.08±1.408 <sup>ax</sup>	5.28±0.49 <sup>bx</sup>
	50.0	7.05±1.09 <sup>ax</sup>	5.83±1.18 <sup>ay</sup>	8.08±0.53 <sup>ax</sup>
Thymus gland relative weight	0.0	0.24±0.035 <sup>ax</sup>	0.33±0.120 <sup>ax</sup>	0.30±0.084 <sup>ax</sup>
	25.0	0.16±0.058 <sup>ax</sup>	0.36±0.067 <sup>ax</sup>	0.26±0.026 <sup>ax</sup>
	50.0	0.36±0.067 <sup>ax</sup>	0.35±0.076 <sup>ax</sup>	0.39±0.022 <sup>ax</sup>
Spleen weight	0.0	3.63±0.78 <sup>ax</sup>	3.05±0.74 <sup>ax</sup>	4.20±0.81 <sup>ax</sup>
	25.0	3.90±0.96 <sup>ax</sup>	3.48±1.05 <sup>ax</sup>	4.13±0.86 <sup>ax</sup>
	50.0	4.18±0.88 <sup>ax</sup>	2.10±0.18 <sup>ax</sup>	3.25±0.71 <sup>ax</sup>
Spleen relative weight	0.0	0.20±0.044 <sup>ax</sup>	0.16±0.035 <sup>ax</sup>	0.23±0.044 <sup>ax</sup>
	25.0	0.21±0.044 <sup>ax</sup>	0.17±0.044 <sup>ax</sup>	0.20±0.040 <sup>ax</sup>
	50.0	0.21±0.044 <sup>ax</sup>	0.12±0.044 <sup>ax</sup>	0.16±0.035 <sup>ax</sup>
Fat weight	0.0	35.62±9.70 <sup>ax</sup>	21.20±4.70 <sup>ax</sup>	22.80±4.88 <sup>ax</sup>
	25.0	27.89±4.74 <sup>ax</sup>	23.03±3.69 <sup>ax</sup>	23.68±4.11 <sup>ax</sup>
	50.0	24.20±4.72 <sup>ax</sup>	17.80±3.15 <sup>ax</sup>	23.58±3.46 <sup>ax</sup>
Fat relative weight	0.0	1.89±0.83 <sup>ax</sup>	1.14±0.32 <sup>ax</sup>	1.26±1.19 <sup>ax</sup>
	25.0	1.54±0.35 <sup>ax</sup>	1.12±0.55 <sup>ax</sup>	1.15±0.46 <sup>ax</sup>
	50.0	1.25±0.83 <sup>ax</sup>	1.06±0.51 <sup>ax</sup>	1.21±0.56 <sup>ax</sup>
Dressing %	0.0	72.50±1.15 <sup>ax</sup>	72.45±0.54 <sup>ax</sup>	73.74±1.29 <sup>ax</sup>
	25.0	72.66±0.93 <sup>ax</sup>	73.11±3.16 <sup>ax</sup>	70.56±0.58 <sup>ax</sup>
	50.0	72.94±0.45 <sup>ax</sup>	72.75±0.95 <sup>ax</sup>	72.81±0.49 <sup>ax</sup>
Liver weight	0.0	43.70±7.83 <sup>ax</sup>	45.60±4.88 <sup>ax</sup>	46.40±7.77 <sup>ax</sup>
	25.0	46.15±8.83 <sup>ax</sup>	41.63±6.67 <sup>ax</sup>	45.83±5.04 <sup>ax</sup>
	50.0	41.45±8.43 <sup>ax</sup>	38.33±5.78 <sup>ax</sup>	44.40±6.13 <sup>ax</sup>
Liver relative weight	0.0	2.38±0.50 <sup>ax</sup>	2.41±0.12 <sup>ax</sup>	2.59±0.36 <sup>ax</sup>
	25.0	2.52±0.31 <sup>ax</sup>	2.11±0.43 <sup>ax</sup>	2.34±0.22 <sup>ax</sup>
	50.0	2.97±0.36 <sup>ax</sup>	2.28±0.25 <sup>ax</sup>	2.14±0.36 <sup>ax</sup>

Values are means ± standard error. Mean values with different letters at the same column (a - c letters) or row (x - z letters) and period differ significantly at (P≤0.05).

It was observed that enzyme supplementation in both corn-soybean based diet or wheat grain containing diet improved ( $P \geq 0.05$ ) thymus gland weight and relative weight when compared with broiler chicken group fed on the same diet without enzyme supplementation. Moreover, wheat inclusion at 25% instead of corn increased ( $P \geq 0.05$ ) thymus weight and relative weight by about 33.0% and 33.3% respectively, while higher inclusion level of wheat grain improved ( $P \geq 0.05$ ) both thymus gland weight and relative weight when compared with broiler chicken group fed on corn-soybean based diet. On the other hand both enzyme supplementation and wheat inclusion instead of corn in broiler chicken diet had no significant effect on spleen weight and relative weight. The present data indicated that enzyme supplementation may slightly improve immune response of broiler chicken. The obtained data are in harmony with Soltan (2009) who indicated that enzyme supplementation to broiler chicken diet with different levels of palm kernel cake (PCK) had no effect on spleen, bursa and thymus gland relative weights when compared with chick group fed on the same diet without enzyme supplementation. Moreover, it was observed that enzyme supplementation had no significant effect on liver weight and dressing percentage of broiler chicken. It was observed that wheat inclusion instead of corn in broiler chicken diet non significantly reduced abdominal fat weight and relative weight when compared with broiler chicken group fed on corn – soybean based diet, moreover, enzyme supplementation in both corn – soybean based diet or with wheat grain non significantly reduced abdominal fat when compared with broiler chicken group fed on the same diet without enzyme supplementation.

Conclusion: From the result of the present study, it can be concluded that wheat grain can be used in broiler chicken ration instead of corn up to 50% without determinately effect of growth performance and liver or kidney functions. Moreover, enzyme supplementation in wheat containing diet improve broiler chicken performance, immune response and

economical efficiency of broiler chicken production.

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