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PROTEIN SOURCES AND/OR ENZYMES EFFECTS ON BROILER PERFORMANCE AND PHYSIOLOGICAL STATUS

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SUMMARY

The aim of this study was to investigate the synergism of protein sources (plant or animal) and enzymes in diet on broiler performance, gastric enzymes activity, some blood parameters and histological observations. A total number of 270 one day unsexed Hubbard chicks were distributed into 6 equal groups. Each group has 3 replicates of 15 birds each. The first group served with plant source protein, the second one served with animal source protein, the rest 4 groups were supplemented with 100 or 200mg commercial enzymes per kg diet (plant or animal source of protein). Our results indicated that the benefits of the use of 100mg enzyme per Kg plant source of protein in diets on broiler performance, economic efficiency and increase in gastric enzymes activities (when comparing with the rest of groups) which explain the improvements in the FCR. The current trial indicated also, that broiler diets supplemented with enzymes increased significantly the total proteins and globulin levels and reducing the cholesterol level in serum, which might supported by the enhancement of some immune organs (spleen and bursa).

Keywords: *enzymes, source of protein, broiler performance, gastric enzyme activity, blood components.*

INTRODUCTION

The attention of protein source and/or enzymes to broiler diets has gained increasing focusing because of both environmental and economic aspects. Broiler industry has been focused to obtain faster broiler growth in minimal time (Longo et al., 2007). In order to achieve this improvement, broilers should be fed diets that meet all their high-quality protein requirements. In addition, enzyme prospect stimulate a better utilization of the diet, because less feed is needed to produce a certain amount of meat and fewer nutrients end up in the litter (Kalmendal and Tauson, 2012). When using the animal protein sources they must be good manufactured product for devoid of disease but this source it is balance in amino acids vs. the plant protein sources. So when added enzymes on plant protein source developed digestive efficiency of protein increased in percentage of amino acids for bird. An animal by-product can be simply defined as a part of a slaughtered animal which is not directly contributing to human nutrition (Hazarika, 1994). Protein supplements of animal origin are obtained from rendering operations, meat packing, poultry and poultry processing, milk and dairy processing, and fish and fish processing (Denton et al., 2005). Although, these by-products are characterized by their high content of good quality protein and energy, but the most protein ingredients supplied to broiler diets are obtained from vegetable proteins, especially soybean meal and corn gluten meal. Vegetable (plant) and animal products are the two most important protein sources in poultry diets. Therefore during broiler diet formulation, choosing ingredients to maximize nutrient availability, rather than simply meeting energy or amino acid levels, is necessary (Ravindran, 2005; Sleman et al., 2015).

Recently, the efficacy of many commercial enzyme products has been well stated, but there is still some vagueness in their mode of action (Bedford, 2002). It is well known that exogenous enzymes have been used to improve the feeding values of feedstuffs are high in soluble non- starch polysaccharides that induce viscosity (Mathlouthi et al., 2002; Lazaro et al., 2003). Moreover, it has been reported that enzyme cocktail (carbohydrase and protease) enhance the productivity (saleh et al., 2005) and digestibility of corn - soybean

meal (Graacia et al., 2003; Olukosi et al., 2007; Cowieson and Ravindran, 2008). In addition, Gao et al. (2007) suggested that enzyme supplementation accelerated the development of the immune organs. In this respect, the author hypothesized that the improvement of the nutrient digestibility might be reflected in enhancing immunity and modifies blood metabolites profile. Therefore, the aim of this study was to investigate the synergism of protein sources (plant or animal) and enzymes in diet on broiler performance, gastric enzymes activity, some blood parameters and histological observations.

MATERIALS AND METHODS

This study was conducted at poultry experimental units and laboratories, faculty of agriculture, Ain Shams University, Egypt. A total number of 270 one day-old Hubbard broiler chicks were randomly divided into 6 equal groups, each was subdivided into 3 replicates with (15 chicks/each). All chicks were vaccinated against the common viral diseases (NDV, IBDV) at the recommended periods. Two feeding phases were applied: starter, from 1-21 days and finisher from 22-35days of age. Experimental diets were formulated to meet the nutrient requirements of the broiler chicks (NRC, 1994) which are presented in Table (1). The first group was served as control and fed basal diets with plant source of protein. While, the second group fed animal protein source, while, the other four groups received the basal diet (plant or animal source of protein) supplemented with commercial enzymatic product (PhytaBex PLUS) 100 or 200mg per Kg diet. Each 1 Kg of this product contains: Xylanase 10000000 IU, Cellulase 500000 IU, Acid Protease 2000000IU, α-Amylase 100000IU, B-Glucanase 500000IU, B-Mannanase 800000IU and phytase 5500000FTU.

Table (1): Feed ingredients and chemical composition of basal diets:

Ingredient	Dietary Treatments Plant Protein		Dietary Treatments Animal Protein	
	Starter	Grower	Starter	Grower
Yellow Corn	54.50	57.50	55.00	58.00
Soybean Meal (44%)	33.00	28.00	33.00	28.00
Corn Gluten Meal (62%)	6.20	6.20	-	-
Poultry By-Product Meal (58%)	-	-	6.60	6.60
Soybean Oil	2.00	4.00	2.00	4.00
Di-Calcium Phosphate	2.10	2.10	1.50	1.50
Calcium Carbonate	1.30	1.30	1.00	1.00
Premix	0.30	0.30	0.30	0.30
NaCl	0.20	0.20	0.20	0.20
Methionine HA	0.20	0.20	0.20	0.20
L-Lysine HCl	0.20	0.20	0.20	0.20
Total	100	100	100	100
	Chemical Analysis			
Crude Protein %	23.00	21.05	23.16	21.21
ME Kcal/ Kg diet	2986	3168	2966	3149
Ca%	1.08	1.06	1.01	1.00
AP%	0.51	0.50	0.50	0.50
Lysine	1.29	1.16	1.43	1.31
Methionine &Cystein	0.95	0.90	0.92	0.87
Price/ Ton (L.E.)	3842	3823	4017	3998

Methionine HA: Methionine Hydroxy-Analogue, ME: metabolizable energy, AP: Available phosphorus.
Each 3 Kg of the premix contains: Vitamins: A: 12000000 IU; Vit. D3 2000000 IU; E: 10000 mg; K3: 2000 mg; B1:1000 mg; B2: 5000 mg; B6:1500 mg; B12: 10 mg; Biotin: 50 mg; Coline chloride: 250000 mg; Pantothenic acid: 10000 mg; Nicotinic acid: 30000 mg; Folic acid: 1000 mg; Minerals: Mn: 60000 mg; Zn: 50000 mg; Fe: 30000 mg; Cu: 10000 mg; I: 1000 mg; Se: 100 mg and Co: 100 mg.

Feed and water were supplied ad-libitium and a constant (22L: 2D) light period was provided during the experimental periods. All chicks were kept under the same managerial, hygienic and environmental conditions. Chicks were individually weight at the beginning of the experiment, then at weekly intervals until the end of experiment, live body weight (LBW), body weight gain (BWG), feed consumption (FC), feed conversion ratio (FCR, g feed/g gain) were recorded during these periods. At the end of the experiment, 10 birds from each experimental group were weighted and slaughtered. Carcasses were manually eviscerated and weighted. Liver, heart, gizzard, spleen, thymus (all lobes of both sides), bursa and abdominal fat were removed and their relative percentage of live body weight was estimated. Representative specimens of liver, and small intestine (jujenium) for different groups were fixed in 10 % formalin-saline solution and prepared by the ordinary histological techniques. The sections were stained with haemotoxylline and eosin (H& E) stains according to the methods of Culling (1983). These sections were examined under X40 power using light microscope and photographed by using a suitable digital Camera. Concerning the digestive tract, the stomach and intestine were emptied by gentle squeezing, contents of individual segments were taken and mixed and about 1g of the mixed content was immediately diluted with 10 ml of distilled water. All samples were centrifuged for 10 minutes. The supernatant fluid was taken and stored in sealed bottles at -20°C until analyzed for enzymatic activity. Protease enzyme activity determination method in digestive content of stomach, and intestine of chicks was as described by Malik and Singh (1982).

Blood samples were collected from the ten slaughtered birds in non-heparinized tubes and were centrifuged at 3000rpm for 15 min. and serum obtained was stored at -20C until analysis. Serum total proteins, albumin, total cholesterol, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were determined calorimetrically. The globulin values were calculated by subtracting the values of albumin from the corresponding values of total proteins. The economic efficiency was calculated according to the price of local market at the time of carrying out the experiment as follows: Economic efficiency = (A-B/B) × 100.

Where: A = Price of kg gain in Egyptian pounds B = Feed cost / kg gain in Egyptian pounds.

Performance index (PI) was calculated according to North (1981).

The production efficiency factor (PEF) was calculated according to Emmert (2000).

Data were analyzed using two-way analysis of variance with source of protein and enzymes and their interaction using the General Linear Model (GLM) procedure of SAS (2002). When significant differences among means were found, means were separated using Duncan's multiple range tests (1955).

RESULTS AND DISCUSSION

Performance traits:

The effects of dietary treatments on BW, FCR, and mortality are shown in Tables (2a,b). The source of plant protein regardless enzyme additive significantly increased LBW and DWG. When added enzyme on source of plant protein we found increased in LBW. The level of 100mg of enzyme with plant source of protein in the diets of broiler affect significantly positively in the BW and the livability of the broilers at the growing period than those fed diets with or without supplementations.

The results showed significantly ($P \leq 0.05$) higher body weight at 21 day of age for the group supplemented with 100 mg of enzyme with plant source of protein than other groups and the same trend was recorded at 35 day of age. The best LBW ($P \leq 0.01$) was recorded for 100 mg of enzyme with plant source of protein group than anther groups. Concerning daily weight gain, the high daily weight gain for chicken fed the plant source of protein regardless enzyme and increased when added enzyme in the group fed 100 mg of enzyme with plant source of protein showed higher daily weight gain (Tables 2a,b). The highest values were recorded for 100 mg of enzyme with plant source of protein group which were equally

similar at 21 and at 35 day of age. The best FCR for birds feed diet supplemented with 100 mg of enzyme with plant source of protein which were equally similar at 21 and 35day of age.

Table (2a): Effect of feeding different levels of enzymes and sources of protein on productive performance of growing chicks:

Item	Trait			
	LBW 21 d	DWG	DFI	FCR
Source of protein				
Animal	659.444^{b}	29.32^{b}	52.462^{a}	1.789^{a}
Plant	716.968^{a}	32.054^{a}	47.957^{b}	1.496^{b}
Treatment (T)				
0	676.917^{b}	30.150^{b}	48.437^{b}	1.608^{b}
100 mg	701.667^{a}	31.325^{a}	51.272^{a}	1.678^{a}
200 mg	686.035^{ab}	30.583^{ab}	50.920^{a}	1.628^{b}
Interaction effect				
Animal -0	663.00^{d}±1.35	29.49^{c}±0.064	50.40±0.055	1.71^{b}±0.040
Animal -100 mg	663.33^{d}±0.96	29.50^{c}±0.046	53.90±0.915	1.83^{a}±0.003
Animal- 200 mg	652.00^{e}±0.58	28.96^{d}±0.026	53.09±0.230	1.83^{a}±0.020
Plant – 0	690.83^{c}±1.83	30.81^{b}±0.087	46.47±1.110	1.51^{c}±0.008
Plant -100 mg	740.00^{a}±3.94	33.15^{a}±1.045	47.94±0.015	1.45^{d}±0.023
Plant –200 mg	720.07^{b}±0.92	32.20^{a}±0.043	49.46±0.592	1.54^{c}±0.006
Probability				
P	0.0001	0.0001	0.0001	0.0499
E	0.0411	0.0419	0.0016	0.0175
P*E	0.0421	0.0417	NS	0.0407

$^{a, b \text{ and } c}$ *Means within the same main effects with different letters are significantly differed, NS= Non-significant.*

Table (2b): Effect of feeding different levels of enzymes and sources of protein on productive performance of growing chicks at 35 day of age

Item	Trait			
	LBW 35 d	DWG	DFI	FCR
Source of protein				
Animal	1358.94^{b}	49.964^{b}	118.357	2.369^{a}
Plant	1455.04^{a}	52.720^{a}	116.373	2.207^{b}
Treatment (T)				
0	1367.61^{b}	49.398^{b}	115.847	2.347^{a}
100 mg	1454.37^{a}	53.777^{a}	115.609	2.158^{b}
200 mg	1399.01^{ab}	50.852^{ab}	120.640	2.385^{a}
Interaction effect				
Animal -0	1384.71±21.311	51.623±0.363	118.943+3.225	2.304±0.055
Animal 100 mg	1385.11±4.105	51.580±0.176	120.667±5.903	2.339+0.058
Animal- 200 mg	1307.00±12.191	46.690±0.728	115.460±0.341	2.473+0.136
Plant – 0	1350.50±12.413	47.173±0.823	112.750+1.963	2.390+0.017
Plant -100 mg	1523.63+13.063	55.973+0.632	110.550±2.696	1.975+0.075
Plant –200 mg	1491.00+8.083	55.013±0.447	125.820±2.731	2.287+0.017
Probability				
P	0.0248	0.0380	NS	0.0410
E	0.0460	0.0447	NS	0.0492
P*E	NS	NS	NS	NS

$^{a, b \text{ and } c}$ *Means within the same main effects with different letters are significantly differed, NS= Non-significant.*

Our results concerning the performance of broilers were in agreement with Cowieson et al. (2006). They indicated that exogenous protease and phytase (Avizyme) can be used successfully in a strategically formulated low nutrient density diet to maintain performance to that of birds fed on a nutritionally adequate diet. In addition, Cowieson and Ravindran (2008) stated that supplementing corn-SBM-based broiler diets with an enzyme product containing protease improved BWG and feed efficiency compared with the un-supplemented diets, but feed intake did not affected. The mode of action of enzymes in corn-SBM-based diets has been linked to improved starch digestibility associated with augmentation of endogenous alpha-amylase or improved digestion of resistant starches, improved access to cell contents via a reduction in cell wall integrity, modification of the intestinal microbial communities, improved protein solubility and digestibility and a reduction in the inimical effects of maize and/or soy-derived anti-nutrient factors. In the same context, Saleh et al, (2005) reported that the commercial enzymes, which are mostly comprised of carbohydrases and contain small amount of protease activity (Energex) improved significantly the productivity (BWG and FCR) of broilers fed corn-SBM-based diets compared to pure carbohydrases (cellulose, hemicellulose and pectinase) supplementation, which tended to affect compared to control group (without enzyme supplementation). However, they noted that feed intakes were not affected by dietary enzymes. Similar results have been found earlier by Zanella et al. (1999) when they supplemented a corn-SBM diet with Avizyme, a commercial enzyme; BWG and FCR were significantly improved by Avizyme. They demonstrated that the energy and amino acid digestibility of a corn-SBM-based diet for broiler could be improved by around 3% when supplemented with xylanase, amylase and protease allowing performance to be maintained on a diet with a lower nutritional plane. In addition, Kalmendal and Tauson (2012) observed that the combination of xylanase and serine protease improved FCR, compared with the control diet but, BW and FI were not affected by enzyme addition sole or mixed. Moreover, Gracia et al, (2003) demonstrated that amylase was a critical enzyme to improve the nutritional value of corn-based broiler diets, improved BWG and FCR by 4 to 9% compared with an un-supplemented control diet. Moreover, Onilude and Oso (1999a) reported that the supplementation of three enzyme mixtures (amylase, cellulase and pectinase) to broiler fiber-containing diets improved live BW, BWG and FCR at 42 d of age. Whereas, Sarica et al. (2005) reported that xylanase supplementation in broiler diets based on wheat-corn-SBM up to 42 d did not affected BWG, feed intake or FCR for control and treated birds.

On the other hand, Meng et al. (2006) stated that 0.05% enzyme (contained cellulase, pectinase, mannanase, xylanase and glucanase as main activities) supplementation to broiler diets based on corn-SBM improved FCR. However, no effects were observed by enzyme supplementation for feed intakes for control vs. supplemented enzyme group, respectively. In addition, Walk et al. (2011) used mono-component xylanase and protease products, they found no positive effects on production performance in broiler chickens fed a corn-SBM-based diet. Also, Barekatain et al. (2013) observed that an admixture of protease to broiler corn-SBM based diets did not result in further improvement in productive performance represented by BWG, feed intake and FCR. If the enzymes were additive in their effect, it would be expected that the sum of the effect attributed to each enzyme individually should not be different from the effect attributed to the use of the enzymes in combination (Olukosi et al., 2007).

Digestive enzymatic activity:

Digestive enzymes activities (protease) in different segments of gastrointestinal tract are presented in Table (3). As a result of stomach pH, among groups,protease activity was significantly different among groups when estimated in stomach and in small intestine. In stomach, protease activity was more than 2 folds in plant source of protein than other groups regardless of enzyme. Protease activity was more than 4 folds in 100 mg of enzyme with plant source of protein than other groups. Whereas, the same trend was found for its activity in small intestine,these data may explain our results for the superiority of supplemented broilers with 100 mg of enzyme with plant source of protein in live body weights and blood profile. These findings may reflect a good feed utilization, absorption and metabolism for birds when diets supplemented with plant source of protein.

Table (3): Effect of different levels of enzymes and sources of protein on protease enzyme activity of broiler

Item	Trait	
	In stomach content:- Protease	In small intestine content:- Protease
Source of protein		
Animal	22.30[b]	14.66[b]
Plant	49.70[a]	23.76[a]
Treatment (T)		
0	27.26[c]	17.25[b]
100 mg	46.14[a]	23.10[a]
200 mg	34.61[b]	17.28[b]
Interaction effect		
Animal -0	27.01[c]±3.14	16.25[b]±2.23
Animal -100 mg	26.33[c]±5.65	17.45[b]±2.75
Animal- 200 mg	13.57[d]±3.75	10.27[c]±3.15
Plant – 0	27.50[c]±3.63	18.25[b]±3.76
Plant -100 mg	65.95[a]±4.89	28.75[a]±2.01
Plant –200 mg	55.65[b]±2.35	24.28[a]±3.24
Probability		
P	0.0249	0.0080
E	0.0465	0.0128
P*E	0.0270	0.0474

[a, b and c] *Means within the same main effects with different letters are significantly differed, NS= Non-significant.*

Blood parameters:

Data presented in Table (4) clarify the effect of feeding broilers on diets with different levels of enzyme with source of proteins on serum protein profile. Significant increases were recorded in total proteins and globulin in 100mg enzyme with plant source of protein group. Whereas, no significant differences in albumin were observed compared with another groups. These findings in turn have influenced the A/G ratio as it declined from 0.52 in the control to the range of 0.22 to 0.29 in the other treatments. The reduction in A/G ratio may reflect an enhancement of broilers immunity.

The values of serum constituents in broilers at 35 days of age (Table 4) were within the normal ranges for serum total cholesterol, total protein and albumin (Meluzzi et al., 1992; Del Bianchi et al., 2005).

Regarding to liver function expressed as serum AST and ALT, the results clearly indicate non-significant variations between treatments. The histological sections on liver confirmed these findings. Serum kidney enzymes activities values (AST and ALT) at 35 d of age of broilers in the current trial are within the normal range (Viveros et al., 2002). Enzyme supplementation of chicken diets is employed in order to increase the availability of starch, protein and other macronutrients that are entrapped by intact cell wall structures or viscous polymers that are resistant to digestion by endogenous host enzymes (Frigard et al., 1994). The current trial indicated also, that broiler diets supplemented with enzymes increased significantly the proteins and globulin levels in serum, which might supported by the enhancement of immune organs (spleen and bursa). It is well stated that gama-globulin is the main component of anti-body production, which presents the humoral immune response. So, findings of globulin levels in serum in the current study are supported by Gao et al. (2007), who suggested that xylanase supplementation, to wheat-based diets for cockerels from 7 to 21 d of age enhanced the humoral immune response.

The current experiment showed a favor effect of enzyme addition to broiler diets up to 35 d of age in reducing the cholesterol level in serum, suggesting that enzyme supplementation might play a role in broiler lipid metabolism. Unfortunately, little information has been published on the effects of enzyme supplementation in broiler diets on blood lipid metabolites. However, Onilude and Oso (1999b) reported that the supplementation of enzyme mixture including amylase, cellulase and pectinase to broiler fiber-containing diets from hatch to 42 d of age reduced blood lipid metabolites including plasma cholesterol

level from 246 to 136 mg/dL at 42 d of age. Also, Cowieson et al. (2013) reported that phytase addition to broiler diets reduced total cholesterol concentration in the blood of chickens fed the positive control diet (adequate in P and Ca) but, increased cholesterol concentrations in the blood of chickens fed the negative control diet (with P and Ca levels reduced by 0.12 and 0.14%, respectively) however, no effects of phytase on total- and HDL-cholesterol were noted.

Table (4): Effect of different levels of enzymes and sources of protein on blood parameters of broiler

Item	Trait					
	T. protein g/dl	Albumin g/dl	Globulin g/dl	Cholesterol mg/dl	AST IU/L	ALT U/L
Source of protein						
Animal	6.8467	4.2344	2.6122	176.17	49.268[a]	45.403
Plant	6.2311	4.0578	2.1744	198.33	29.188[b]	47.122
Treatment (T)						
0	6.2167	4.2517	1.9650[b]	191.00	40.490	46.350
100mg	6.7433	4.0333	2.7117[a]	173.00	41.447	42.608
200mg	6.6567	4.1533	2.5033[ab]	197.75	35.747	49.830
Interaction effect						
Animal-0	6.587±0.292	4.197±0.332	2.390±0.040	185.50±6.640	49.830±9.660	46.680±0.416
Animal-100mg	7.347±0.459	4.400±0.300	2.947±0.159	145.00±12.124	58.657±6.411	42.370±9.694
Animal- 200mg	6.607±0.193	4.107±0.003	2.500±0.196	198.00±2.887	39.317±3.617	47.160±0.208
Plant – 0	5.847±0.338	4.307±0.153	1.540±0.185	196.500±8.949	31.150±0.260	46.020±0.727
Plant-100mg	6.140±0.641	3.667±0.061	2.477±0.580	201.000±37.528	24.237±4.050	42.847±9.339
Plant –200mg	6.707±0.136	4.200±0.064	2.507±0.072	197.500±22.228	32.177±0.476	52.500±3.429
Probability						
P	NS	NS	NS	NS	0.0005	NS
E	NS	NS	NS	NS	NS	NS
P*E	NS	NS	NS	NS	NS	NS

[a, b and c] *Means within the same main effects with different letters are significantly differed, NS= Non-significant.*

Carcass traits:

The main results of carcass traits are set out in Table (5). Carcass, breast and thigh weights (as a percentage of carcass) were significantly ($P \leq 0.01$) greater in birds fed supplement of enzyme than another group. No effect of treatment on % gizzard, liver, heart, spleen, bursa, abdominal fat and dram. Dressing of broilers in the current trial represented by carcass relative weight was increased in response to dietary plant source of protein and increased when added enzymes, so the best value of dressing percentage for chicken fed 200 mg enzyme with plant source of protein.

Onilude and Oso (1999a) reported that the supplementation of three enzyme mixture (amylase, cellulase and pectinase) to broiler fiber-containing diets increased carcass weight. Moreover, Café et al. (2002) noted a significant increase in dressing percentage in broilers given a corn-SBM diet supplemented with commercial enzymes. These also, are in agreement with Saleh et al. (2005) who reported that carcass relative weight was higher for broilers fed pure carbohydrases (cellulase, hemicellulose and pectinase) than control group with broilers fed a commercial enzymes. They attributed the improvement of carcass yield to the effects on crude protein metabolize ability. An increase in carcass is a typical response to increased protein: ME ratio (Donaldson, 1985). In the current experiment, internal organs were not affected by the enzyme addition. These results are in agreement with Saleh et al. (2005), who stated no differences in liver relative weight in response to dietary mixed enzymes. Also, Gao et al. (2007) observed that xylanase supplementation in broiler diets based on wheat-corn-SBM did not affect gizzard relative weight. In addition, Sarica et al. (2005) reported that xylanase supplementation in broiler diets based on wheat-corn-

SBM did not affect heart, liver or gizzard relative weights. Similar results were reported by Gracia et al. (2003) and Barekatain et al. (2013).

Table (5): Effect of different levels of enzymes and sources of protein on carcass traits

Item	Trait					
	LBW	Hot carcass	Dressing percentage	Abdominal fat	Liver	Gizzard
Source of protein						
Animal	1474.78[b]	1041.67[b]	70.66	1.14[b]	2.32	1.44[a]
Plant	1590.00[a]	1127.44[a]	70.91	1.71[a]	2.34	1.16[b]
Treatment (T)						
0	1524.17	1074.33	69.95[b]	1.33	2.27	1.52[a]
100mg	1563.83	1094.50	70.48[ab]	1.33	2.29	1.22[b]
200mg	1509.17	1083.83	71.93[a]	1.63	2.44	1.17[b]
Interaction effect						
Animal-0	1486.67±20.48	1051.33±12.98	69.24±0.567	0.88±0.262	2.14±0.111	1.74±0.132
Animal-100mg	1499.33±14.45	1038.00±5.77	70.73±0.629	1.14±0.172	2.34±0.139	1.31±0.107
Animal-200mg	1438.33±37.23	1035.67±20.58	72.01±0.897	1.41±0.382	2.49±0.075	1.27±0.076
Plant – 0	1561.67±49.10	1097.33±36.78	70.66±0.822	1.77±0.341	2.39±0.114	1.29±0.049
Plant-100mg	1628.33±34.92	1151.00±32.08	70.22±0.270	1.51±0.196	2.25±0.115	1.12±0.045
Plant – 200mg	1580.00±71.47	1134.00±40.10	71.84±0.803	1.85±0.134	2.39±0.209	1.06±0.059
Probability						
P	0.0060	0.0026	NS	0.0220	NS	0.0013
E	NS	NS	0.0388	NS	NS	0.0027
P*E	NS	NS	NS	NS	NS	NS

[a, b and c] *Means within the same main effects with different letters are significantly differed, NS= Non-significant.*

Table (6): Effect of different levels of enzymes and sources of protein on giblets and some carcass characteristics

Item	Trait				
	Heart	Spleen	Thymus	Bursa	Giblets
Source of protein					
Animal	0.46	0.107	0.223	0.108	4.222
Plant	0.52	0.148	0.240	0.102	4.018
Treatment (T)					
0	0.490	0.125	0.212	0.125	4.272
100mg	0.483	0.113	0.255	0.080	3.993
200mg	0.485	0.143	0.228	0.110	4.095
Interaction effect					
Animal-0	0.433±0.047	0.123±0.012	0.237±0.048	0.130±0.058	4.320±0.218
Animal-100mg	0.443±0.037	0.103±0.020	0.277±0.029	0.070±0.001	4.093±0.112
Animal- 200mg	0.490±0.015	0.093±0.023	0.157±0.052	0.123±0.032	4.253±0.012
Plant – 0	0.547±0.049	0.127±0.018	0.187±0.069	0.120±0.031	4.223±0.151
Plant-100mg	0.523±0.030	0.123±0.038	0.233±0.069	0.090±0.017	3.893±0.170
Plant – 200mg	0.480±0.065	0.193±0.068	0.300±0.087	0.097±0.032	3.937±0.306
Probability					
P	NS	NS	NS	NS	NS
E	NS	NS	NS	NS	NS
P*E	NS	NS	NS	NS	NS

[a, b c] *Means within the same row with different letters are significantly differ.*

The present study showed that 100mg enzyme with plant source of protein to corn-based diets significantly increased the relative weight of the spleen and bursa suggesting that enzyme supplement accelerated the development of the immune organ. However, Gao et al. (2007) observed that xylanase supplementation to wheat-based diets for cockerels from 7 to 21 d of age significantly increased the relative weight of the spleen. They attributed that to the improvement of feed digestion, the enhancement of nutrients absorption and the regulation of metabolic hormones in response to the addition of the enzyme, which in turn could have an effect on body immunity.

Table (7): Effect of different levels of enzymes and sources of protein on Economic Efficiency.

Item	Trait							
	Average Feed consumed	Total Feed cost	Final Wight Gain	Total Return	Net Return	EE	PI	PEF
Source of protein								
Animal	2.759	10.0814	1.311	18.353	8.272	82.048	62.306	207.888
Plant	2.636	9.7110	1.440	20.151	10.440	107.509	78.654	267.032
Treatment (T)								
0	2.635	9.615	1.344	18.811	9.195	95.636	68.482	228.901
100mg	2.686	9.582	1.430	20.024	10.172	103.246	75.598	256.655
200mg	2.767	10.205	1.351	18.921	8.716	85.413	65.766	220.974
Interaction effect								
Animal-0	2.724	9.894	1.336	18.714	8.821	89.154	65.525	217.872
Animal-100mg	2.821	10.311	1.337	18.718	8.407	81.539	63.365	212.906
Animal- 200mg	2.731	10.031	1.259	17.626	7.595	75.714	58.044	192.998
Plant – 0	2.547	9.337	1.351	18.907	9.570	102.490	71.721	240.983
Plant-100mg	2. 554	9.339	1.524	21.331	11.932	126.946	90.854	311.629
Plant – 200mg	2.800	10.368	1.444	20.216	9.848	94.976	74.471	252.829

The highest values of Economic Efficiency, performance index (PI) and Production Efficiency Factor (PEF) were recorded for chicken given100 mg of enzymes with plant protein source. However, chickens fed animal protein source with 200 mg enzymes reflected the lowest figures compared with other treatments.

Histological results:

Histological examinations of the intestinal sections are illustrated in Figure (1). It is clear from the transverse sections (T.S) that the villi height were well developed in all birds even they were crowded in sections taken from groups served plant source of protein supplemented with both 100 and 200mg enzymes. Furthermore, there were great variations in the size and number of Crypts of Lieberkuhn associated with the supplemental additives. These Crypts are known to secrete fluids containing different vital substances essential for the internal micro-environment of the small intestine segments (Hodges, 1974). While the sections from the liver parenchyma of the control treatment Fig. (2) has normal hepatocytic structure with dilated central vein engorged with blood. Also, there were dark stained eosinophilic cells surrounding or near the central veins. There is moderate hypertrophy of liver cells especially in (groups fed animal source of proteins) which may reveal hyperactivity of the liver cells or a compensatory effect due to more degenerative (necrotic) areas in these sections. The above mentioned changes in liver sections may be related to the higher metabolic activity associated with the higher growth rates of broilers which depends on their genetic background.

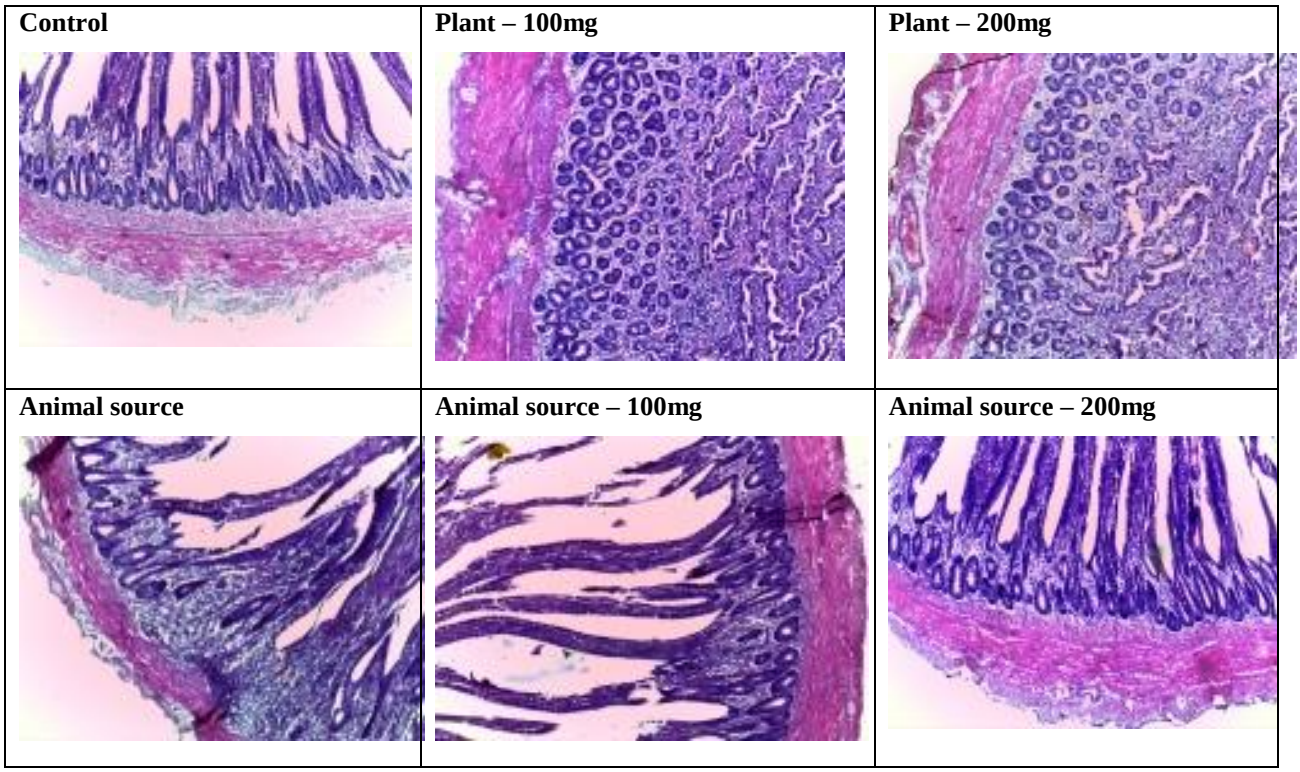

Fig. (1): The histological structure (at 40X) of the small intestine from broilers fed different biological additives and a control group.

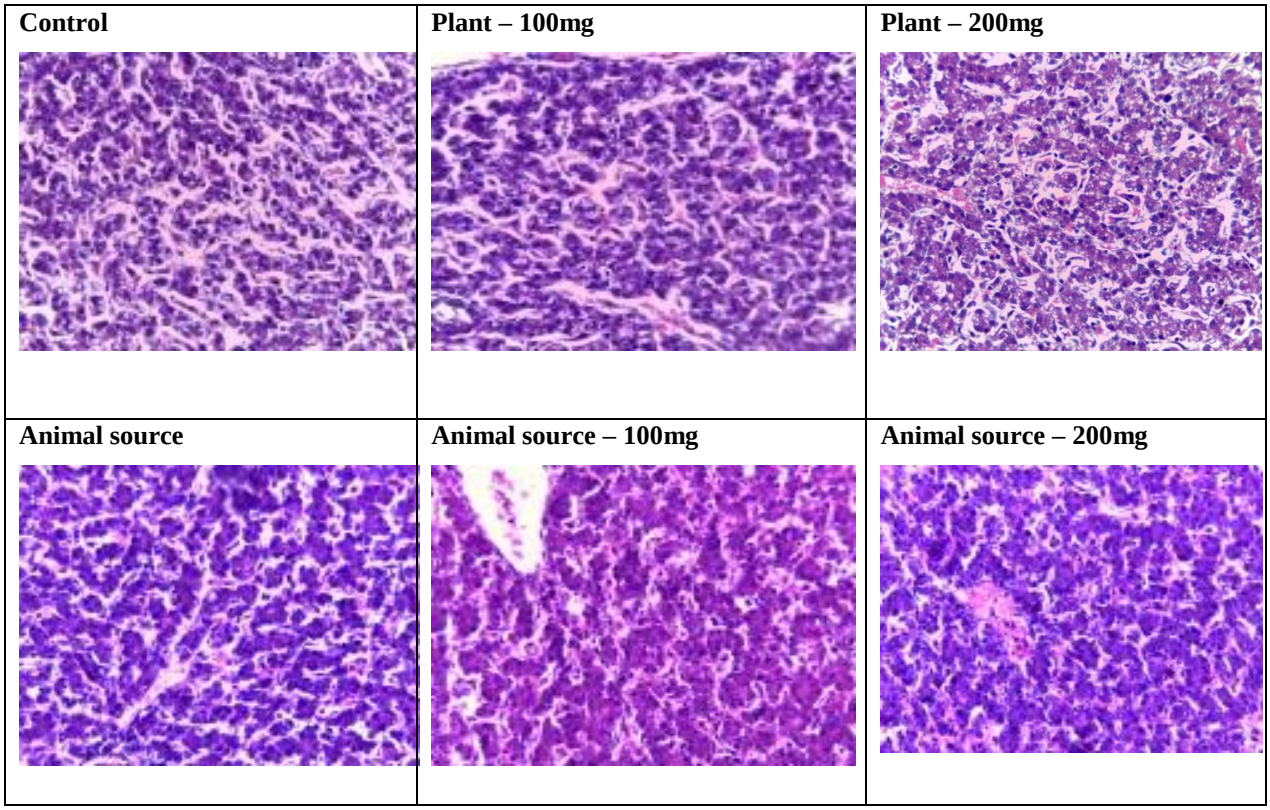

Fig. (2): The histological structure (at 40X) of the liver from broilers fed different biological additives and a control group.

CONCLUSIONS

It could be concluded that broiler diets supplement with 100 mg of enzyme with only plant source of protein improved broiler might improve broiler immunity by accelerating the improvement of immune organs and increasing the total proteins and globulins levels in serum. Also, dietary 100 mg of enzymes with only plant source of protein has a favor effect on enzymes activity by the increase in stomach and intestine enzymes in 100 mg of enzyme with plant source of protein groups are good explanations for the improvements in the FCR. The histological and blood pictures showed that enzyme products are very safe on birds and accordingly birds are safe for human consumption. Good of economic efficiency for 100 mg of enzyme with plant source of protein/ Kg diet. Therefore, more studied are required to explain the mode of action of the effect of enzyme supplementation on immunity and blood constituents in broilers.

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تأثير مصادر البروتين و/أو الانزيمات علي الحالة الانتاجيه و الفسيولوجية لدجاج التسمين

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تهدف هذه الدراسة الي دراسة تلازم مصادر البروتين (سواء النباتيه أو الحيوانية) مع الانزيمات بالعليقة علي الاداء الانتاجي , النشاط الانزيمي بالامعاء , بعض مكونات الدم و كذلك القطاعات الهستولوجية بدجاج التسمين. تم تقسيم 270 كتكوت (هابرد) تسمين عمر يوم واحد الي 6 مجموعات متساوية. تمت تغذية المجموعة الاولي علي عليقة تحتوي علي مصدر بروتيني من أصل نباتي, أما الثانية فتمت تغذيتها علي بروتين من أصل حيواني . أما باقي المجموعات تمت اضافة 100 أو 200ملجم/كجم انزيمات تجارية للعليقة (سواء كان بروتين العليقة من أصل نباتي أو حيواني).

وضحت نتائجنا علي أفضلية اضافه 100 ملجم/ كجم انزيمات علي العليقة المحتويه علي بروتين من أصل نباتي لافضل اداء انتاجي لدجاج التسمين , هذا بالاضافة الي أفضل تحويل غذائي و زيادة في النشاط الانزيمي بالامعاء مقارنة بالمجموعات الاخري بهذه الدراسة. وضحت هذه الدراسة أيضا علي أفضلية اضافة الانزيمات لعلائق دجاج التسمين التي حسنت معنويا مستويات البروتينات الكليه و الجلوبيولين و كذلك خفضت مستوي الكولستيرول بسيرم الدم علاوة علي بعض الاعضاء المناعية مثل الطحال و غدة البرسا.