

INFLUENCE OF DIETARY POLYZYME ON THE PERFORMANCE OF BROILERS FED LOW CRUDE PROTEIN

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ABSTRACT: *The present experiment was carried out in order to investigate the effect of polyzyme (multi enzymes) in broiler diets with low protein levels on growth performance, carcass traits, some blood components and economic efficiency. A total of 180 Ross 308 unsexed one - day old broiler chicks, were distributed at random into 3 groups each in 3 replicates, 20 chicks each. Treatments were: T₁: positive control, without Polyzyme, T₂: negative control, without Polyzyme and T₃: negative control, with 0.5 g Polyzyme/ kg diet through the starter (1- 21 d) and finisher (22- 35 d) period. Results revealed that, chicks fed the negative control with polyzyme addition had significantly ($P \leq 0.05$) higher body weight, body weight gain and performance index, while daily feed intake decreased. Feed conversion ratio, European efficiency index and economic efficiency improved also with the supplementation. Dressing, liver and heart % were increased in the same treatment (T₃), while gizzard % was not affected. Polyzyme supplementation to low protein diet decreased serum total cholesterol and increased aspartic transaminase (AST) enzyme compared to the positive control (T₁), meanwhile total protein, creatinine, glucose and alanine transaminase (ALT) enzyme were not affected, indicating the safety use of polyzyme at the level of 0.5 g/ kg diet. Data indicated that the beneficial economical effect of using polyzyme with low protein diet is to reach almost the same performance with normal protein diet.*

Key words: *Polyzyme, broilers, growth, carcass, serum components, diet.*

INTRODUCTION

Reducing the dietary CP level by 3% with amino acid supplementation during the grower period, followed by a control or low – energy diet, could improve overall protein utilization and reduce fat accumulation via increasing lipolysis and / or disruption of triglyceride transportation in broiler chickens. Understanding the mechanism and factors involved in this improvement requires focusing on gene expression related to lipids and energy metabolism (Jariyahatthakij *et al.*, 2016).

In the last decades, it has been done a lot of research in the chicken nutrition about using the exogenous enzymes, either in single or mixture enzyme products, on feed utilization and most of

them recorded beneficial effects of it as a very important tool for keeping the gut healthy, improving the utilization of nutrients, reduced environmental contamination and lowering feed cost, thus, allowing for more flexibility in diet formulation. This is reflected in better flock performance, better litter quality and improved bird health which in turn, has a positive influence on total production cost (Selim *et al.*, 2010 and Ferket, 2011) and many commercial enzyme products are currently available for the chicken nutrition (Adeola and Cowieson, 2011). Different kinds of interactions can occur between various supplemental enzymes. Carbohydrates may require the use of enzymes with divers activities that are able to target different sugar components of feed stuffs

used in poultry diets. When an enzyme cocktail containing several activities is used in a broiler diet, it's more likely to have greater effect than when enzymes are added separately (Odetallah *et al.*, 2003; and Gracia *et al.*, 2003).

Protein is very expensive component in broiler diets (Kamran *et al.*, 2004). Therefore, reducing crude protein (CP) without deleterious effects in broiler performance is a great challenge for broiler nutritionist. Not only reduced protein regimens diets in poultry nutrition are considered an alternative application to reduce feed costs, but also to reduce the environmental pollution (Kobayashi *et al.*, 2013). The industry also uses feed additives such as vitamin premixes, amino acids and enzymes which are largely procured from indigenous sources. Feedstuffs are consistently increasing in their cost and the trend that has exacerbated in recent years. Parallely increased public concern regarding the environment impact of animal and agriculture has increased the need to reduce nutrient in waste by food animals. Research on the use of exogenous enzymes is going on and many commercial enzymes are presently available to the poultry industry (Dongare *et al.*, 2017).

Despite of that, one applicable argument for using exogenous protease is using it in low protein diets. Enzyme supplementation should allow a reduction in CP level in feed whereas individually amino acid were not improved equally by supplementation and should be balanced (Zanella *et al.*, 1999). This study aimed to investigate the effect of polyzyme (multi enzymes) in broiler diets with low protein levels on growth performance, carcass traits, some blood components and economic efficiency.

MATERIALS AND METHODS

The present study was conducted at a private farm in Menoufia Governorate, Egypt, throughout the experimental period (October to November 2017). One hundred and eighty, one day old unsexed Ross broiler chicks were used in this study. Chicks were individually weighed, wing – banded, and randomly assigned to three treatment groups nearly similar in average body weight, (3 replicates pens of 20 birds each, ~ 40g). Polyzyme* was added at the levels of 0 and 0.5 g/ Kg diets to the low crud protein diet (20 and 17%) compared to the normal crud protein (23 and 20%), respectively at the starter (1 - 21 d) and at the finisher (22 – 35 d) period. Birds received their diets to save the nutrient according to the NRC (1994).

The compositions and chemical analysis of the experimental diets are shown in (Table 1). Feed and water were provided *ad libitum* during the experimental period (1 - 35 d of age). Artificial light was used to provide 24 hour photo period. The experimental groups were follows: Starter period, T₁: positive control 23% crude protein (CP), T₂: negative control, low CP diet (LCP) 20% without polyzyme, T₃: negative control, low CP diet (LCP) 20% with 0.5 g polyzyme/ Kg diet. The finisher period, T₁: positive control 20% CP, T₂: negative control, low CP diet (LCP) 17% without polyzyme, T₃: negative control, low CP diet (LCP) 17% with 0.5g polyzyme/ Kg diet. Body weight (BW), body weight gain (BWG) and feed intake (FI) was recorded weekly. Feed conversion ratio (FCR) was calculated during the experimental period. At the end of the experimental (5 weeks of age), 6 birds from each treatment around the average live body weight were chosen, fasted for

* Xylanase 2000000; Protease 375000; Lipase 25000; Pectanase 120000; Mannase 100000; Amylase 750000; Cellulase 200000; Phytase 25000; β Glucanase 200000 and Galctocidasse 200000U/ 0.5 kg Polyzyme.

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about 12 hours, weighed and slaughtered to complete bleeding, followed by plucking the feathers. Dressing and giblets weight were expressed related to live body weight were recorded. Blood samples were collected into tubes without heparin and separated by centrifugation at 3000 rpm for 15 minutes and frozen at -20 C° until analysis. Serum total lipids, cholesterol, creatinine and

glucose were determined using commercial kits. Also, liver enzymes including AST, ALT were calorimetrically estimated. The economic efficiency of the experimental diets used in the present study was calculated from the input – output analysis (Heady and Jensen, 1954), assuming that the other head costs were constant.

Table 1. Composition and chemical analysis of the experimental diets fed during starting (1 - 21) and finishing periods (22 - 35) days of age.

Ingredients	Starter diets		Finisher diets	
	Positive	Negative	Positive	Negative
Yellow corn, 8.5%	47.20	56.70	56.70	65.41
Soybean meal, 44%	41.30	34.50	34.50	26.15
Corn gluten, 60%	1.32	---	---	---
Vegetable oil	6.35	4.97	4.97	3.80
Limestone, ground	1.35	1.35	1.35	1.97
Di- Calcium phosphate	1.88	1.88	1.88	2.00
Vitamins and minerals mixture ¹	0.30	0.30	0.30	0.30
Salt (Sodium chloride)	0.30	0.30	0.30	0.30
DL- Methionine ²	----	----	----	0.07
Total	100	100	100	100
<u>Calculated analysis (air dry basis)³:</u>				
Crude protein, %	23.00	20.00	20.00	17.07
ME, k cal/ kg diet	3104	3100	3100	3104
C/ P ratio	135	155	155	182
Calcium, %	1.00	1.00	1.00	1.24
Available phosphorous, %	0.46	0.46	0.46	0.46

¹Vitamins and minerals mixture at 0.30 % of the diet supplies the following/ kg of the diet: Vit. A, 12000 IU; Vit. D₃, 2500 IU; Vit. E, 10 mg; Vit. K₃, 3 mg; Vit B₁, 1 mg; Vit. B₂, 4 mg; Pantothenic acid, 10 mg; Nicotinic acid, 20 mg; Folic acid, 1 mg; Biotin, 0.05 mg; Niacin, 40 mg; Vit.B₆, 3 mg; Vit B₁₂, 0.02mg; Choline chloride, 400 mg; Mn, 62 mg; Fe, 44 mg; Zn, 56 mg; I, 1 mg; Cu, 5 mg and Se, 0.01 mg.

²DL – Methionine: 98% feed grade (98 % Methionine).

³Calculated according to NRC (1994).

Statistical Analysis:

Data were statistically analyzed by the completely randomized design using SPSS (2011) program and the differences among means were determined using Duncan's multiple range test (Duncan, 1955). Percentages were transformed to the corresponding arcsine values before performing statistical analysis (Snedecor and Cochran, 1982).

The model applied was:
 $Y_{ij} = \mu + \alpha_i + E_{ij}$, Where:- Y_{ij} = an observation. μ = Overall mean. α_i = effect of treatment (I = 1, 2, 3, 4,.....7), and E_{ij} = Random error.

RESULTS AND DISCUSSIONS

Growth performance: Results of growth performance for Ross broiler chicks are shown in Tables (2 and 3). In the starter period, chicks fed the negative control diet, 20% without polyzyme had significantly lower ($P \leq 0.05$) BW (885g) and BWG (845g) compared to the positive control group (957g and 917g), respectively but nearly identical to the control group (T_1) in chicks fed the negative control diet + 0.5g polyzyme (948g and 909g).

Table 2. Effect of dietary polyzyme supplementation on the performance of growing broiler chicks at 3 and 5 weeks of age (Means \pm S.E).

Treatments ¹	Body weight (g)		Body weight gain (g)		Performance Index (PI, %) ⁴	
	3 weeks	5 weeks	3 weeks	5 weeks	3 weeks	5 weeks
T1	957 ^{a2,3} \pm 13.00	1951 ^b \pm 26.11	917 ^a \pm 13	1911 ^b \pm 26.41	72.56 ^b \pm 2.32	67.73 ^b \pm 4.12
T2	885 ^b \pm 7.93	1865 ^c \pm 16.91	845 ^b \pm 7.93	1825 ^b \pm 16.90	65.05 ^c \pm 1.12	60.97 ^c \pm 1.91
T3	949 ^a \pm 13.67	2092 ^a \pm 20.17	909 ^a \pm 1.12	2052 ^a \pm 20.52	81.42 ^a \pm 3.03	91.15 ^a \pm 2.25

¹T₁: Positive control, T₂: Negative control, T₃: Negative control + 0.5g polyzyme.

² means \pm S.E. of 3 replicates/ treatment.

³a,b,c.....etc: Means within the same column with different superscripts are significantly different ($P < 0.05$).

⁴ Performance Index (PI) = (live body weight (kg) \times 100 / feed conversion).

Table 3. Effect of dietary polyzyme supplementation on feed intake, feed conversion ratio* at 3 and 5 weeks and European Efficiency index at 5 weeks of broiler chicks of age (Means \pm S.E).

Treatments ¹	Feed intake (g/ bird/ day)		Feed conversion ratio		European efficiency index, % ⁴
	3 weeks	5 weeks	3 weeks	5 weeks	5 weeks
T1	55.31 ^{a2,3} \pm 1.59	95.80 ^a \pm 1.42	1.27 ^a \pm 0.11	1.77 ^a \pm 0.38	190.27 ^b \pm 7.12
T2	52.09 ^b \pm 0.50	92.81 ^a \pm 1.60	1.30 ^a \pm 0.12	1.81 ^a \pm 0.18	174.19 ^b \pm 5.45
T3	49.37 ^c \pm 2.19	88.49 ^b \pm 2.12	1.18 ^b \pm 0.014	1.54 ^b \pm 0.31	251.74 ^a \pm 6.23
SIG	*	*	*	*	*

¹T₁: Positive control, T₂: Negative control, T₃: Negative control + 0.5g polyzyme.

² means \pm S.E. of 3 replicates / treatment.

³a,b,c.....etc: Means within the same column with different superscripts are significantly different ($P < 0.05$).

⁴ European efficiency index, EEI= (Mean body weight (kg) \times livability %) / (marketing age, days \times feed conversion ratio) \times 100, cited by Soltan and Kusainova, 2012.

* feed conversion ratio= (feed intake, g/ d) / (body weight gain, g/ d)

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Whereas, performance index was significantly increased by the supplementation, T_3 (81.42%) in comparison with chicks fed the positive control and the low protein diets (72.56 and 65.05%), respectively. These results confirm that, the effects of enzyme could extend to the overall growth period. Our results are agree with the results of Zhu *et al.* (2014) who noted that enzyme supplementation had no major effects on average daily gain (ADG), average daily feed intake (ADFI) and feed: gain ratio. The reason might be due to that the study tested only the first 21 day. Similarly, previous studies of Kocher *et al.* (2002); Meng and Slominski, (2005) showed that enzyme addition to the corn – soybean diet did not affect chick performance.

During the finishing periods chicks fed the supplemented diet had significantly the highest BW and BWG (2092g and 2052g) followed by chicks fed the positive control diet (1951g) compared to those fed the negative control diet (1865g and 1825g). The same trend was noticed at performance index (PI) which recorded 91.15, 67.73 and 60.97 % at chicks fed the supplemented, positive control and un-supplemented diets, respectively.

Chicks fed the negative control diet without and with 0.5g polyzyme had significantly decreased feed intake to reach approximately 94.18, 89.26% and 96.88, 92.37%, respectively of the control group (100%) at the starting and the finishing periods. Feed conversion ratio (FCR) was significantly improved by the supplementation. There were no significant differences in both positive and negative control groups in FCR, but it recorded a significant improvement with the supplementation.

The best FCR was 1.54 for chicks fed diets supplemented with 0.5 polyzyme followed by 1.77 for the positive control group. While, the worst FCR was for chicks fed the negative control diets; being 1.81.

European efficiency was increased by the supplementation being 251.74% in chicks fed the negative control diet + 0.5g polyzyme compared to 190.27 and 174.19% for chicks fed the positive and negative control diets, respectively. The improved BW in supplemented groups may indicate that the addition of enzymes is essential for the utilization of low protein diet. Also, enzyme supplementation improved the digestibility of corn and soybean meal. Thus, the effect of enzymes with corn soybean meal resulted in higher BW than the enzyme negative control or the positive control groups.

These findings are supported with those obtained by (Noy and Sklan, 1995 and Uni *et al.*, 1995) who suggested that the inclusion of enzymes during the initial phase of the chickens may improve the digestibility of the diet and the performance of the bird.

On the other hand, Aboudabos and Yehia (2013), Zhu *et al.* (2014) and Fernandes *et al.* (2015) did not observe differences in the performance of broilers that were supplemented with enzymatic complex for 1- 21 days of age with diets based on corn and soybean meal.

Our results indicated that reducing crude protein (CP) caused poorer BWG and FCR because lowering the CP levels in the diet decreased protein intake and decreased the final body weight and considering the economic efficiency values of birds fed the reduced CP diets were more costly than those in the other groups.

Elmakeel *et al.* (2007), Giraldo *et al.* (2008) and Abdel- Hafeez *et al.* (2016) noted that the impact of enzyme is attributed to its activity in breaking the polymeric chains of non-starch polysaccharides (NSP) found in feed, so improving the digestibility rate and the nutritive value.

Carcass characteristics:

The effect of polyzyme supplementation on carcass characteristics of growing broilers at 5 weeks of age is presented in Table (4). There were significant differences between the treatments on all observed carcass characteristics. Supplementation of 0.5g polyzyme to low protein diets significantly increased dressing percentage and the percent of liver and heart which suggests that there was a positive response to increase calorie – protein ratio when enzyme was supplemented.

Gizzard percentage was affected by the supplementation to the low protein diets but not significant. It's clear that

inadequate CP consequently amino acid negatively influence the broiler carcass composition (dressing and giblets percentage) in comparison with the control and the negative control diet supplemented with exogenous enzymes. These results are coincides with Si *et al.* (2001), Cafè *et al.* (2002), and Furlan *et al.* (2004) who reported that chicks fed diets supplemented with enzyme had significantly carcass yields.

Blood components:

The present results of some blood components Table (5) showed that polyzyme supplementation to low protein diet improved serum total protein, creatinine and glucose but significantly reduced total cholesterol. Enzyme supplementation had significantly reduced blood serum AST, while ALT concentrations was not significant when compared to chicks fed the same diet without polyzyme supplementation (27.50 and 14 U/ L vs. 32.00 and 18.50 U/ L), respectively.

Table 4. Effect of dietary polyzyme supplementation on carcass characteristics of growing broiler chicks at 5 weeks of age (Means ± S.E).

Treatments ¹	Carcass traits (%)			
	Dressing	Giblets		
		Liver	Heart	Gizzard
T1	88.47 ^{ab2,3} ± 0.42	2.78 ^a ± 0.01	0.42 ^a ± 0.03	2.22 ± 0.12
T2	88.03 ^b ± 0.20	2.43 ^b ± 0.07	0.29 ^b ± 0.01	1.99 ± 0.23
T3	89.74 ^a ± 1.26	2.84 ^a ± 0.06	0.49 ^a ± 0.04	2.30 ± 0.10
SIG	*	*	*	NS

¹T₁; Positive control, T₂; Negative control, T₃; Negative control + 0.5g polyzyme.

² means ± S.E. of 3 replicates / treatment.

³a,b,c.....etc: Means within the same column with different superscripts are significantly different (P < 0.05).

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Table 5. Effect of dietary polyzyme supplementation on some blood components at 5 weeks of age (Means ± S.E).

Treatments ¹	Total protein	Creatinine	Glucose	Total cholesterol	AST	ALT
	----- mg/ dl -----			----- U/ L -----		
T1	4.50 ^{2,3} ± 0.17	0.70 ± 0.01	248.00 ± 18	145.00 ^a ± 1.20	25.50 ^b ± 1.50	17.50 ± 4.50
T2	3.95 ± 0.19	0.86 ± 0.03	228.00 ± 16	139.50 ^b ± 1.50	32.00 ^a ± 2.10	18.50 ± 3.50
T3	4.60 ± 0.15	0.687 ± 0.06	240.50 ± 3.5	121.00 ^c ± 3.50	27.50 ^{ab} ± 5.07	14.00 ± 2.00
SIG	NS	NS	NS	*	*	NS

¹T₁; Positive control, T₂; Negative control, T₃; Negative control + 0.5g polyzyme.

² means ± S.E. of 3 replicates/ treatment.

³a,b,c.....etc: Means within the same column with different superscripts are significantly different (P < 0.05).

Measurements of AST and ALT activities are indicative of liver damage in broiler chicks and it's therefor available tool for determination of a safe inclusion rate for feed additives. Based on these findings, enzyme blend administered at the level of 0.5g/ kg diet in this study may not exert adverse effects on broiler chickens. The obtained data are in harmony with those reported by Hamada *et al.* (2015) and Jariyahatthaakij *et al.* (2018).

Economic efficiency:

Results of feeding cost for chicks fed the experimental diets are presented in Table (6). These results considerable saving with using polyzyme and low protein level as compared to the positive control group (T₁).

Differences in feeding cost showed that diets supplemented with exogenous enzyme decreased the feeding cost/ kg live body weight (18.04 L. E.) compared to the control group (21.11 L. E.) about 14.50% saving. This saving in feeding cost was due to the decreasing cost of

low protein diets and due to the increase in live body weight and the improvement in FCR using the polyzyme.

On the contrary, using low protein diets without enzyme supplementation resulted in increasing feed cost/ kg body weight compared to the supplemented group. Data also showed that the highest relative economic efficiency value was observed with chicks fed low protein diets supplemented with 0.5g polyzyme (154.44%) vs. 100% and 115.56% at chicks fed the control and the low protein diets.

Also, European efficiency index confirms these results and tested chicks had 251.74 compared to 190.27 and 174.19 % in the control and low protein groups.

CONCLUSION:

Data indicated that the beneficial economical effect of using polyzyme with low protein diet is to reach almost the same performance with normal protein diet.

Table 6. Effect of dietary polyzyme supplementation on the economic efficiency of the experimental diets at 35 days.

Items	Treatments ¹		
	T ₁	T ₂	T ₃
Initial body weight, (g)	40.00	40.00	40.00
Final body weight, (kg)	1.95	1.87	2.09
Body weight gain, (kg)	1.91	1.83	2.05
Total revenue,(L.E) ²	40.11	38.43	43.05
Feed intake, (kg)	3.35	3.25	3.10
Price of kg feed,(L.E)	6.30	5.77	5.82
Feed cost, (L.E)	21.11	18.75	18.04
Net revenue,(L.E) ³	19.00	19.68	25.01
Economical efficiency ⁴ .	0.90	1.04	1.39
Relative economic efficiency, (%)	100	115.55	154.44
European efficiency index ⁵	190.27	174.19	251.74

¹T₁; Positive control, T₂; Negative control, T₃; Negative control + 0.5g polyzyme.

Price of one kg live body weight was 21 L. E.

Price of one kg polyzyme powder 100.90 L.E.

²Total revenue = live body weight gain × marketing price

³Net revenue = Total revenue – Feed cost.

⁴Economical efficiency = Net revenue / Feed cost.

⁵ European efficiency index, EEI= (Mean body weight, kg × livability %) / (marketing age, days × feed conversion ratio) × 100, cited by Soltan and Kusainova, 2012.

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تأثير إضافة البولييزايم إلى العلائق المنخفضة البروتين على أداء كتاكيت التسمين

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الملخص العربي

Influence of dietary polyzyme on the performance of broilers fed low crude

أجريت هذه التجربة لدراسة تأثير إضافة البوليزايم (polyzyme) في علائق كتاكيت التسمين المنخفضة البروتين على كل من كفاءة النمو، صفات الذبيحة، بعض مكونات الدم والكفاءة الاقتصادية. استخدم عدد 180 كتكوت روس 308 غير مجنس عمر يوم - قسمت عشوائيا إلى 3 مجموعات بكل منها 3 مكررات (20 كتكوت/ مكررة)، وكانت المعاملات التجريبية على النحو التالي: المعاملة الأولى: عليقة المقارنة (23، 20% بروتين خام)، المعاملة الثانية: عليقة منخفضة البروتين (20، 17% بروتين خام) وبدون إضافة البوليزايم، المعاملة الثالثة: عليقة منخفضة البروتين (20، 17% بروتين خام) مضاف إليها 0,5 جم بوليزايم/كجم عليقة خلال فترة البادئ (1- 21 يوم) وفترة الناهي (22- 35 يوم) على التوالي. وقد أوضحت النتائج أن إضافة البوليزايم إلى العليقة المنخفضة البروتين (المعاملة الثالثة) كانت الأعلى معنويا في وزن الجسم والزيادة في وزن الجسم ودليل الأداء - بينما أدت الإضافة إلى تقليل الغذاء المأكول وتحسن كل من: معدل تحويل الغذاء، دليل الكفاءة الأوروبية والكفاءة الاقتصادية، كما زادت نسبة كل من: التصافي والكبد والقلب - بينما لم تتأثر نسبة القانصة - وانخفض معنويا مستوى الكوليستيرول الكلي وازداد مستوى إنزيم AST معنويا - ولم يتأثر مستوى كل من: البروتين الكلي، الكرياتينين، الجلوكوز وكذلك إنزيم ALT - مما يشير إلى إمكانية استخدام البوليزايم بمستوى 0,5 جم/كجم عليقة تسمين منخفضة في محتواها من البروتين دون حدوث أضرار. وبصفة عامة أوضحت النتائج أن العليقة المنخفضة البروتين (20، 17% بادئ وناهى) مع إضافة البوليزايم بمستوى 0,5 جم/كجم عليقة يتساوى أو يفوق عليقة المقارنة (23، 20% بادئ وناهى) بدون إضافة البوليزايم. ويوضح ذلك الفائدة الاقتصادية لاستخدام البوليزايم مع العلائق المنخفضة البروتين.

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