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A STUDY ON SYNERGISM BETWEEN TWO DIFFERENT SUPPLEMENTAL CARBOHYDRASES ENZYMES TO STANDARD OR LOW ENERGY BROILER DIETS

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ABSTRACT: The objective of the present study was to investigate the synergism relationship between xylanase (Xyl.) and pectinase (Pec.) enzymes when applied in different levels of metabolizable energy (ME) broiler diets based on corn-soybean meal. The study included two experiments. Experiment1 (EXP1) studied the effects of Xyl. and/or Pec. supplementation to corn-soybean standard diet. Experiment 2 (EXP2) examined the enzymes supplemented to different reduced ME diets (lower 100 kca/kg, NC100; and lower 150 kcal/kg, NC150) than control. Growth performance, carcass characteristics, and meat quality of broiler were carried out. The obtained results were: EXP1 showed that enzymes supplementation (Xyl., Pec. or both of them) to broiler diets resulted in significant improvement of body weight (BW), body weight gain (BWG), feed conversion ratio (FCR) and breast meat yield%, while feed intake (FI) were decreased. Enzyme combinations (Xyl. and Pec.) improved dressing percentage, also lead to significant improvement in total protein (TP) of broiler meat, whereas the individual enzyme had no effect. EXP2: Reducing ME values from normal to NC150 level caused significant reduction in final BW, BWG, carcass edible parts percentage, MDA and TP of broiler meat, and cause significant increase in FI and drip loss of breast meat and drum stick samples percentage of broiler meat. The combination of Xyl. and Pec. supplemented to NC100 and NC150 diets significantly improved growth performance more than the individual enzymes, while, the best FCR was recorded in NC100 supplemented with the combination of Xyl. and Pec. enzymes. The NC150 supplemented with Xyl. and Pec. showed better carcass total weights and dressing percentage compared with other treatments, and the lowest abdominal fat recorded in NC100P. and NC150. In conclusion, the combination of Xyl. and Pec. is more effective than the individual in growth performance and quality of carcass and meat.

Keywords:broiler-pectinase-xylanase-metabolizable energy-performance- meat quality.

INTRODUCTION

The major cost in poultry production is the feed cost (about 70%), with dietary energy sources occupying the greatest portion (70 to 75% of the diets) (Van der Klis *et al*., 2010). Dietary energy level is the main factor influencing feed intake, as birds tend to eat feeds mainly to satisfy their energy requirements and once this is met, they will not consume any more feeds, even if the requirements of other nutrients like protein, vitamins or minerals have not been met (Singh and Panda 1992). For this reason, numerous studies have been carried out to investigate the possibility of reducing energy by adding feed enzymes (Pack and Bedford 1997; Min *et al*., 2011 and Abou El-Wafa *et al*., 2013 and Selim *et al*. 2015).

In Egypt, poultry diets are based on corn and soybean meal generally which, in spite of presenting high digestibility, can be better utilized. The metabolizable energy (ME) of soybean meal is notably low for poultry due to Non-Starch Polysaccharides (NSPs) Pierson *et al*., (1980). Studies with broilers fed those ingredients have confirmed that the use of exogenous enzymes individually or in combination improves dietary nutrient utilization, resulting in more uniform animal performance (Cowieson and Adeola 2005; Lu *et al*. 2013; Selim *et al*., 2015 and 2016), and this exogenous carbohydrates could increase AME contents (Kocher *et al*., 2002; Gracia *et al*., 2003) and degradation of NSPs (Mandels 1985; Brenes *et al*., 1993; Frigard et al., 1994; Mcknight 1997; Bedford and Schulze 1998; Mathlouthi *et al*., 2003 and Saki *et al*., 2005).

Reducing ME of broiler diets 100 and 150 kcal/kg diet than standerd resulted in significant depression of BWG and FCR (Downs *et al*., 2006; Golian *et al*., 2010; O'Neill *et al*., 2012; Abou El-Wafa *et al*., 2013 and Selim *et al* 2015), while, adding xylanase enzyme to low (by 100 kcal/kg diet) metabolizable energy corn-soybean meal broiler diets improved growth performance (Nian *et al*., 2011; and Williams *et al*., 2014; Pirgozliev *et al*., 2015 and selim *et al*., 2015). On the other hand, pH value of broiler meat increased by the increasing energy level of diets (Tang *et al*. 2007), while, xylanase supplementation to low energy broiler diets resulted in decreasing pH value of broiler meat (Selim *et al*., 2015) with increase shelf-life of muscles (Allen *et al*., 1997).

Exogenous carbohydrases has been reported to improve energy utilization and the performance of broilers (Bedford 1995; Olukosi and Adeola 2008 ;Cowieson 2010 and Williams *et al*., 2014) by hydrolysis of cell wall arabinoxylans and improve the access of endogenous digestive enzymes to cellcontents (Kocher *et al*., 2003; Meng *et al*., 2005; Omar *et al*., 2008 and Francesch and Geraert 2009).

Lima (2005) reported that the harm effects of antinutritional factors could be reduced by using supplemental enzymes which break specific links and change the overall chemical structure of that antinutrional factors, consequently the available nutrients in the diets increased. Also the author showed that the endogenous enzymes by digestive tract cannot or have limited ability to do that analysis. Youssef *et al*., (2011) observed that using a mixture of enzyme preparation containing xylanase and amylase added to corn-based diet could reduce the dietary energy level (about 150

kcal/kg) without a negative effect on growth performance or carcass traits. While, Gehring *et al*., (2013) concluded that responses to exogenous enzyme supplementation are influenced by type of feed ingredients (not constant) and other factors. Besides, Abudabos (2012) concluded that adding a commercial enzyme (containing 30 U g^{-1}unites pectinase) to both low density diet (less ME diets by 180,150 and 170 kcal/kg for starter, grower and finisher respectively) could significantly improve the body weight of broilers at 42 days, the values of overall feed conversion ratio were improved by either enzyme supplementation, increasing ME of diet, or their interaction.

According to Barbosa *et al*., (2012); Brito *et al*., (2006) and Leite *et al*., (2011) the addition of enzyme blends (including xylanase) can be added to diets with reduced nutrient levels (metabolizable energy, calcium, and phosphorus content) aiming to maintain broiler growth performance. Nunes *et al*., (2015) reported that the supplementation of enzyme blends containing xylanase to diets with reduced calcium, phosphorus and energy levels allowed provided better performance results than the enzyme complex containing pectinase.

Previous work showed that reducing ME (by 100 or 150 kcal/kg) lead to reduce broiler BW and BWG and increase FCR and FI. Supplementing xylanase to broiler corn-Soybean meal standard or low 100 or 150 kcal/kgm ME diets improved growth performance and meat quality, whereas pectinase enzyme supplementation to standard or low ME diets had no effect on growth performance and improved meat quality. Besides the physical and chemical examination of broiler meat quality showed clear improvement of meat quality by xylanase or pectinase supplementation. Reducing ME had a bad effects on meat quality, these effects could be treated with enzyme supplement (xylanase or pectinase) (Selim *et al*., 2015 and 2016).

The objective of the present study was to investigate the effect of applying two different carbohyrases enzymes (xylanse, pectinase and their combination) in diets with standard or low energy levels on the performance, carcass characteristics, and meat quality of broilers. If there a synergism relationship between xylanase and pectinase or not? Is a question to answer?

MATERIALS AND METHODS

Experiment 1:

Experimental diets and birds:

A total of 160 day-old Arbor Acres broiler chicks were allocated in four dietary treatment groups. Each treatment had four replicates, with 10 birds per replicate. The control fed with corn soybean basal broiler starter, grower, and finisher diets until the age of 10, 24 and 40 days, respectively. The second group fed on the control diet supplemented with 16000 U xylanase/kg diets (Econase XT25 at level 100 mg/kg diet). The third group fed on control diet supplemented with 1000U pectinase enzyme/kg diet. The fourth group fed on the control diet supplemented with both previous doses of xylanase (Xyl.) and pectinase (Pec.) enzymes. All chicks were reared on wire battery cages and subjected to the same managerial and veterinary procedures throughout the growth trial term during the experimental period (1-40 d of age). Birds were fed *ad libitum* diets and had access to water. The compositions of the experimental basal diets are shown in Table 1. Parameters of growth

performance including body weight (BW) and feed intake (FI) were recorded, also, body weight gain (BWG) and feed conversion ratio (FCR) were calculated during the experimental period.

Slaughtering and carcass characteristics:

At the end of the experimental period, four birds with the same body weight of each group mean were slaughtered to record carcass characteristics. In carcass characteristics, carcass, abdominal fat and edible parts (liver, gizzard and heart) were recorded. All carcass characteristics were calculated as percentage of birds body weight. For each sample, front quarter (FQ) which include breast quarter muscles and skin (without wing) and skinned back quarter (BQ) which include drumstick and thigh were weighted and calculated as a percentage from carcass weight. Drumstick (DS) and thigh samples were weighted and calculated as percentage of BQ. Skin weight of every part was recorded as percentage of its part. All the samples of carcass cuts were kept for 24 h at 4°C to complete the physical and chemical evaluation of broiler meat.

Meat quality measurements

Physical measurements of broiler meat: Drip loss of the 48 meat samples (16 breast, 16 thigh and 16 drumsticks) were described by Saenmahayak *et al*., (2012). Samples preparation and drip loss calculation were done according to Selim et al., (2013). During 24 h of chilling the samples on 4°C were used to measure ultimate pH (pHu) as described by Selim *et al*., (2013).

Chemical measurements of broiler meat: Breast, thigh and drum stick (DS) meat of each slaughtered bird were stored on -20°C for 60 days before used to determine total protein (TP), low density lipoprotein (LDL) and high density lipoprotein (HDL) and malondialdehyde (MDA), in meat extract by Colorimetric methods using analytical commercial kits produced by Biodiagnostic Company, Egypt (Selim et al., 2014).The extract of broiler meat was prepared using phosphate buffer pH 7.4 (10 ml/1g of sample)

Statistical analysis:

Statistical analyses were performed using one-way analysis of variance using the general liner model (GLM) procedure of SAS User's guide (SAS, 2001). The significance of difference between means was determined by Duncan's multiple range test (Duncan, 1955) and the least significant difference at $p \leq 0.05$ as the following model:

$Yi = \mu + Ti + ei$

Where: Yi=The observation; μ=Overall mean; Ti=Effect of treatments (i=1,2,3,4); ei=Random error.

Experiment 2:

Experimental diets and birds:

A total number of 360 one day old Arbor Acres broiler chicks were equally subjected to 9 treatment (2 reduced metabolizable energy × 4 enzyme supplementation+ control group). The control group fed on standard broiler diets without supplementation during all stages of the experiment. The low ME (negative control) levels were lower than standard values at each period by 100 Kcal (NC100) and 150 Kcal (NC150). The enzymes supplementations were: without (free), 16000 U of Xyl/kg diet (X), 1000 U pectinase/kg diet (P), and both of supplemental doses of xylanase and pectinase (XP). The starter period (1-10 d) diets contained 3000, 2900 or 2850 kcal/kg, the grower period (11-22 d) contained 3100, 3000 or 2950 kcal/kg and the finisher period (23-40 d) contained

3200, 3100 or 3050 kcal/kg. The compositions of the experimental diet are shown in Table 1. The chicks were reared for 40 days on wire battery cages and subjected to the same managerial and veterinary procedures throughout the growth trial term, diets and water were fed ad libitum. Parameters of growth performance including body weight (BW) and feed intake (FI) were recorded, also, body weight gain (BWG) and feed conversion ratio (FCR) were calculated during the experimental period.

Slaughtering and carcass characteristics:

At 40 days of age, a total number of 36 birds (four birds / treatment) were slaughtered and carcass traits were carried out as in the first experiment

Meat quality measurements

Physical and chemical examination of 72 samples of broiler meat were carried out as described in the first experiment.

Statistical analysis:

The effect of pectinase and/or xylanase supplementation were statistically analyzed in a two way analysis of variance using the general liner model (GLM) procedure of SAS User's guide (SAS, 2001) as the following model:

$Y_{ijk}= \mu+X_i +M_j +(XM)_{ij} + e_{ijk}$.

Where:

Y_{ijk}= Trait measured

μ = Overall mean

X_i = Enzymes supplementation i= (1, 2,3,4)

M_j=Metabolizable energy levels j= (1,2)

$(XM)_j$ = Interaction between xylanase supplementation and metabolizable energy level.

e_{ijk}= Experimental error

All the experimental treatments (9) were analyzed using the one way analysis of variance to detect the differences between all treatments as following model:

$Y_{ij} = \mu+Ti+e_{ij}$

Where: μ = overall mean of Yij, T_i = effect of treatment, i = (1,…, 9)

e_{ij} = Experimental error

Duncan's multiple range test (Duncan, 1955) were applied to separate the means. A probability (P) value of less or equal to 0.05% was considered statistically significant.

RESULTS AND DISSCUSION

Experiment 1 (Exp1)

Growth performance

The results of growth performance including the final live body weight (BW), feed intake (FI), body weight gain (BWG), and feed conversion ratio (FCR) of broiler chicks in Exp1 at 40 days of age are shown in Table (2). All enzymes supplementation treatments led to improve growth performance compared to control group. The supplementation of Xyl, Pec or a mixture of both to corn-soybean basal broiler diets were significantly increase BW and BWG, and reduced the values of FI compared with the control. While, FCR improved by 7.4 and 6.3% with xylanase or pectinase supplementation, respectively. The supplemented diets with both Xyl and Pec resulted in increasing BW and BWG and reducing FI which led to 9.7% improvement in FCR compared with the control.

The results showed that, the combination of Xyl and Pec that supplemented to the soybean basal diet is more effective in improving growth performance than the individual supplementation of either of them.

In connection with xylanase enzyme supplementation, results obtained here were in agree with those reported previously by (Cowieson, 2005; Nian *et*

al., 2011; and Williams *et al*., 2014; Selim, *et al*., 2015, and Pirgozliev *et al*., 2015). While, Cowieson (2010) reported an increasing in BWG and improving in FCR with no effect on FI. While, Kocher *et al*., (2003) and Singh *et al*., (2012) failed to record significant effect of Xyl on growth performance of broilers.

In connection with pectinase enzyme supplementation, the improvement of broiler performance were reported by Igbasan and Guenter (1996); Cowan *et al*., (1999); and Khalil *et al*. (2014). While, Igbasan *et al*., (1997) and Selim *et al*., (2015) reported that pectinase supplementation had no effect on broiler performance when supplemented to corn soybean diet at level of 1000 U/kg diet.

The recorded higher improvement effect on growth performance parameters by using supplemented diet with a mixture of both Xyl and Pec enzymes compared with control or individual enzyme supplement were inagreement with Khalil *et al*., (2014) who found that the combination of pectinase and xylanase greatly reduced water-holding capacity, viscosity and cell wall content, compared to pectinase or xylanase alone.

Perez-Maldonado *et al*., (1999) and Jia *et al*., (2008) reported that supplementing legume grains with exogenous enzymes can break down the non starch polysaccharides (NSPs) and releasing cell contents and overcoming the negative effects of supplementing legume grains (Ali *et al*., 2009). Furthermore, enzymes will work synergistically (Ravindran *et al*., 1999 and Wu *et al*., 2004) to break down pectins and xylans complex compounds, and explained the synergism effect by breaking the pectin main chain compound firstly by supplemental Pec which increase the increase the ability of supplemental Xyl to break the xylan side branches attached to main pectin backbone.

Carcass characteristics:

The recorded results of carcass characteristics of broiler chicks of experimental treatments at 40 d of age are presented in Table (3). The obtained results showed significant enhancement of dressing % by adding either Xyl+Pec or Xyl alone, while pec. supplementation could not cause any significant change of dressing % compared to control unsupplemented group. Supplementation of Xyl+Pec significantly increased dressing % by 4.6% compared to unsupplemented control group. There were no significant effects of experimental treatments on either abdominal fat % or edible parts %. The determined mean values of breast meat yield % significantly increased by 24.9, 12.4 and 14.7% for chicks fed on diet supplemented with Xyl., Pec., and Xyl.+Pec., respectively compared to value of control group. On the other side, the determined values of thigh, DS and total of BQ weights did not changed as a response to experimental treatments. Regarding to the all treatments the chicks fed on Xyl and Xyl+Pec significantly increased the % of thigh skin by 20.6 and 16.44%, respectively compared with control, while supplemented Pec. didn't affected it. These may be prove that single supplementation of Pec. could not release more energy from feed as Xyl. supplementation either alone or with combination with Pec. Previous studies (Selim *et al*., 2015) recorded the enzyme supplementation field to show that Xyl. supplementation significantly enhanced both dressing % and breast meat yield%, while single Pec. supplementation had no effect on the same measurements (Selim *et al*., 2016) when added to corn soybean

broiler diet. The results obtained here confirmed the reported results by some workers about response of carcass measurements to Xyl. supplementation whereas it significantly increased dressing (Medhi *et al*., 2003; and Khan *et al*., 2006), and that may be due to the ability of Xyl to increase energy utilization of broiler diets (Williams *et al*., 2014).While other worker suggested that this dressing improvement might be due to improve the growth rate of broiler (Medhi *et al*., 2003 and Khan *et al*., 2006) by improving absorption of starch, fat and amino acids (Oloffs *et al*., 1999; Bedford, 2000; Ziggers, 2006 and Serena *et al*., 2009).

Meat quality:

The determined results of some meat quality parameters of broiler chicks fed on diet supplemented with Xyl., Pec., or Xyl.+Pec. are shown in Table (4). The pH_u of breast, thigh and DS did not affected significantly by any of experimental treatments. The control group had shown generally lower values of drip loss in thigh and drum stick as compared to other supplemented groups.

There were no significant effects of the exogenous supplementation of Xyl., Pec. or Xyl.+Pec. enzymes on the MDA, LDL and HDL in broiler meat extract, while, TP was significantly increased by 29.2% with Xyl.+Pec. supplemented diet compared to control unsupplemented group.

These results were in agreement with those reported by (Allen *et al*., 1997; Tang *et al*., 2007 and Zakaria *et al*., 2010) about effect of xylanase on broiler meat. In previous study, Selim *et al*., (2015) reported that xylanase supplementation to corn-soybean meal broiler diets significantly reduced pHu of both breast meat and thigh accompanied with significant reduction of drip loss % of thigh. Also, the chemical quality measurements of broiler meat including MDA, LDL, and HDL did not changed with enzymes single supplementation of Xyl or Pec (Selim *et al*., 2015 and 2016). On the same trend, some workers reported that supplemented diet with mixture of enzymes did not significantly improve meat quality traits (Jeroch *et al*., 1991 and Zakaria *et al*. 2010).

Xylanase supplementation increased drip loss %, which increased the ability of breast and thigh meat to processing whereas water holding capacity and glycogen were increased (Barbut, 1993 and Zhang and Savage, 2010).

The overall results of Exp1 showed clear synergism effect between both Xyl and Pec enzymes when supplemented to corn-soybean broiler diets. The combination of both enzymes could enhance the energy utilization, which reflected on growth performance, carcass and meat quality.

Experiment 2 (EXP2)

Growth performance

The recorded results of broiler growth performance parameters at 40 days of age are shown in Table (5). The results of main effect of reduction of ME of feed showed significant depression of all growth performance parameters due to decreasing ME to NC100 and NC150. Among enzyme supplementations, the best overall growth performance recorded by using combined supplementation (XP) compared with other supplementations. The free supplementation group recorded the worst values of all growth performance parameters, while there is no significant difference between main effects of X and P which reflect the synergism between X and P when supplemented to reduced ME diet as

reported in Exp1 by using standard (sufficient energy) diets.

Among experimental treatments, all treatments recorded better BW and BWG except treatment NC150 when compared with control group. The best values of all growth performance parameters recorded by chicks fed on NC100XP (FCR= 1.71), while birds of NC150XP consumed the highest quantity of feed which led to increase the value of FCR (1.76).

The overall growth performance showed that, although the combination of XP could enhance the growth performance when supplemented to NC100 diet, while that combination failed to release sufficient ME from feed when added to NC150 diet to compensate the reduction of ME in NC150 diet.

Regarding to the effect of ME reducing on broiler performance the recorded results in this study confirmed those published by Downs *et al*., (2006); Golian *et al*., (2010); O'Neill *et al*., (2012) and Selim *et al*., (2015). Also, Abou El-Wafa *et al*., (2013) found that the effect of dietary ME level reveal a significant decrease in total BWG associated with a corresponding increase in total FI and a worsen in FCR as the dietary ME decreased 50 and 100 kcal/kg diet.

O'Neill *et al*., (2012) reported that supplemented xylanase could improve the value of overall FCR when supplemented to 100 kcal reduced ME broiler diet. In addition more recent studies confirmed the same trend (Abou El-Wafa *et al*., 2013; Pirgozliev *et al*., 2015).

Supplemented pectinase enzyme to low ME diets by 100 or 150 kcal/kg diet could not improve growth performance parameters (Igbasan *et al*. 1997 and Selim *et al*., 2015). On the contrary, Cowan *et al*. (1999); Igbasan and Guenter (1996) and Khalil *et al*. (2014) reported that supplementing diets with pectinase enzyme improved broiler performance.

Many researchers reported significant improvements of growth performance of broilers by using mixture of enzymes supplemented to lower ME diets (Kocher *et al*., 2002 ;Gracia *et al*., 2003; Tahir *et al*., 2008; and Abudabos, 2012) . Khalil *et al*. (2014) confirmed the same trend when using a mixture of xylanase and pectinase enzymes to broiler diets contained lupine kernel. They reported that synergism effect might be due to that the changes in physico-chemical properties of lupine kernel were less when both enzymes were individually supplemented than that achieved by combination of them (Khalil *et al*. 2014). The positive effect of exogenous enzymes supplementation to broiler diets on growth performance may be due to that supplementing diets with exogenous enzymes can break down the NSPs (Perez-Maldonado *et al*., 1999; Jia et al., 2008), releasing cell contents and overcoming the negative effects (Ali *et al*., 2009).

Carcass characteristics:

Results in table 6 clearly showed that, reduction of ME of broiler diets from 100 to 150 kcal caused a significantly reduction (6.4%) in carcass edible parts %, however values of dressing and abdominal fat % did not significantly affected. The values of FQ and BQ did not significantly changed by reducing ME from 100 to 150 kcal. The results of main effect of enzyme supplementation showed that both X and XP supplementation caused significant increase of dressing and abdominal fat % compared to without or P groups. That meaning that P as single enzyme supplementation could not able to release more ME from feed ingredients.

Generally, chicks fed on NC150XP or NC150X significantly gained the highest dressing % compared to other treatments, followed by NC100X. Regarding to abdominal fat % birds fed on NC100, NC150, NC100P and NC150P recorded lower % compared to other treatments , which confirmed the lower ability of P supplementation to release more ME in the diet. There was significant increase of BQ% from carcass due to feeding on NC150XP or NC150P compared with both control and NC100 groups. While the rest of examined carcass parts did not significantly affected by different treatments.

These results are in match with AbouEl-Wafa *et al*., (2013) who found no significant effect of xylanase on dressing % but the % of abdominal fat was decreased by 25.41% compared with unspplemented group in broiler fed with diets based on corn or corn/rye at recommended and reduced ME levels (50 and 100 kcal/kg). Previuous reports by NarsimhaRao (1998) and Hanumantharao *et al*., (2003) stated no effect of carbohydrases on dressing % and general carcass traits of broilers.

The overall carcass traits results showed that the interaction (P x ME) might be increased the breast meat yield, BQ and DS % quarter by the enzyme which could be able to restore the nutritional value in the low density diet by releasing the entrapped protein during disruption of the cell wall matrix (Chesson, 2001 and Bedford, 2000). Min *et al*., (2011) obtained that when broiler were fed on diet supplemented with Rovabio Max 200g/tone (containing pectinase) and low energy diet (40 kcal/kg diet) didn't differ significantly compared with control group in dressing and breast meat yield %.

In the present research the combination of pectinase and xylanase improved dressing % compared to control, it seems likely that some enzymes will work synergistically (Ravindran *et al*., 1999; Wu et al., 2004) to break down pectins and xylans compounds. Also, Khalil *et al*. (2014) reported that when pectinase and xylanase were used alone, the changes in physico-chemical properties of diets were less than that achieved by combination of two enzymes.

Meat quality:

Table (7) presents the effect of reduction of ME, enzymes supplementations and different treatments on some physical and chemical broiler meat quality properities. Reduction in ME from NC100 to NC150 levels increased the drip loss % of breast meat and DS, while drip loss of thigh and pHu of all examined carcass parts did not affected. The main effect of enzyme supplementation showed clear superior effect of X to increase drip loss of breast meat and thigh while drip loss of DS showed the opposite trend. Using NC150XP diets gave higher values of drip loss of breast meat samples, while thigh drip loss values by NC100X and DS drip loss by NC150 significantly higher as compared with values of other treatments.

The chemical composition did not affected by the reduction of ME from 100 to 150 except for MDA and TP or by the examined enzyme supplementation. While the one way analysis of variance of all treatments showed significant reduction of MDA by applying NC150XP and significant increase of TP by applying NC150P. Birds fed on control diets showed the highest LDL value in broiler meat extract compared with all other treatments. On the other side, the determined values of HDL in broiler meat

did not changed as a response to any treatment.

The overall results of broiler physical and chemical meat quality examination showed clear improvement of breast meat pHu by both xylanase supplementation and low ME level and general enhancement of chemical quality and self life of produced meat and this was in agreement with our previous works (Selim *et al*., 2015 and 2016). Also, the recorded results were in agreement with those reported by (Allen *et al*., 1997 and Tang *et al*., 2007) who reported that pH value of broiler meat increased by the increasing energy level of diets, and pH value of broiler meat decreasing with xylanase supplementation to low energy broiler diets. On the other side, Zakaria *et al*. (2010) reported that the addition of mixture enzymes (containing pectinase) did not significantly impact meat quality traits of broiler (pH, cooking loss and water holding capacity).

Tahir *et al*., (2006); Kocher *et al*., (2002) and Saleh *et al*., (2005) used multi-enzyme preparations obtained positive effect on protein digestibility. Also, Slominski and Campbell, (1990) and Simbaya *et al*., (1996) explained that improvements by the digestion of pectin polysaccharides and increase of the available nutrients which reflect on the growth and muscles formation. Furthermore, Tahir (2008) recorded that the enzyme preparation stimulated protein digestion when added to broiler diets and indicated that degradation of pectin compound enables protein and energy, especially the protein of broiler feed, to become more digestible.

Enzyme supplementation decreased glycogen storage in breast meat and releases energy to chicks' metabolic use, as there was a reported relation between breast pH and glycogen storage by Le-Bihan-Duval *et al*. (2008). The reduction of breast pHu due to enzymes supplementation which increased the glycogen storage in breast meat (Le-Bihan-Duval *et al*., 2008), and ability of breast meat to possessing might be increased with increasing water holding capacity and glycogen (Barbut, 1993; and Zhang and Savage, 2010).

CONCLUSION

According to the obtained results of this study, it could be conclude that the combination of xylanase and pectinase is more effective than the individual enzyme supplementation on growth performance, carcass traits, meat quality when added to standard or reduced ME corn soybean diet. So it could be recommended adding xylanase at 16000 U/kg and pectinase at level of 1000 U/kg diet to either control or NC100 corn-soybean meal broiler diets to enhance the quality of produced broilers accompanied with saving in feeding cost

Table (1): Composition and calculated analysis of experimental diets (EXP1 and EXP2).

Ingredient	Starter (1-10 days)			Grower (11-22 days)			Finisher (23-40 days)		
	control	NC100	NC150	Control	NC100	NC150	control	NC100	NC150
Yellow corn	55.10	55.00	54.90	60.65	62.31	62.55	64.74	66.51	65.7
Soybean meal (44%)	31.00	33.57	36.17	23.84	24.44	25.86	19.25	19.48	23.09
Corn gluten meal (60%)	7.50	5.88	4.00	8.57	8.00	7.00	8.16	8.0	5.30
Soybean oil (SO)	1.80	0.95	0.45	2.27	0.60	0.00	3.28	1.44	1.44
Di-Ca-P	2.00	2.00	2.00	1.75	1.75	1.75	1.55	1.55	1.55
Limestone	1.15	1.20	1.15	1.46	1.46	1.46	1.58	1.58	1.58
NaCl	0.24	0.24	0.24	0.24	0.24	0.24	0.24	0.24	0.24
Vit.&Min. pre-mix*	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40
Sodium bicarbonate	0.23	0.23	0.23	0.23	0.23	0.23	0.23	0.23	0.23
DL Methionine	0.24	0.26	0.27	0.19	0.19	0.19	0.17	0.17	0.19
L-Lysine HCl	0.34	0.27	0.19	0.40	0.38	0.32	0.40	0.40	0.28
Total	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0
Calculated analysis									
Crude protein%	23.00	23.00	23.00	21.00	21.00	21.00	19.00	19.00	19.00
ME (kcal/kg diet)	3000	2904	2850	3093	3000	2950	3200	3100	3050
Crude fiber%	3.68	3.81	3.99	3.32	3.38	3.49	3.06	3.06	3.30
Ether extract%	4.52	3.97	2.97	5.05	3.52	2.92	6.21	4.37	4.21
Calcium %	0.98	0.97	0.98	1.00	1.00	1.00	1.00	1.00	1.00
Available P %	0.52	0.52	0.53	0.47	0.46	0.47	0.42	0.42	0.42
Sodium %	0.17	0.17	0.17	0.17	0.17	0.17	0.17	0.17	0.17
Lysine %	1.38	1.38	1.38	1.24	1.26	1.24	1.11	1.11	1.11
Methionine %	0.67	0.68	0.67	0.60	0.60	0.60	0.55	0.55	0.55
Methionine+Cystine%	1.05	1.05	1.05	0.95	0.96	0.95	0.87	0.87	0.87

* Vitamins and minerals premix will provide each kg of diet with: Vit. A, 11000 IU; Vit. D3, 5000 IU; Vit. E, 50 mg; Vit K3, 3mg; Vit. B1, 2mg; Vit. B2 6mg; B6 3 mg; B12, 14 mcg; Nicotinic acid 60 mg; Folic acid 1.75 mg, Pantothenic acid 13mg; Biotine 120 mcg ; Choline 600 mg; Copper 16mg; Iron 40mg; Manganese 120 mg; Zinc 100mg; Idoine 1.25mg; and Selenium 0.3 mg.

Table (2): Effect of supplemental xylanase and pectinase enzymes on growth performance of broiler chicks at 40 days of age (EXP1).

Treatment	Final weight (g)	Feed intake (g)	Body weight gain (g)	Feed conversion Ratio
Control	1910.50[b]	3279.88[a]	1870.50 [b]	1.75[a]
Xyl.	1970.00[a]	3135.67[b]	1930.00 [a]	1.62[b]
Pec.	1955.50 [ab]	3154.13 [b]	1915.50 [ab]	1.64 [b]
Xyl+Pec	2000.00 [a]	3091.12 [b]	1960.00[a]	1.58[c]
Mean of SE	±17.49	±21.64	±17.49	±0.02
Probability	0.02	0.0003	0.02	0.0001

a,b,...= Means in the same column with different superscripts, differ significantly (P<0.05)
Xyl. = 16000U Xylanase/kg diet Pec. = 1000U Pectinase/kg diet.

Table (3): Effect of supplemental xylanase and pectinase enzymes on carcass characteristics of broiler chicks at 40 days of age (EXP1).

Treatment	Carcass			Front quarter (FQ)		Back quarter (BQ)				
							Thigh		Drums stick (DS)	
	Dressing (% from live weight)	Edible Parts (% from carcass)	Abdominal fat (% from carcass)	Breast weight (% from carcass)	Skin (% from Breast)	Total weight (% from carcass)	Weight (% from BQ)	Skin (% from Thigh)	weight (% from BQ)	Skin (% from DS)
Control	71.48b	5.63	1.15	16.40b	7.69	17.49	61.34	6.69 ab	38.66	8.80
Xyl.	73.82a	5.34	1.01	20.48 a	8.29	18.74	64.08	8.07 a	35.92	8.28
Pec.	72.01b	6.41	1.26	18.44 ab	6.52	17.32	61.71	6.13 b	38.29	8.58
Xyl+Pec	74.81a	5.69	1.14	18.82 ab	6.77	18.17	61.47	7.79 a	38.53	8.22
Mean of SE	±0.39	±0.32	±0.20	±0.79	±1.24	±0.42	±1.71	±0.44	±1.71	±1.17
Probability	0.0002	NS	NS	0.04	NS	NS	NS	0.04	NS	NS

a,b,...= Means in the same column with different superscripts, differ significantly (P<0.05)
Xyl. = 16000U Xylanase/kg diet Pec. = 1000U Pectinase/kg diet

Table (4): Effect of supplemental xylanase and pectinase enzymes on physical and chemical meat quality of broiler chicks at 40 days of age (EXP1).

Treatment	Physical measurements						Chemical measurements			
	Breast		Thigh		Drum Stick					
	Drip Loss	PH_u	Drip Loss	PH_u	Drip Loss	PH_u	MDA	T.P	LDL	HDL
Control	2.59^{ab}	7.36	2.06^{b}	6.27	0.59^{b}	6.44	477.93	47.11^{b}	1419.52	691.43
Xyl.	3.63^{ab}	7.55	4.31^{a}	6.44	1.51^{a}	6.42	415.96	52.65^{b}	1309.52	614.60
Pec.	2.49^{b}	7.71	3.40^{ab}	6.33	1.50^{a}	6.34	423.94	50.98^{b}	1282.33	693.76
Xyl+Pec	4.11^{a}	6.79	3.12^{ab}	6.24	1.12^{ab}	6.36	415.96	60.88^{a}	1230.95	672.22
Mean of SE	±0.38	±0.31	±0.45	±0.09	±0.22	±0.07	±42.07	±2.46	±59.20	±27.25
Probability	0.04	NS	0.04	NS	0.05	NS	NS	0.02	NS	NS

a,b,...= Means in the same column with different superscripts, differ significantly ($P<0.05$) Xyl. = 16000U Xylanase/kg diet
Pec. = 1000U Pectinase/kg diet

Table (5): Effect of xylanase and pectinase enzymes supplementation to low ME diets on growth performance of broiler chicks at 40 days of age (EXP2).

Treatments	Final weight (g)	Feed intake (g)	Body weight gain (g)	Feed conversion Ratio
Main effect		**- ME Reduction**		
NC100	1953.09 [a]	3335.59 [b]	1913.09 [a]	1.74 [b]
NC 150	1924.20 [b]	3466.84 [a]	1884.20 [b]	1.84 [a]
Mean of SE	±9.79	±15.30	±9.79	±0.01
Probability	0.05	0.0001	0.05	0.0001
		-Enzyme supplementation		
Free	1853.61[c]	3417.80	1813.61 [c]	1.89 [a]
With X	1946.64 [b]	3371.29	1906.64 [b]	1.77 [b]
With P	1944.98 [b]	3404.39	1904.98 [b]	1.79[b]
With XP	2009.34 [a]	3411.38	1969.34 [a]	1.73 [c]
Mean of SE	±13.85	±21.63	±13.85	±0.1
Probability	0.0001	NS	0.0001	0.0001
		- Treatments		
Control	1900.50[c]	3289.88[d]	1860.50 [c]	1.77[bcd]
NC 100	1932.22[bc]	3367.04[bcd]	1892.22[bc]	1.78[bc]
NC 100X	1959.03[bc]	3303.75[d]	1919.03[bc]	1.72 [de]
NC 100P	1929.93 [c]	3346.04 [cd]	1889.93 [c]	1.77[bcd]
NC 100XP	1991.18 [ab]	3325.51 [d]	1951.18 [ab]	1.71 [e]
NC 150	1775.00 [d]	3468.55 [ab]	1735.00 [d]	2.00 [a]
NC 150X	1934.26[bc]	3438.83[abc]	1894.26[bc]	1.81 [b]
NC 150P	1960.03[bc]	3462.74 [ab]	1920.03[bc]	1.80[bc]
NC 150XP	2027.50 [a]	3497.25 [a]	1987.50 [a]	1.76 [cd]
Mean of SE	±18.76	±35.83	±18.76	±0.02
Probability	0.0001	0.009	0.0001	0.0001

a,b,...= Means in the same column with different superscripts, differ significantly (P<0.05) NC100 = negative control lower 100 kcal of ME than control NC150 = negative control lower 150 kcal of ME than control Free = without supplementation. X = Xylenase P=pectinase control = standard requirement

Table (6): Effect of xylenase and pectinease on Carcass characteristics of broiler chicks at 40 days of age (EXP2).

Treatments	Dressing%	Edible Parts%	Abdominal fat%	Breast		Back quarter weight%	Thigh		Drums stick (DS)	
				weight%	Skin%		Weight%	Skin%	weight%	Skin%
Main effect				**- ME Reduction**						
NC 100	72.27	5.45 ^{a}	1.02	19.23	5.24	17.83	57.59	6.38	42.41	10.66
NC 150	72.58	5.10 ^{b}	0.98	19.10	5.24	18.46	57.10	6.70	42.90	10.22
Mean of SE	±0.34	±0.11	±0.08	±0.27	±0.29	±0.23	±0.79	±0.48	±0.79	±0.45
Probability	NS	0.03	NS	NS	NS	NS	NS	NS	NS	NS
- Enzymes Supplementation										
Free	71.00^{b}	5.36	0.84 ^{b}	19.43	5.70	17.82	58.33	6.86	41.67	9.83
With X	73.57 ^{a}	5.06	1.34 ^{a}	18.84	5.10	17.65	57.00	5.90	43.00	11.59
With P	71.65 ^{b}	5.36	0.80 ^{b}	19.49	4.58	18.64	56.51	6.93	43.49	10.57
With XP	73.48 ^{a}	5.31	1.01 ^{ab}	18.89	5.59	18.49	57.54	6.40	42.46	9.79
Mean of SE	±0.48	±0.15	±0.11	±0.38	±0.41	±0.33	±1.12	±0.68	±1.12	±0.64
Probability	0.001	NS	0.009	NS	NS	NS	NS	NS	NS	NS
- Treatments										
Control	72.06^{abc}	5.08	1.45 ^{ab}	17.06	6.89	16.99 ^{c}	56.68	7.16	37.79	9.30
NC 100	71.75^{abc}	5.25	0.79 ^{c}	19.92	6.39	17.11^{bc}	59.88	7.08	40.12	11.13
NC 100X	73.23 ^{ab}	5.24	1.65 ^{a}	18.74	4.44	17.68^{abc}	56.52	5.46	43.48	11.64
NC 100P	71.24^{bc}	5.60	0.63 ^{c}	19.66	4.57	18.66 ^{a}	56.49	6.89	43.51	11.33
NC 100XP	72.85^{bc}	5.71	1.01^{bc}	18.60	5.55	17.89^{abc}	57.45	6.08	42.55	8.55
NC 150	70.24 ^{c}	5.47	0.88 ^{c}	18.94	5.00	18.54 ^{ab}	56.78	6.65	43.22	8.52
NC 150X	73.91 ^{a}	4.87	1.03^{bc}	18.95	5.76	17.62^{abc}	57.48	6.35	42.52	11.53
NC 150P	72.06^{abc}	5.12	0.96^{bc}	19.32	4.58	18.62 ^{a}	56.53	6.96	43.47	9.81
NC 150XP	74.10 ^{a}	4.91	1.02^{bc}	19.18	5.63	19.09 ^{a}	57.62	6.72	42.38	11.03
Mean of SE	±0.65	±0.21	±0.16	±0.57	±0.67	±0.44	±1.65	±0.91	±1.65	±0.90
Probability	0.005	NS	0.003	NS	NS	0.03	NS	NS	NS	NS

a,b,...= Means in the same column with different superscripts, differ significantly (P<0.05) NC100 = negative control lower 100 kcal of ME than control
NC150 = negative control lower 150 kcal of ME than control Free = without supplementation. X = Xylenase P=pectinase control = standard requirement

Table(7): Effect ofxylenase and pectinease on physical and chemical meat quality of broiler chicks at 40 days of age (EXP2).

Treatments	Breast		Thigh		Drum Stick (DS)		MDA (nmol/100 g meat)	T.P (mg/100g meat)	LDL (mg/100g meat)	HDL (mg/100g meat)
	Drip Loss	pH_u	Drip Loss	pH_u	Drip Loss	pH_u				
Main effect					**- ME Reduction**					
NC 100	3.29 ^{b}	5.81	4.37	6.43	0.79 ^{b}	6.47	340.49 ^{a}	75.71 ^{a}	971.67	690.25
NC 150	4.98 ^{a}	6.02	4.74	6.44	1.12 ^{a}	6.52	303.64 ^{b}	80.11 ^{b}	969.05	724.78
Mean of SE	±0.20	±0.16	±0.30	±0.06	±0.09	±0.03	±8.69	±1.04	±25.14	±16.56
Probability	0.0001	NS	NS	NS	0.02	NS	0.008	0.009	NS	NS
					- Enzymes Supplementation					
Free	3.31 ^{b}	5.62	2.85 ^{b}	6.60 ^{a}	1.29 ^{a}	6.43	335.92	76.23	966.43	666.42
With X	3.82 ^{b}	5.97	5.55 ^{a}	6.32 ^{b}	0.99 ^{ab}	6.58	336.86	78.31	955.95	740.05
With P	3.74 ^{b}	5.74	4.95 ^{a}	6.48 ^{ab}	0.75 ^{b}	6.52	310.33	79.44	987.38	720.19
With XP	5.66^{a}	6.33	4.86 ^{a}	6.34 ^{b}	0.79 ^{b}	6.45	305.17	77.67	971.67	703.19
Mean of SE	±0.28	±0.22	±0.42	±0.08	±0.13	±0.04	±12.28	±1.47	±35.55	±23.41
Probability	0.0001	NS	0.002	0.08	0.04	NS	NS	NS	NS	NS
					Treatments					
Control	3.06 ^{cd}	6.86 ^{a}	2.89 ^{cd}	6.67	0.87^{bc}	6.54	465.27^{a}	52.44^{c}	1407.86 ^{a}	681.76
NC 100	3.23 ^{cd}	5.67 ^{b}	1.89 ^{d}	6.52	1.03 ^{ab}	6.47	364.32 ^{b}	74.90^{b}	1005.71 ^{b}	640.44
NC 100X	2.44 ^{d}	6.27 ^{ab}	6.02 ^{a}	6.39	1.06 ^{ab}	6.54	358.22^{bc}	75.20 ^{b}	885.24 ^{b}	727.83
NC 100P	3.57 ^{cd}	5.67 ^{b}	5.39 ^{ab}	6.49	0.69^{bc}	6.49	318.78^{bcd}	76.47^{ab}	1010.95 ^{b}	687.50
NC 100XP	3.92 ^{c}	5.65 ^{b}	4.18^{abc}	6.33	0.38 ^{c}	6.39	320.66^{bcd}	76.27 ^{ab}	984.76 ^{b}	705.22
NC 150	3.39 ^{cd}	5.58 ^{b}	3.82^{bc}	6.68	1.54 ^{a}	6.39	307.51^{bcd}	77.55 ^{ab}	927.14 ^{b}	692.39
NC 150X	5.20 ^{b}	5.67 ^{b}	5.09 ^{ab}	6.26	0.93^{bc}	6.62	315.49^{bcd}	81.42 ^{ab}	1026.67 ^{b}	752.28
NC 150P	3.92 ^{c}	5.82 ^{b}	4.52^{abc}	6.46	0.82^{bc}	6.54	301.88 ^{cd}	82.40 ^{a}	963.81 ^{b}	752.89
NC 150XP	7.41 ^{a}	7.02 ^{as}	5.54 ^{ab}	6.35	1.21 ^{ab}	6.52	289.67^{d}	79.07 ^{ab}	958.57 ^{b}	701.55
Mean of SE	±0.41	±0.30	±0.59	±0.11	±0.18	±0.06	±17.43	±2.03	±52.21	±32.41
Probability	0.0001	0.01	0.002	NS	0.02	NS	0.0001	0.0001	0.0001	NS

a,b,...= Means in the same column with different superscripts, differ significantly (P<0.05) NC100 = negative control lower 100 kcal of ME than control. NC150 = negative control lower 150 kcal of ME than control. Free = without supplementation. X = Xylenase. P=pectinase. control = standard requirement.

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الملخص العربى

دراسة علي التأثير التكاملي بين نوعين من إنزيمات تحليل الكربوهيدرات المضافه لعلائق دجاج التسمين القياسية و المنخضة في الطاقة

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تهدف هذه الدراسة إلي دراسة التأثير التكاملي بين إنزيمي الزيلينيز والبكتينيز وذلك عند أضافتهم لعلائق دجاج التسمين (الذرة والصويا) المختلفة في مستوي الطاقة . وشملت الدراسة تجربتين، درست التجربة الأولي إضافة الزيلينيز أو البكتينيز أو كلاهما إلي علائق الذرة والصويا القياسية، أختبرت التجربة الثانية إضافة الأنزيمات إلي علائق منخفضة في محتواها من الطاقة (أقل 100 كيلو كالوري/كجم و 150 كيلو كالوري/كجم) عن عليقة المقارنة، وتمت دراسة الأداء الإنتاجي وخصائص الذبيحة وجودة اللحم. وكانت النتائج كالتالي: أظهرت النتائج في التجربة الأولي أن إضافة أنزيم الزيلينيز أو البكتينيز أو كلاهما إلي علائق الدواجن أدت إلي تحسن معنوي في وزن الجسم والزياده في وزن الجسم و ومعامل التحويل الغذائي والنسبة المئوية لوزن الصدر وتقليل الغذاء المأكول بينما اضافة مخلوط الإنزيمين معا أدي إلي تحسن النسبة المئويه للذبيحه وايضاً حسن البروتين الكلي للحم في حين أن الإنزيمات الفردية ليس لها تأثير. في التجربة الثانية أدي نقص مستوي الطاقة بمعدل 150 كيلو كالوري/كجم إلي نقص الوزن النهائي للجسم ووزن الجسم المكتسب ونسبة الأجزاء المأكولة بالذبيحة والمالونالدهيد والبروتين الكلي للحم وأدي إلي زيادة معدل الغذاء المأكول ومعدل الفقد في الماء المحتجز للحم الصدر والدبوس في لحم الدجاج، إضافة مزيج من إنزيم الزيلينيز وإنزيم البكتينيز إلي علائق منخفضة الطاقة بمعدل 100 كيلو كالوري/كجم أدي إلي تحسن معنوي في الأداء الإنتاجي بصورة أكبر من إضافة الأنزيمات الفردية، في حين أن أفضل معامل تحويل غذائي كان للمجموعة المغذاة علي علائق منخفضة في الطاقة بمعدل 100 كيلو كالوري/كجم ومضاف إليها الزيلينيز والبكتينيز، سجلت المجموعة المغذاة علي العليقة المنخفضة في الطاقة بمعدل 150 كيلو كالوري/كجم والمضاف إليها الزيلينيز والبكتينيز أفضل وزن كلي للذبيحة ونسبة الذبيحة مقارنة مع المعاملات الأخري، وقد سجلت المجاميع المغذاة علي علائق منخفضة في الطاقة (100 كيلو كالوري/كجم و 150 كيلو كالوري/كجم) أقل معدل دهن بطن. نستخلص أن إضافة الخليط من إنزيمي الزيلينيز والبكتينيزله تأثير أكبر من إضافة الإنزيمات منفرده الي العليقه علي الأداء الإنتاجي وجودة اللحم وإعطاء ذبيحة أفضل.