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Different Dietary Source of Non-Starch Polysaccharides Supplemented with Enzymes Affected Growth and Carcass Traits, Blood Parameters and Gut Physicochemical Properties of Broilers

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ABSTRACT

An experiment was conducted to evaluate the effect of different dietary source of Non-starch polysaccharides supplemented with enzymes on growth, carcass and physiological characteristics of broilers. A total number of 625 unsexed broiler chicken (Ross 308) were randomly assigned to 5 treatments, 5 replicates and 25 birds per each unit, using a CRD statistical design. Treatments were included control, wheat (W), wheat+ enzyme (WE), barley (B), and barley+ enzyme (BE). Total feed intake and body weight gain were significantly increased, but feed conversion ratio decreased by diets supplemented with enzymes rather diets without enzymes ($P<0.05$). The inclusion of W and B in diets led to significantly decreased in carcass yield and the percentage of breast, leg, fat pad, and liver compare to the control ($P<0.05$). The percentage of pancreas was higher in W and B diets than control or enzyme supplemented diets ($P<0.05$). Serum levels of glucose (GLU), triglyceride (TG), and cholesterol (CHO) significantly decreased in diet contained W and B ($P<0.05$). Digesta viscosity significantly increased, but pH decreased after feeding the chickens by W and B diets rather control ($P<0.05$). The results of present study have shown that supplementation of W and B with multi-enzymes completely restored the situation and neutralized the negative effects of W and B on growth and carcass traits, blood parameters and gut physicochemical properties of broiler chickens.

Keywords: chicken, carcass, growth, NSP, physicochemical.

INTRODUCTION

The two major corn-producing countries, United State and Brazil, have been recently shifted using of corn to ethanol fuel producing. This diversion along with increased world's demand for this cereal has resulted in rise of corn price and consequently feed cost for poultry industry (Donohue and Cunningham, 2009). Although wheat and barley are common cereals used in many countries due to close energy level to corn, capability of grow on native lands with lower water requirement and meanwhile reduce feed costs (Ahmadi *et al.*, 2013), but their high levels of xylans and β-glucans, the principal water-soluble non-starch polysaccharides (NSP), limits their use in poultry feeding (Kalantar *et al.,* 2014). The presence of xylans and β-glucans increases the viscosity of the gut content, impeding the digestion and absorption of nutrients and causing low performance (Brenes *et al.,* 1993; Choct *et al.,* 1997; 2006).

Wheat and barley can locally grow in most parts of the world as well as they have remarkably lower water requirement than corn (Yin *et al.,* 2000; Lin *et al.,* 2010). These cereals as alternative crops can be easily replaced for corn in poultry diets, if truly supplemented with exogenous enzymes known as carbohydrases or multi-glycanase (Selle *et al.,* 2003; Mirzaie *et al.,* 2011; 2012).

Increase feeding levels of NSPs in diets directly influences on growth response and carcass traits as well as affected other physicochemical properties of birds such as digesta viscosity and pH (Silva and Smithard, 1996; Jaroni *et al.,* 1999; Olukosi *et al.,* 2007). Reports indicate that a complex blend of NSPs degrading enzymes requires obtaining satisfactory gain and other performances (Ravindran *et al.,* 1999; Slominski, 2011). In our previous study, effect of different dietary source of Non-starch polysaccharides supplemented with enzyme on growth, gut microbial and serum enzyme activity has demonstrated.

In the present study, equal amount of wheat and barley and approximately similar fractions of NSP content from wheat and barley were included in broiler diets with and without multi-enzyme to compare the effects on growth and carcass traits, blood parameters and gut physicochemical properties of broilers.

MATERIAL AND METHODS

The Animal Ethics Committee of the Shahrekord University, Shahrekord-Iran approved all procedure of the experiment.

Experimental Design and Birds

Through a Completely Randomized Design (CRD) 625 unsexed broiler chicken (Ross-308) were randomly divided to 5 treatments with 5 replicates of 25 birds in each. Treatments were included of control (corn-soy basal diet), and the inclusion of wheat (W), wheat+ enzyme (WE), barley (B), and barley+ enzyme (BE) at levels of 15 and 20 percent in starter and grower periods respectively. Diets were designed in two phases (starter and grower) based on NRC (1994) recommendations (Table 1). Combo® multi-enzyme was used contained 1000 unit phytase and 180 unit multi-glycanase activities per each gram.

Growth and Carcass Traits

Feed and water offered *ad libitum* in all period of experiment. Total body weight gain (TBWG), total feed intake (TFI), and total feed conversion ratio (TFCR) were measured at the end of period based on weekly recorded weights of feed intake and body weight gain of chickens. The lighting schedule was 23 h light per 1 h darkness at 32°C in the first day. This was subsequently reduced 3°C each week until week five.

On 42 days of age, 3 birds from each replicate were randomly selected, tagged, and were fasted for 8h (no limitation of water access). Birds were weighted and slaughtered by serving both of the right and left carotid artery and jugular vein in a single cut and bled for 180s. Carcass weights were measured after removal of feather, head, legs and abdominal contents. The empty body weight, breast, legs, and abdominal fat pad were recorded and calculated as the percentage to the carcass weight. In addition, liver and pancreas weights as well as, intestine length were recorded according to the method reported by the Rezaei *et al.,* (2014). Means of 3 observations were considered to statistical analysis.

Table 1: Diet composition at different periods of the experiment

Diets	experimental diets during 1-21days					experimental diets during 22-42days				
Ingredients(%)/Treatment	C[1]	W[2]	B[3]	WE[4]	BE[5]	C[1]	W[2]	B[3]	WE[4]	BE[5]
Corn grain	56	44.55	45	44.55	45	58	40	42.41	40	42.41
Soybean meal (45% CP/kg)	36.8	35.1	33.9	35.1	33.9	32	30.5	29.6	30.5	29.6
Soybean oil	2	1.35	2	1.35	2	2.9	2.85	3.47	2.85	3.47
Wheat	-	15	-	15	-	-	20	-	20	-
Barley	-	-	15	-	15	-	-	20	-	20
Enzyme[6]	-	-	-	+	+	-	-	-	+	+
Dicalcium phosphate	1.83	1.78	1.78	1.78	1.78	1.81	1.74	1.71	1.74	1.71
Oyster shell	1.12	1.14	1	1.14	1	1.13	1.14	1.13	1.14	1.13
Sodium chloride	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3
Potassium carbonate	0.1	0.13	0.13	0.13	0.13	0.12	0.12	0.11	0.12	0.11
DL-Methionine	0.17	0.15	0.15	0.15	0.15	0.25	0.25	0.05	0.25	0.05
L-Lysine HCL	0.1	-	0.1	-	0.1	0.15	0.1	0.05	0.1	0.05
Premix[7]	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Inert	1.08	-	0.14	-	0.14	2.84	2.5	0.67	2.5	0.67
Total	100	100	100	100	100	100	100	100	100	100
Calculated Analysis										
Metabolizable energy (kcal/kg)	2900	2900	2900	2900	2900	2950	2950	2950	2950	2950
Crude protein	21	21	21	21	21	19	19	19	19	19
Met + Cys	0.86	0.84	0.82	0.84	0.82	0.85	0.85	0.84	0.85	0.84
Lysine	1.2	1.19	1.18	1.19	1.18	1.2	1.19	1.11	1.19	1.11
Calcium	0.95	0.94	0.92	0.94	0.92	0.95	0.95	0.87	0.95	0.87
Available phosphorus	0.43	0.43	0.45	0.43	0.45	0.45	0.42	0.43	0.42	0.43
Sodium	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Chloride	0.22	0.23	0.24	0.23	0.24	0.22	0.23	0.23	0.23	0.23
Potassium	0.95	0.95	0.96	0.95	0.96	0.87	0.87	0.87	0.87	0.87
DEB=(Na+K)-Cl (meq/kg)[8]	247.8	247.9	247.8	247.9	247.8	231.2	231.6	231.5	231.6	231.5
Total NSP	12.43	12.89	12.96	12.89	12.96	11.62	12.11	12.65	12.11	12.65

[1]Control, [2]Wheat [3]Wheat+enzyme, [4]Barley,and [5]Barley+Enzyme.[6]Enzymes contained 1000 unit Phytase and 180 unit Multi-glycanase and added at level 0.1% on top of ingredients in enzyme supplemented diets. [7]Supplied the following per kilogram of diet: vitamin A 44,000 IU, vitamin D3 17,000 IU, vitamin E 440 mg, vitamin K3 40 mg, vitamin B12 70 mg, vitamin B1 65 mg, vitamin B2 32 mg, Pantothenic acid 49 mg, Niacin 122 mg, vitamin B6 65 mg, Biotin 22 mg, Choline Chloride 27 mg: 650 mg of Mn, 250 mg of Zn, 125 mg of Fe, 110 mg of Cu, 60 mg of Se, 68 mg of I, and 21 mg of Co. [8]DEB: Dietary Electrolyte Balance.

Gut Physicochemical Properties

At the end of trial (day 42), 3 additional chicks per pen were randomly selected and killed to obtain samples for the measurement of intestinal pH and viscosity. Whole ileal digesta were individually collected, homogenized at 4^{o}C and immediately measured for pH and viscosity using digital pH meter (Lutron, model pH-201, CE, Taiwan) and viscometer (Brookfield, model DV-II, MA, USA) respectively, according to described method of Langhout *et al.*, (1999). Means of 3 observations were considered to statistical analysis.

Statistical Analyses

All data were analyzed for normal distribution using the NORMAL option of the UNIVARIATE procedure of GLM procedure of SAS software (SAS Inst. Inc., Cary, NC). A completely randomized design was employed. Pen was used as the experimental unit and data were analyzed by GLM procedure. Duncan's multiple range test were used for comparison of means ($P<0.05$).

RESULTS

The effect of dietary treatments on chicken growth response is shown at Table 2. The results indicated that the diets contained W or B led to significantly decrease in TFI and TBWG as well as significantly increase in TFCR rather other diets ($P<0.05$). Enzyme supplemented W and B diets with multi-enzymes resulted in significantly increase in TFI and TBWG as well as significantly decrease in TFCR rather than the same diets without enzymes ($P<0.05$).

Table 2: Effect of different sources of NSP on growth response of chickens at 42 day of age

Treatment	TFI[1] (g/ per bird)	TBWG[2] (g/ per bird)	TFCR[3]
Control	3893.40 ^{a}	2235.66 ^{a}	1.74 ^{b}
W[4]	3680.46 ^{b}	1970.64 ^{b}	1.87 ^{a}
WE[5]	3868.20 ^{a}	2158.80 ^{a}	1.79 ^{b}
B[6]	3688.86 ^{b}	1974.42 ^{b}	1.87 ^{a}
BE[7]	3871.14 ^{a}	2155.02 ^{a}	1.79 ^{b}
SEM	62.51	64.32	0.05
P-value	0.037	0.041	0.016

Means with common letters in the same column are not significantly different (P<0.05). SEM: Standard error of the means. [1]Total Feed intake, [2] Total Body weight gain, [3] Total Feed conversion ratio, [4]Wheat, [5]Wheat+enzyme, [6]Barley,and [7]Barley+Enzyme.

Table 3 showed the effect of diets on chicken carcass characteristics at 42 days of age. Diets contained W or B caused to significantly decreases in carcass yield and percentages of breast, leg and fat pad of chickens at 42 days of age (P<0.05). Liver percentage significantly decreased, but inversely, pancreas percentage significantly increased by feeding the W and B diets to chickens at 42 days of age (P<0.05). Intestine length didn't show significant differences through experimental diets.

Table 3: Effect of different sources of NSP on carcass characteristics of chickens at 42 day of age

Treatment	Empty Carcass (%)	Breast (%)	Leg (%)	Fat pad (%)	Liver (%)	Pancreas (%)	Intestine length (cm)
Control	65.92 ^{a}	35.56 ^{a}	33.67 ^{a}	1.26 ^{a}	2.38 ^{a}	0.07 ^{b}	85.20
W[1]	62.66 ^{c}	33.35 ^{b}	31.19 ^{b}	1.06 ^{b}	2.05 ^{b}	0.12 ^{a}	88.80
WE[2]	63.92 ^{bc}	34.66 ^{a}	32.30 ^{ab}	1.15 ^{ab}	2.15 ^{ab}	0.08 ^{b}	87.40
B[3]	64.51^{b}	33.42 ^{b}	31.15 ^{b}	0.87 ^{c}	2.09 ^{b}	0.15 ^{a}	86.90
BE[4]	65.08^{a}	34.54 ^{a}	32.29 ^{a}	1.18 ^{ab}	2.13 ^{ab}	0.09 ^{b}	88.60
SEM	1.13	1.24	1.05	0.08	0.11	0.02	3.30
P-value	0.045	0.031	0.033	0.022	0.050	0.001	0.242

Means with common letters in the same column are not significantly different (P<0.05). SEM: Standard error of the means. [1]Wheat, [2]Wheat+enzyme, [3]Barley,and [4]Barley+Enzyme.

The effects of diets on the serum levels of GLC, TG, and CHO presented at Table 4. The inclusion of W and B in diets led to significantly decreases in serum levels of GLC, TG, and CHO rather control (P<0.05). W and B diets which supplemented with exogenous enzymes led to significantly increases in serum levels of GLC, TG, and CHO rather the same diets without enzymes (P<0.05).

Table 4: Effect of different sources of NSP on blood parameters of chickens at 42 day of age

Treatments	GLU[1] (mmol/L)	TG[2] (mg/dl)	CHO[3] (mg/dl)
Control	15.11 ^{a}	131.16 ^{a}	134.83 ^{a}
W[4]	12.31 ^{b}	116.63 ^{c}	116.14 ^{c}
WE[5]	14.32 ^{a}	121.25 ^{b}	125.50 ^{b}
B[6]	11.58 ^{b}	115.45 ^{c}	114.11 ^{c}
BE[7]	14.21 ^{a}	119.32 ^{b}	122.33 ^{b}
SEM	0.48	1.24	1.54
P-value	0.003	0.025	0.031

Means with common letters in the same column are not significantly different (P<0.05). SEM: Standard error of the means. [1]Glucose, [2]Triglyceride, [3]Cholesterol, [4]Wheat, [5]Wheat+enzyme, [6]Barley,and [7]Barley+Enzyme.

In table 5, the effects of diets on digesta pH and viscosity were presented. The inclusion of W and B in diets led to significantly decreases in digesta pH, but inversely, significantly increased digesta viscosity rather control (P<0.05). Supplemented W and B diets with exogenous multi-enzyme led to significantly increases in pH, but inversely, significantly decreased digesta viscosity rather the same diets without enzymes (P<0.05).

Table 5: Effect of different sources of NSP on gut physicochemical properties of chickens at 42 days of age

Treatments	pH	Viscosity (cP) [1]
Control	6.89 [a]	1.59 [c]
W[2]	5.93 [b]	2.17 [a]
WE[3]	6.39 [a]	1.60 [b]
B[4]	5.67 [b]	1.95 [a]
BE[5]	6.41 [a]	1.60 [b]
SEM	0.16	0.04
P-value	0.011	0.003

Means with common letters in the same column are not significantly different (P<0.05). SEM: Standard error of the means. [1]cP : centipoise, [2]Wheat, [3]Wheat+enzyme, [4]Barley,and [5]Barley+Enzyme.

DISCUSSION

According to the results presented in table 2, birds fed on W or B diet consumed lower feed intake, consequently they had lower BWG compared to other diets. Soluble NSPs of wheat and barley including xylans and β-glucans, the principal water-soluble non-starch polysaccharides (NSP), limits digestion of nutrients through the gut, prevents the normal motion of digesta and mixing to endogenous enzymes and conduces to negative effects on feed intake and broiler growth performance (Yin *et al.*, 2000; Lin *et al.*, 2010; Mirzaie *et al.*, 2012). The presence of xylans and β-glucans increases the digesta viscosity, motivates the gut microbial propagation resulted to produces high amount of short chain fatty acids which lowers digesta pH (Jaroni *et al.*, 1999; Choct *et al.*, 2006), according to the results reported in Table 5.

Several reports indicated that water soluble NSPs have more deleterious impact on physicochemical properties and microbial proliferation of digesta in the intestinal tract. These types of NSPs creates ideal environment for maximum proliferation of bacteria. A slow moving digesta with low oxygen tension could provide a stable media where fermentative microbes such as anaerobic bacteria can establish. These microbial changes result in reduced nutrients available for host and produces of detrimental byproducts (Langhout *et al.,* 1999; Jaroni *et al.,* 1999). Supplementation of W and B diets with multi-enzymes significantly reduced the negative effects of soluble NSPs on digesta viscosity and pH through breakdown of NSP polymers. These findings are in accordance with other reports (Lin *et al.,* 2010; Mirzaie *et al.,* 2011; 2012).

Results reported in table 3, indicated that W and B diets have more deleterious impacts on carcass yield and organ percentages of breast, leg, fat pad as well as organelle percentages of liver and pancreas. Intestinal length of chickens didn't influenced by dietary source of NSP. Birds fed on W or B diet due to presence of soluble NSPs in diets consumed lower feed intake, consequently they had lower BWG and carcass efficiency compared to other diets. The growth and carcass traits are consistent with the viscosity of gut digesta, which negatively affected by soluble NSPs (Silva and Smithard, 1996; Jaroni *et al.,* 1999; Selle *et al.,* 2003). Diets contained W and B caused a significant decrease in liver percentage rather control or enzyme supplemented diets. Decrease in lipid metabolism thereafter reduction of lipid uptake by liver might result in lower liver percentage (Saki *et al.,* 2011; Rezaei *et al.,* 2014). Results showed that high secretive activity of the pancreas due to NSP contents of W and B resulted in significant differences in pancreas percentage compared to other diets. Soluble NSPs impedes enzyme capability to hydrolyze nutrients. This in turn, creates high degree of hyperplasia resulted in higher weight and pancreas percentage compared to other diets (Li *et al.,* 2004; Olukosi *et al.*, 2007; Mirzaie *et al.*, 2011). Supplementation of diets with exogenous enzymes releases the encapsulated nutrients and reduces digesta viscosity. These processes are further facilitated by the action of phytases (Ravindran *et al.*, 1999; Slominski,, 2011).

Results reported in table 4 showed that, birds fed on W or B diet presented lower serum concentrations of GLU, TG and CHO compared to other diets. Research data indicated that changing the blood parameters such as GLU, TG and CHO are proper indices to recognize of bird reaction to consumed diet and to the food components like fiber or NSP (Selle *et al.,* 2003; Saki *et al.,* 2011). Inclusion of NSPs in diets could reduce serum glucose concentration. It can refer to decrease in carbohydrates hydrolysis to simple monosaccharide due to impede enzyme capability to hydrolyze nutrients (Jaroni *et al.,* 1999; Lin *et al.,* 2010). Another probable mechanism is strong anti-oxidant action of NSPs components in diets contained W and B (Saki *et al.,* 2011; Rezaei *et al.,* 2014). The use of diets with fibrous structure in broiler feeding can induce lowering effect on serum TG and CHO concentrations (hypotriglyceridemia and hypocholesteridemia) in broilers fed on W and B. This effect refers to a few mechanisms such as decrease in lipid absorption through the gut surface posterior blocking the bile salts by NSP lattice and lowering the lipid metabolism thereafter reduction in lipid uptake (Yin *et al.,* 2010; Rezaei *et al.,* 2014). Enzyme supplementation of W and B diets modulates these changes. These observations were in line with other reports (Jaroni *et al.,* 1999; Mirzaie *et al.,* 2012; Kalantar *et al.,* 2014).

CONCLUSION

Results of the present study indicated that the adverse effects of W and B diets on broiler growth and carcass yield as well as blood parameters and gut physicochemical properties are due to deleterious effects of NSP content of these cereals. But these conversions can be remarkably relieved by supplementing W and B diets with NSP-degrading multi-enzymes.

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