

## The effects of enzymes supplementation on bio-productive performance, intestinal viscosity, and sanguine indices on broilers fed with wheat and barley based diets

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### Abstract

The purpose of the study was to determine the effect of various types of enzymes, such as mono-enzymes represented by xylanase, beta-glucanase or poly-enzymes complex on productive performances as well as on intestinal viscosity and on some biochemical indices of sanguine indices. This experiment was carried out on a group of 200 chickens distributed in four experimental groups (10 replicate with 5 chickens/cage) as follows: the experimental group EG1, fed with 600g of wheat/kg complete feed and 100 mg/kg of xylanase, the experimental group EG2 fed with 600g of wheat/kg complete feed and 250 mg/kg poly-enzymatic complex, the experimental group EG3, fed with 600g of wheat/kg complete feed and the experimental group EG4, fed with 600g of wheat/kg complete feed and 250 mg/kg of poly-enzymatic complex. Complete feed based on enzymes like wheat and barley increases feed consumption throughout the experimental period. Adding poly-enzymatic complex in complete feed based on barley leads to a higher body weight until 3 weeks, compared to the use of beta glucanase; incorporating poly-enzymatic complex in wheat-based compound leads to a lower body weight until 4 weeks. Feed conversion rate, viscosity in the gut are influenced positively by adding enzymatic complexes in complete feed based on barley and of xylanase in wheat-based compound. Adding of poly-enzymatic complex in complete feed based on wheat increases the triglycerides content and in barley-based complete feed lowers triglycerides and does not change the cholesterol content of the blood serum.

**Key words:** poly-enzymatic complex, beta glucanase, xylanase, NSP, cholesterol, triglycerides

### Introduction

Due to the high energy content, corn grain is considered ideal for bird feed. However, there are cases when the situation requires the use of other grains in amounts up to 60%, such as wheat or barley. Using them in this quantity is only possible by adding enzyme due to the increased non-starch polysaccharides (NSP) content, which in large quantities have an anti-nutritive effect. Usage of enzyme should not be taken randomly but according to the substrate on which they operate. In recipes based on wheat and barley, both mono-enzyme and several enzymes were used. Wheat and barley however contain high amounts of antinutritive carbohydrates that normally impair feed utilization (Aman & Graham, 1987, Campbell *et al.*, 1989; Choct *et al.*, 1996). The water soluble NSP present in barley and wheat increases the intestinal viscosity in the gastrointestinal tract and impedes digestive enzymes to be in contact with substrates (White *et al.*, 1983; Choct & Annison, 1992; Almirall *et al.*, 1995). Beta glucanase hydrolyses glucans and improves digestive use of barley by breaking polymer chains into smaller pieces and reduce the gut viscosity, hence improves the nutritive value of grains rich in NSP (Bedford *et al.*, 1991; Almirall *et al.*, 1995; Yu *et al.*, 1997), (Smits & Annison, 1996). Water soluble glucan in barley causes increased gastrointestinal viscosity in animals and poultry. (Anderson *et al.*, 1990), (White *et al.*, 1983). Anyway there are only few researches on the effect of complex polyenzymatic in broilers.

The purpose of this experiment was to estimate whether different grain types (wheat, barley) and adding of exogenous enzymes influence the performance and the sanguine indices of broiler chickens as regards intestinal viscosity.

## **Materials and methods**

This experiment lasted for a period of six weeks and was carried out on a group of chickens by S.C. Alis Deva, distributed in four experimental groups (EG1, EG2, EG3 and EG4). Four experimental groups were formed as follows: the experimental group EG1, fed with 600g of wheat/kg complete feed and 100 mg/kg of xylanase, the experimental group EG2 fed with 600g of wheat/ kg complete feed and 250 mg/kg poly-enzymatic complex/kg Kemzyme Plus Dry (Endo-1,4-beta-glucanase (complex cellulose), alpha-amylase, Bacillolysine (protease) and Endo-1,4-beta-xylanase), the experimental group EG3, fed with 600 g of barley /kg complete feed and 100 mg/kg beta glucanase, and the experimental group EG4, fed with 600 g of barley/kg complete feed and 250 mg/kg of the same poly-enzymatic complex/kg. At the ages of 3 and 6 weeks, after slaughter, intestinal content samples were taken in order to determine intestinal viscosity. Each experimental group is formed of five replicates, each one consisting of 10 chickens and each experimental lot had similar weights. The birds were kept in poultry arks cages from hatching to 42 days. The feed and water were given ad libitum, following with two stage feeding system (hatching to 3 weeks and 4 to 6 weeks).

### ***Biological material used in experiment***

The hybrids used for meat production in intensive system are tetra-linear biracial, obtained through the simple hybridization of two White Cornish lines (paternal genotype) and two White Plymouth Rock lines (maternal genotype). The experiments were carried out on broiler chickens, the hybrid Ross 308, at the Department of Animal Nutrition and Alimentation, From the Didactic Station of Banat University of Agricultural Sciences and Veterinary Medicine, Timișoara. The experiments were carried according to the ethical norms (Law no. 9/2008).

### ***Nutritive content analysis of the complete feed***

To determine the nutritive value of the complete feed offered to broiler chickens in our experiments, we applied the standard methods according to WEENDE scheme, respectively: Dry matter (DM) (g/kg) – stove-drying at 105 ° C, Crude protein (CP) (g/kg ) – Kjeldahl method, Crude fat (CF) (g/kg ) – Soxhlet method, Crude fibre (CF) (g/kg) – Van Soest method.

### ***Analysis of NSP content of complete feed used during the experiment***

Determination of the NSP of complete feed was made in the University of Dublin laboratory. The four based wheat and barley diets were analyzed for non starch polysaccharides in hatching and 3 weeks old broilers. The procedure is described by Dusel *et al.*, 1997 and is based on the methods described by Englyst, 1989.

### ***Determination of nutritive, bio-productive and digestive indices at broiler chickens***

In this experiment on broiler chickens, the following indices were determined: feed intake, weight gain, feed conversion rate, intestinal viscosity.

To determine the feed intake for each period separately, the amounts allocated for each variant were weighed at the beginning of the experiment, and at 3 weeks old we weighed the amount of feed left. The same method was applied for the period of 3-6 weeks. The weight gain was determined by weighing at hatching, at the age of 3 weeks and at 6 weeks.

### ***Analysis of viscosity at intestinal level***

The method used for determining the viscosity was made by Dusel *et al.*, 1997 and Englyst 1989. For digestive viscosity determination 32 broilers were slaughtered (8 broilers per group) by cervical dislocation, at 42 days old. The birds were dissected duodenum and jejunum segments were quickly tied together in order to prevent post-mortem digestion. For

viscosity analysis approximately 2 g of the fresh digestion was placed in the centrifuge tube and was centrifuged at 10,000 revolutions per minute for 10 minutes. The supernatants were stored on ice until viscosity measurements were made. Viscosity was measured with Brookfield viscosity meter (LD DV II +).

#### *Study of sanguine indices*

A blood sample was collected from a wing vein with a heparinised syringe from 32 birds (8 birds per replicate) per group on age of 42 days. Blood samples were collected on ice, centrifuged and plasma stored at -20<sup>0</sup> C. Measurements were performed using Fully Vet analyzer, which is an automated system for testing blood serum biochemical parameters of 13 species of farm animals. Following determinations were made from blood serum cholesterol and triglycerides.

#### *Result processing methods*

The testing of differences between batches, in terms of production indices, was performed with the help of Mann Whitney (Wilcoxon) test and with MINITAB 14 software as well. The regression equations were established with Data Fit 9 programs.

The experimental design is presented in table 1, and the nutritional characteristics of the complete feed used in this experiment are presented in table 1.

**Table 1.** The experimental design

EG1	EG2	EG3	EG4
<b>Period 0-3 weeks</b>			
Complete Feed with 600 g/kg wheat + xylanase 100 mg/kg	Complete Feed with 600 g/kg wheat + poly-enzymatic complex 250 mg/kg	Complete Feed with 600 g/kg barley + betaglucanase 100 mg/kg	Complete Feed with 600 g/kg barley + poly-enzymatic complex 250 mg/kg
<b>Period 3-6 weeks</b>			
Complete Feed with 600 g/kg wheat + xylanase 100mg/kg	Complete Feed with 600 g/kg wheat + poly-enzymatic complex 250 mg/kg	Complete Feed with 600 g/kg barley + betaglucanase 100 mg/kg	Complete Feed with 600 g/kg barley + poly-enzymatic complex 250 mg/kg

The chickens in the four experimental groups were fed as follows: during the first growing period, namely from hatching to the age of 3 weeks, the complete feed provided 12.52-13,28 MJ metabolizable energy (ME) and a crude protein (CP) content of 215.2-229.7 g/kg. During the second growing period from 3 to 6 weeks, the complete feed provided 12.97-13.49 MJ ME and 189.4-215.2 g/kg CP (table 2).

**Table 2.** The structure and the nutritive characteristics of complete feed used in the experimental groups

Specification	EG1-EG2		EG3 - EG4	
	Period 0-3 weeks	Period 3-6 weeks	Period 0-3 weeks	Period 3-6 weeks
Corn	-	58	-	50
Barley	-	-	600	600
Wheat	600	600	-	-
Soybean meal	280.5	248.4	270.5	246.4
Fish meal	50	20	50	20
Sunflower oil	35	40	45	50
Calcium carbonate	12	12	12	12
Mono calcium phosphate	8	8	8	8

DL Methionine	2	2	2	2
Salt	2.5	1.6	2.5	1.6
Vitamin- mineral premix* + poly- enzymes** or ***xylanase or betaglucanase (g/kg CF)	10	10	10	10
<b>Nutritional characteristics</b>				
M E (MJ/kg complete feed)	13.28	13.49	12.52	12.97
Crude protein (g/kg)	229.7	201.1	215.2	189.4
Total lysine (g/kg)	11.5	9.4	11.8	9.8
Total methionine + cystine (g/kg)	9	7.2	9	7.2
Calcium (g/kg)	10.84	8.9	11.2	9.4
Total Phosphorus (g/kg)	7.26	6.2	6.9	5.8

\*Providing per kilogram of complete feed – retinyl acetate 12042.00 UI; cholecalciferol 3010.50 UI; DL  $\alpha$  –tocopheryl acetate 40.14 mg; menadione sodium bisulphite 3.12 mg, thiamin mononitrate 3.01 mg; riboflavin 7.03 mg; niacin 40.14 mg; calcium pantothenate 12.04 mg; pyridoxine 5.02 mg cyanocobalamin 0.03 mg; biotin 0.12 mg; folic acid 1.00 mg, Fe 22.37 mg; Mn 30.00 mg; Zn 21.74 mg; Cu 2.38 mg; Co 0.34 mg; I 0.10 mg; Se 0.15 mg.

\*\*Kemzyme Plus Dry : minimal guaranteed enzyme activity, Endo 1,3(4) beta –glucanase (beta-glucanase) – 2350 Units/g, Endo1,4 beta-glucanase (celulase) 4000 Units /g, Alpha –Amylase 400 Units /g, Bacilolysin (protease) 450Units/g Endo 1,4 beta-xylanase (xylanase) 20000 Units/g

\*\*\*xylanase: minimal guaranteed enzyme activity 80000 Units/g, or beta-glucanase: minimal guaranteed enzyme activity 4000 Units/g

## Results

Determination of the NSP of the raw materials was made in the University of Dublin laboratory. The obtained values are shown in table 3.

**Table 3.** The feed contents in NSP

Growth period	Specification	NSPs* (g/kg)	NSPi** (g/kg)	NSPt*** (g/kg)
Period hatching-3 weeks	600 g/kg wheat	21±2.1	95±2.5	116±4.2
	600 g/kg barley	27.64±3.2	112.74±0.45	140.38±9.4
Period 3-6 weeks	600 g/kg wheat	20.3±2	90.3±2.2	110.6±4.3
	600 g/kg barley	32.91±6	113.2±5	146.11±0.5

\* NSP soluble,

\*\* NSP insoluble,

\*\*\* NSP total

From the data in table 3 it can be noticed that the content of soluble NSP was between 20.3 to 21 g/kg of complete feed, obtained by adding 600 g/kg of wheat to the complete feed, and between 27.64 to 32.91 g/kg of complete feed by adding barley. The content of NSPi was between 90.3-95 g/kg complete feed when wheat was added and between 112.74-113.2 g/kg of complete feed when barley was added.

### Feed consumption

A first indicator discussed is the evolution of feed consumption which is presented in data table 4.

**Table 4.** Mean ( $\pm$ SEM) feed intake of four experimental groups differing in enzymes kind

Specification	Wheat					Barley				
	Feed intake, mean kg/day/chicken									
	EG <sub>1</sub>		EG <sub>2</sub>		P	EG <sub>3</sub>		EG <sub>4</sub>		P
mean	SEM	mean	SEM	mean		SEM	mean	SEM		
period 0-3 weeks	0.935 <sup>a</sup>	0.005	0.974 <sup>b</sup>	0.004	0.012*	0.883 <sup>a</sup>	0.005	0.898 <sup>a</sup>	0.014	0.444 <sup>is</sup>
period 3-6 weeks	2.940 <sup>a</sup>	0.021	2.99 <sup>a</sup>	0.058	0.504 <sup>is</sup>	2.820 <sup>a</sup>	0.026	2.89 <sup>a</sup>	0.024	0.130 <sup>is</sup>
period 0-6 weeks	3.875 <sup>a</sup>	0.021	3.964 <sup>a</sup>	0.010	0.064 <sup>is</sup>	3.703 <sup>a</sup>	0.011	3.792 <sup>a</sup>	0.026	0.087 <sup>is</sup>

<sup>a-b</sup> Means within the same row bearing different superscripts differ significantly:

\*  $P < 0.05$ , \*\*  $P < 0.01$ , \*\*\*  $P < 0.001$ , <sup>is</sup>  $P > 0.05$  –insignificant difference

To determine the amount of feed consumed the quantity of feed allocated for each experimental group during the 0-3 weeks was weighed. The same was done for 3-6 weeks. At the end of each experimental period the amount of feed consumed was weighed. In the data shown in the above table it can be noted that during 0-3 weeks of feed consumption for the experimental variant that consumed wheat-based feed plus an enzymatic complex there was a higher feed consumption ( $P < 0.05$ ) than EG<sub>1</sub> which consumed the same feed but contained only xylanase. The same result is found during 4-6 weeks and during 0-6 weeks where the difference is higher ( $P > 0.05$ ). EG<sub>4</sub> chickens that consumed the complete feed based on barley plus enzyme complex also consumed more feed ( $P > 0.05$ ). from hatching to six weeks. Therefore it appears that the use of enzyme complex in barley or wheat based complete feed causes an increase in broilers feed intake ( $P > 0.05$ ).

### Body weight

The determination of the development of body mass and weight gain during the experiment was done by an individual weekly weighing of chickens in each variant (table 5).

**Table 5.** The means ( $\pm$  SEM) body weight evolution during the experiment

Specification	EG <sub>1</sub>		EG <sub>2</sub>		P	EG <sub>3</sub>		EG <sub>4</sub>		P
	mean	SEM	mean	SEM		mean	SEM	mean	SEM	
Weight at 1 week (g)	150.84 <sup>a</sup>	±3.15	121.11 <sup>d</sup>	±1.91	0.0000	97.65 <sup>a</sup>	±3.36	137.11 <sup>d</sup>	±5.04	0,0000
Weight at 2 weeks (g)	370.4 <sup>a</sup>	±9.94	305.78 <sup>c</sup>	±9.73	0.0017	269.1 <sup>a</sup>	±10.3	342.5 <sup>c</sup>	±19.1	0.0014
Weight at 3 weeks (g)	759.7 <sup>a</sup>	±13.8	681.9 <sup>b</sup>	±15.9	0.0141	595.8 <sup>a</sup>	±16.2	673.2 <sup>b</sup>	±26.2	0.0143
Weight at 4 weeks (g)	1302.2 <sup>a</sup>	±36	1136.0 <sup>b</sup>	±60.4	0.0373	1071.6 <sup>a</sup>	±35.1	1156.0 <sup>a</sup>	±39.2	0.550
Weight at 5 weeks (g)	1760.2 <sup>a</sup>	±46.9	1571.3 <sup>a</sup>	±72.2	0.055	1360.0 <sup>a</sup>	±44	1498.2 <sup>a</sup>	±42.8	0.0662
Weight at 6 weeks (g)	2275.3 <sup>a</sup>	±37.3	2207.1 <sup>a</sup>	±44.2	0.407	1918.3 <sup>a</sup>	±62.4	2051.3 <sup>a</sup>	±81.9	0.1468

<sup>a,b</sup>  $-p < 0.05$ , <sup>a,c</sup>  $-p < 0.01$ , <sup>a,d</sup>  $-p < 0.001$ , <sup>a,a</sup>  $-p \geq 0.05$

The data table on the evolution of body weight shows broilers' body weight that consumed wheat-based complete feed plus an poly enzymatic complex (EG2) weighted less than broilers that consumed a wheat based compound feed only wheat plus xylanase, this new difference is statistically significant ( $P < 0.001$ ). At the age of two weeks the difference between the two experimental groups was statistically significant ( $P < 0.01$ ). The body weight in EG1 at the age of 3 weeks was also higher than EG2, a statistically insignificant difference ( $P > 0.05$ ). At four weeks old, chickens from EG1 also had higher body weight, a difference which is statistically significant ( $P < 0.05$ ). A higher body weight recorded on EG 1 at the age of 5 weeks and 6 weeks, these differences being statistically insignificant ( $P > 0.05$ ). Therefore it appears that in the wheat-based complete feed is recommended the use of xylanases mono-enzymes, and the use of poly-enzymatic complex cannot be justified.

In the case of broilers fed **barley**-based complete feed (EG4) at one week, the body weight was higher than in EG3 that consumed the same barley based complete feed only with beta glucanase added in its structure. This difference is statistically significant ( $P < 0.001$ ). This difference is maintained at the age of 2 weeks ( $P < 0.01\%$ ) and at 3 weeks, also statistically significant ( $P < 0.05\%$ ).

From the age of 4, 5 and 6 weeks the difference is reduced ( $P > 0.05$ ). Therefore, it appears that with barley-based complete feed the use of poly-enzymatic complex is recommended.

### ***The feed conversion rate (FCR)***

Feed consumption efficiency was based on feed consumption data and total average gain, the feed conversion ratio being calculated as kg feed / kg gain (table 6).

**Table 6.** Feed conversion rate evolution in chickens from the experimental groups

Specification	Feed Conversion rate (kg complete feed/kg growth)									
	EG <sub>1</sub>		EG <sub>2</sub>			EG <sub>3</sub>		EG <sub>4</sub>		P
	mean	SEM	mean	SEM		mean	SEM	mean	SEM	
period 0-3 weeks	1.30 <sup>a</sup>	0.005	1.51 <sup>b</sup>	0.012	0.004 **	1.59 <sup>a</sup>	0.015	1.41 <sup>b</sup>	0.008	0.002 **
period 4-6 weeks	1.94 <sup>a</sup>	0.008	1.96 <sup>a</sup>	0.008	0.184 is	2.13 <sup>a</sup>	0.008	2.10 <sup>a</sup>	0.008	0.135 is
period 0-6 weeks	1.73 <sup>a</sup>	0.006	1.83 <sup>b</sup>	0.006	0.000 ***	1.97 <sup>a</sup>	0.010	1.89 <sup>b</sup>	0.008	0.008 **

<sup>a-b</sup> Means within the same row bearing different superscripts differ significantly:

\*  $P < 0.05$ , \*\*  $P < 0.01$ , \*\*\*  $P < 0.001$ , <sup>is</sup>  $P > 0.05$  –insignificant difference

The data table 6 shows that the use of complex enzymatic in complete feed based on wheat (EG2), increases the feed conversion ( $P < 0.01\%$ ) rate during the growing period from hatching to 3 weeks, from 4 weeks to slaughter ( $P > 0.05\%$ ) and for the entire growing period ( $P < 0.001\%$ ). Using the same enzyme complex complete feed based on barley compared with those using a single enzyme beta-glucanase, resulted in reduced feed conversion rate in the first growing period ( $P < 0.01\%$ ), in the second period ( $P > 0.05\%$ ) and for the entire growing period (from hatching to six weeks) ( $P < 0.01\%$ ). Therefore, the enzymatic complex supplementation in wheat based complete feed increases the feed conversion rate and in the barley-based complete feed it reduces the feed conversion rate.

**Viscosity at intestinal level**

After the slaughter of broilers at the age of 6 weeks we collected intestinal contents from duodenum and jejunum in order to determine viscosity values.

**Table 7.** The intestinal viscosity at jejunum and duodenal level for broilers at six weeks of age

Specification	Wheat					Barley				
	Intestinal viscosity at duodenum									
	EG <sub>1</sub>		EG <sub>2</sub>		P	EG <sub>3</sub>		EG <sub>4</sub>		P
mean	SEM	mean	SEM	mean		SEM	mean	SEM		
	2.41 <sup>a</sup>	0.024	2.61 <sup>b</sup>	0.012	0.019*	2.50 <sup>a</sup>	0.012	2.35 <sup>b</sup>	0.008	0.002**
Specification	Intestinal viscosity at jejunum									
	EG <sub>1</sub>		EG <sub>2</sub>		P	EG <sub>3</sub>		EG <sub>4</sub>		P
	mean	SEM	mean	SEM		mean	SEM	mean	SEM	
	2.08 <sup>a</sup>	0.014	2.53 <sup>b</sup>	0.030	0.006**	2.20 <sup>a</sup>	0.008	2.17 <sup>a</sup>	0.017	0.309 <sup>is</sup>

<sup>a-b</sup> Means within the same row bearing different superscripts differ significantly:

\* P<0.05, \*\* P<0.01, \*\*\* P<0.001, <sup>is</sup> P>0.05 –insignificant difference

From the above data table it appears that the incorporation of an enzymatic complex into complete feed based on wheat leads to a duodenal viscosity ( $P < 0.05\%$ ) and jejunum viscosity higher ( $P < 0.01\%$ ) compared to chickens that ate complete feed containing wheat but with the addition of xylanase. Incorporating the same enzymatic complex in barley-based complete feed decreases intestinal viscosity in duodenum ( $P < 0.01\%$ ) and in the jejunum ( $P > 0.05\%$ ).

**Regression equations of intestinal viscosity depending on the content of soluble NSP**

It is known that the viscosity of the gut is given by soluble NSP. Therefore, there is a direct correlation between the content of soluble NSP and viscosity at the duodenum and jejunum level. Regression equations and R<sup>2</sup> value are given below in table 8 and 9.

**Table 8.** The correlation between duodenum viscosity and soluble NSP

Specification	Equation	R <sup>2</sup>	Prob(F)
Duodenum viscosity (y)- soluble (x)	Y = 147.375 + 0.167543*exp(x) + 400.2373*ln(x)/x	0.9986	0.03678 *

**Table 9.** The correlation between jejunum viscosity and soluble NSP

Specification	Equation	R <sup>2</sup>	Prob(F)
Jejunum viscosity (y)- soluble (x)	Y = 114.7505 + 0.164369x <sup>3</sup> - 42.6661x/ln(x)	0.99851	0.02226 *

**Determination of biochemical indices of blood serum in chickens from the experimental groups**

Following slaughter, blood was collected from a total of five broilers in each experimental variant. Cholesterol and triglyceride determinations were made and the values are given in Table 10.

**Table 10.** The biochemical indices of blood serum in the broilers from experiment (mg/100 ml)

Specification	Reference values	Experimental group			
		EG1 Mean $\pm$ SEM	EG2 Mean $\pm$ SEM	EG3 Mean $\pm$ SEM	EG4 Mean $\pm$ SEM
Cholesterol	105 $\pm$ 15	120.5 <sup>a</sup> $\pm$ 1.92	126.5 <sup>a</sup> $\pm$ 4.60	114.5 <sup>a</sup> $\pm$ 1.92	112 <sup>a</sup> $\pm$ 4.24
Triglycerides	60 $\pm$ 20	73.5 <sup>a</sup> $\pm$ 1.71	88 <sup>b</sup> $\pm$ 4.04	72 <sup>a</sup> $\pm$ 5.55	61.5 <sup>a</sup> $\pm$ 0.70

<sup>a-a</sup> ( $P > 0.05$ ) <sup>a-b</sup> ( $P < 0.05$ ) Means within the same row bearing different superscripts differ significantly

The data table shows that the incorporation of enzymatic complexes in wheat-based complete feed increases the cholesterol content of the blood serum ( $P > 0.05\%$ ). and for incorporation in complete feed based on barley it lowers the cholesterol content ( $P > 0.05$ ). Regarding the content of triglycerides we can note that adding enzyme preparations to wheat-based complete feed increases the content ( $P < 0.05$ ) and in barley-based complete feed it lowers the triglycerides ( $P > 0.05$ ).

## Discussions

This study showed that adding the xylanase enzyme in wheat-based complete feed (600 g/kg) improves bio-productive performances. The same results were obtained by Gao *et al.*, 2008, Onderci 2008, (Brenes *et al.*, 1993; Choct *et al.*, 1999, Wang *et al.*, 2005). Adding complex enzyme Endo-1 ,4-beta-glucanase (cellulase complex), alpha-amylase, Bacilloylisin (protease) and Endo-1 .4-beta-xylanase in the barley based complete feed, compared with the simple addition of betaglucanase is justified by the fact that the cellulose is also an NSP derivative that is used efficiently by the enzymes present in the enzymatic complex. Improvements in gain, feed/gain (FCR), intestinal viscosity is associated with multi-enzyme addition (Gohl *et al.*, 1978), Jozefiak, 2006). The use of beta-glucanase enzymes is important in broiler diets containing high levels of barley, or in some cases of wheat. Barley contains between 2% and 6% beta-glucan and yields highly viscous solutions (Dunne, 1995). Wheat and rye contain between 4% and 8% of pentosans (arabinoxylan) gums which are not readily metabolizable by the young chickens (Marquardt *et al.*, (1996). Research (Gohl *et al.*, 1978; Belyavin, 1994; Pack *et al.*, 1998; Lobo, 1999) has demonstrated the benefits of adding enzyme when directed at a specific feed. Using enzymatic complexes in barley-based complete feed decreases intestinal viscosity in the duodenum and jejunum, while the use of xylanase in complete feed lowers the viscosity at duodenum and jejunum, while in barley based complete feed the intestinal viscosity decreases in the duodenum and jejunum. Soluble arabinoxilans in wheat are responsible for increasing the intestinal viscosity (Choct & Annison, 1992). The reduction of the intestinal viscosity is one of the main functions of exogenous enzymes (Josefiak *et al.*, 2007). Dusel *et al.*, 1998 shows that adding xylanase decreases intestinal viscosity, same results having been obtained in this paper as well. Enzyme addition improves performance, especially in the first weeks, opposite results obtained by Dusel *et al.*, 1998. Campbell *et al.*, 1989 shows that the use of barley in poultry feed is limited due to this particularly water soluble NSP mainly (1-3),(1-4) glucans. They are also present in grain and increase digesta viscosity, which interferes with the activity of intestinal enzymes in the gastrointestinal tract. Feeding barley increases the incidence of sticky manure, reduces the extent of digestion and absorption of nutrients, and impairs broiler performance (Annison & Choct, 1991). Supplementation of glucanase has been shown to minimize the negative effects of barley glucan (Wiedmer & Volker, 1989; Brufau *et al.*, 1992; Broz *et al.*, 1994;



Marquardt *et al.*, 1996; Perttila *et al.*, 2001 Onderci *et al.*, 2008). Improved performance by using enzyme production can be explained by the following: solubilisation of cell wall NSP, eliminating encapsulated nutrients, hydrolysis of certain types of carbs-protein links, obtaining available sugars by hydrolysis oligosaccharides (Slominski, 1995). The results from this study showed that incorporation of poly-enzymatic complex in complete feed based on wheat increases the triglycerides content and incorporation of enzymes in complete feed based on wheat and barley does not change the cholesterol content in blood serum. Similar results were obtained by Wang, 1992.

In conclusion, the addition of poly-enzymatic complex in the structure of the complete feed based on barley determine improved performance of broiler production. The addition of beta glucanase in the structure of the complete feed based on wheat determines improved performance of the broiler production. It is not necessary to use enzymatic complex in the structure of the complete feed based on wheat only in the structure of the complete feed based on barley.

## Conclusions

1. Adding poly-enzymatic complex in complete feed based on barley leads to higher body weight ( $P < 0.05$ ) at 1,2,3 weeks and to the age of 4,5,6 weeks ( $P > 0.05$ ), compared to the use of beta glucanase; incorporating poly-enzymatic complex in wheat-based complete feed, lead to lower body weight ( $P < 0.05$ ) at the age of 1, 2,3,4 weeks ( $P < 0.05$ ) and 5,6 weeks ( $P > 0.05$ ) compared to the use of xylanase in the complete feed. The feed conversion rate and the viscosity in the gut are influenced positively by the introduction of enzymatic complexes in complete feed based on barley and of xylanase in wheat-based complete feed.

2. Incorporation of poly-enzymatic complex in complete feed based on wheat increases the triglycerides content ( $P < 0.05$ ) and in barley-based complete feed decreases the triglycerides ( $P > 0.05$ ).

3. Incorporation of enzymes in complete feed based on wheat and barley does not change the cholesterol content in blood serum.

4. It is necessary to use xylanase in the structure of the complete feed based on wheat and the enzymatic complex in the structure of the complete feed based on barley.

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## References

1. ALMIRALL M, FRENCESCH M, PEREZ-VENDRELL AM, BRUFAU J. & ESTEVE-GARCIA E. 1995. The differences in intestinal viscosity produced by barley and b-glucanase alter digesta enzymes activities and ileal nutrient digestibility more in broiler chicks than in cocks. *Journal Nutrition* 125, 947–955.
2. AMAN P & GRAHAM H. 1987. Analysis of total and insoluble mixed-linked (1,3),(1,4)-b-glucans in barley and oats. *Journal . Cereal Science* 3, 704–709.
3. ANDERSON JW, DEAKINS DA. & BRIDGES, SR., 1990. Soluble fiber: hypocholesterolemic effects and proposed mechanisms. In: Kritchevsky, D., Bonfield, C., Anderson, J.W. (Eds.), *Dietary Fiber, Chemistry, Physiology and Health Effects*. Plenum Press, New York, pp. 339–363.
4. ANNISON G & CHOCT M.,1991. Anti-nutritive activities of cereal non-starch polysaccharides in broiler diets and strategies minimizing their effects. *Worlds Poultry Science Journal*. 47, 222–242.

5. BEDFORD, M.R., CLASSEN, H.L. & CAMPBELL, G.L. 1991. The effect of pelleting, salt and pentosanase on the viscosity of intestinal contents and the performance of broilers fed rye. *Poultry Science*. 70, 1571–1577.
6. BELYAVIN CG. 1994. In-Feed enzymes. *Poultry International*. 35 (11), 54–56.
7. BRENES A, SMITH M, GUENTER W. & MARQUARDT, RR. 1993. Effect of enzyme supplementation on the performance and digestive tract size of broiler chickens fed wheat- and barley- based diets. *Poultry Science*. 72, 1731–1739.
8. BROZ J, OLDALE P, PERRIN-VOLTZ A-H., 1994. Effects of *Trichoderma viride* enzyme complex in broiler chickens. *Archives. Geflügel*. 58, 130–134.
9. BRUFAU J, FRANCESCH M, LEGARDA JE, PEREZ-VENDRELL, AM & ESTEVE-GARCIA E., 1992. The effect of an enzyme supplement on the apparent metabolisable energy of a wheat in broiler diets. In: *Proceedings, 19th World's Poultry Congress, 20–24 Sep, Amsterdam, Netherlands*, p. 452.
10. CAMPBELL GL, ROSSNAGEL BF, CLASSEN HL. & TACKER PA., 1989. Genotypic and environmental differences in extract viscosity of barley and their relationship to its nutritive value for broiler chickens. *Animal Feed Science Technology*. 26, 221–230.
11. CHOCT M & ANNISON G., 1992. Antinutritive activity of wheat arabinoxylans: role of viscosity and gut microflora. *British Poultry Science*. 33, 821–834.
12. CHOCT M. & ANNISON G., 1992. Anti nutritive effect of wheat pentosans in broiler chickens: roles of viscosity and gut microflora. *British Poultry Science*. 33, 821-834.
13. CHOCT M., HUGHES R.J. & BEDFORD MR. 1999. Effects of a xylanase on individual bird variation, starch digestion throughout the intestine, and ileal and caecal volatile fatty acid production in chickens fed wheat. *British Poultry Science*. 40, 419–422.
14. CHOCT M, HUGHES RJ, WANG J, BEDFORD MR, MORGAN AJ. & ANNISON G. 1996. Increased small intestinal fermentation is partly responsible for the anti-nutritive activity of non-starch polysaccharides in chickens. *British Poultry Science*. 37, 609–621.
15. DUNNE J., 1995. History of enzyme production and usage. In: *Official Proceedings 13th Annual Pacific Northwest Animal Nutrition Conference, October 10–12, Marriott Hotel, Portland, OR, USA*, pp. 87–102.
16. DUSEL G, KLUGE H, GLASE KAROLA, SIMON O, HARTMANN G, LENGGERKEN JV. & JEROCH H. 1997. An investigation into the variability of extract viscosity of wheat- relationship with the content of non starch polysaccharide fractions and metabolisable energy for broiler chickens, *Archives Animal Nutrition*, vol. 50, pp 121-135.
17. ENGLYST H. 1989. Classification and measurement of plant polysaccharides. *Animal Feed Science and Technology*. 23: 27-42.
18. GAO F, JIANG Y., ZHOUL GH, HAN ZK. 2008. The effects of xylanase supplementation on performance, characteristics of the gastrointestinal tract, blood parameters and gut microflora in broilers fed on wheat-based diets ,China
19. GOHL B, ALDEN S, ELWINGER K. & THORKE S. 1978. Influence of beta-glucanase on feeding value of barley for poultry and moisture content of excreta. *British Poultry Science*. 19, 41–47.
20. JOSEFIK D, RUTKOWSKI A, JENSEN BB. & ENGBERG RM. 2006. Effects of dietary inclusion of triticale, rye and wheat and xylanase supplementation on growth performance of broiler chickens and fermentation in gastrointestinal tract, *Livestock Science*, volume 108, issue 1-3, pp. 102-105
21. LOBO P, 1999. Using enzymes to enhance corn and soy-based broiler diets. *Feed Manage*. 50 (10), 17–20.
22. MARQUARDT RR, BRENES A, ZHANG Z. & BOROS D. 1996. Use of enzymes to improve nutrient availability in poultry feedstuffs. *Animal Feed Science and Technology*. 60, 321–330.
23. ONDERCI M, SAHIN N, CIKIM G, AYDIND A, OZERCAN I, OZKOSE E, EKINCI S, HAYIRLI A. & SAHIN K. 2008. Beta -Glucanase-producing bacterial culture improves performance and nutrient utilization and alters gut morphology of broilers fed a barley-based diet, *Animal Feed Science and Technology*, 146, 87–97
24. PACK M, BEDFORD M. & WYATT . 1998. Feed enzymes for corn–soybean broiler diets. *Poultry Digest* (June/July), 34–37.
25. PERTTILA S, VALAJA J, PARTANEN K, JALAVA T, KIISKINEN T. & PALANDER S. 2001. Effects of reservation method and beta -glucanase supplementation on ileal amino acid digestibility and feeding value of barley for poultry. *British Poultry Science*. 42, 218–229.
26. SLOMINSKI BA. 1995. Carbohydrates in feedstuffs in relation to dietary enzyme use. In: *Official Proceedings 13th Annual Pacific Northwest Animal Nutrition Conference, October 10–12, Marriott Hotel, Portland, OR, USA*, pp. 140–162.
27. SMITS CHM. & ANNISON G. 1996. Non-starch plant polysaccharides in broiler nutrition: towards a physiologically valid approach to their determination. *Worlds Poultry Science Journal*. 52, 203–221.

28. WANG L, NEWMAN RK., NEWMAN WC. & HOFER P. 1992. Barley beta glucans alter viscosity and reduce plasma cholesterol concentrations in chicks. , *Journal of Nutrition, American Institute of Nutrition*, pp 2292-2297
29. WANG ZR, QIAO SY, LU WQ. & LI DF. 2005. Effects of enzyme supplementation on performance, nutrient digestibility, gastrointestinal morphology, and volatile fatty acid profiles in the hindgut of broilers fed wheat based diets. *Poultry Science*, 84, 875–881.
30. WHITE WB, BIRD HR, SUNDE ML, MARLETT JA, PRENTICE NA. & BURGER WC. 1983. Viscosity of beta-glucan as a factor in the enzymatic improvement of barley for chicks. *Poultry. Science*. 62, 853–862.
31. WIEDMER H. & VOLKER L. 1989. Enzyme supplementation of a barley based diet fed to broiler chickens under practical conditions. In: *Proceedings, 7th European Symposium on Poultry Nutrition*, 19–21 June, pp. 252–253.
32. YU B, HSU JC. & CHIOU PWS. 1997. Effects of beta-glucanase supplementation of barley diets in growth performance of broilers. *Animal Feed Science and Technology* 70, 353–361.