

## **EFFECT OF XYLANASE SUPPLEMENTATION TO CEREAL-BASED DIETS ON APPARENT FECAL DIGESTIBILITY AND GROWTH PERFORMANCE OF PIGS**

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### **Abstract**

A total of 300 growing-finishing pigs (30–110 kg BW), the offspring of Naima sows and Duroc boars, were allocated to three dietary treatments (five pens per treatment, 10 males and 10 females per pen). The growth performance of pigs was determined, and the apparent digestibility of nutrients and energy in animals fed grower diets was calculated by the simple balance method. Complete diets with a high content of triticale (70%–73%) and barley (6.8%–12%) were supplemented with endo-1,4-beta-xylanase (6200 EPU per g). The minimum xylanase activity per kg feed was 1050 and 1500 EPU. Xylanase, at activity levels of 1050 and 1500 EPU kg<sup>-1</sup> complete diet, significantly ( $P \leq 0.05$ ) increased the apparent digestibility of total protein and highly significantly ( $P \leq 0.01$ ) that of crude fat. No significant differences in the digestibility of N-free extractives and energy were found between groups. The increased activity of xylanase (1500 EPU kg<sup>-1</sup> feed) contributed to an increase in the digestibility of dry matter, crude fibre ( $P \leq 0.05$ ) and organic matter ( $P \leq 0.01$ ), compared with the control group. Higher ( $P \leq 0.01$ ) daily gains and average final body weights were noted in pigs fed diets supplemented with the mono-enzyme preparation. The feed conversion ratio (FCR) tended to improve in pigs receiving xylanase-supplemented diets, but the observed differences were statistically non-significant.

**Key words:** pigs, xylanase, fecal digestibility, growth performance

Due to the specific structure of their gastrointestinal tract, pigs do not produce sufficient endogenous enzymes that efficiently degrade non-starch polysaccharides (NSPs), and therefore they are unable to absorb and fully utilize the nutrients supplied in the diet.

Cereal grains and cereal products are characterized by particularly high levels of non-starch polysaccharides (NSPs) such as (1–3), (1–4)- $\beta$ -glucans, cellulose, pectins

and arabinoxylans, which affect their nutritional value (Barrera et al., 2004). According to Diebold et al. (2005), the cell walls of cereal grains may contain up to 15% NSPs. In wheat and triticale grain, which is most commonly used in monogastric animal diets, the major NSPs are arabinoxylans (Diebold et al., 2005) degraded by endo-xylanase (Goesaert et al., 2002). Soluble polysaccharides absorb water, which increases digesta viscosity. High digesta viscosity can lead to digestive disorders. There is a close correlation between NSP concentrations and the degree of digesta viscosity (Dusel et al., 2006; Nortey et al., 2007 a, b). Inefficient enzymatic hydrolysis of NSPs decreases the digestibility of nutrients, including some amino acids and energy (Yin et al., 2000; Omogbenigun et al., 2004; Nortey et al., 2007 a; Sterk et al., 2007). Arabinoxylans, considered to be the main antinutritional factor in rye products, adversely affect the growth performance of pigs and feed conversion. Pig diets can be supplemented with mono- and multi-enzyme preparations to increase their nutritional value and fattening results. Systemic enzyme support enables improving nutrient digestibility and utilization as well as the energy value of diets. As a result, better fattening performance can be expected, particularly in young animals. The relevant literature data and research findings are statistically significant.

The objective of this study was to determine the effect of different levels of endo-1,4-beta-xylanase on nutrient digestibility in complete diets with a high triticale content, and the growth performance of pigs under commercial production conditions.

## Material and methods

### Digestibility trial

The apparent digestibility of nutrients and energy in animals fed grower diets was calculated by the simple balance method. A five-day experimental period was preceded by a seven-day adaptation period. The experimental material comprised 24 male pigs with average body weight of 45 kg, the offspring of Naima sows and Duroc boars, allocated to three dietary treatments, with eight animals per treatment. The pigs were kept in individual metabolism cages, and they were fed grower diets with different activity levels of endo-1,4-beta-xylanase (1050 and 1500 EPU kg<sup>-1</sup> feed). The composition of complete pelleted diets (per kg<sup>-1</sup>) was as follows: triticale – 700 g, barley – 68 g, soybean meal – 100 g, rapeseed meal – 93 g, soybean oil – 6 g (Table 1). The metabolizable energy (ME) and total protein content per kg feed was 13.2 MJ and 170 g, respectively; content of calculated total non-starch polysaccharides (NSPs) reached 129 g. The digestibility trial was conducted simultaneously with an on-farm trial, which was performed on the same experimental material, using an identical experimental design.

The nutrient content of feed components, diets and feces was determined by standard methods (AOAC, 1990). The gross energy content of average diet and fecal samples was calculated based on the total heat of combustion of organic matter in a KL-5 calorimeter.

Table 1. Formulation and calculated nutrient content of diets

Ingredients (g kg <sup>-1</sup> )	Diet	
	growing pigs	finishing pigs
Triticale	700.0	733.7
Barley	68.0	120.0
Soybean meal	100.0	-
Rapeseed meal	93.0	120.0
Soybean oil	6.0	-
L-lysine	3.8	3.7
DL-methionine	0.6	0.4
L-threonine	1.0	1.2
Limestone	6.4	4.7
Monocalcium phosphate	8.2	4.1
Salt	3.0	2.2
Mineral and vitamin premix <sup>a</sup>	10.0	10.0
Calculated nutrient content (g kgc <sup>-1</sup> ) <sup>b</sup> :		
Crude protein	170.0	140.0
Lysine	10.2	8.1
Methionine+cystine	6.3	5.5
Threonine	6.5	5.6
Tryptophan	2.0	1.7
Ca	7.5	6.0
P, available	3.2	2.3
Na	1.5	1.2
Metabolizable energy (MJ kg <sup>-1</sup> )	13.2	12.8

<sup>a</sup> Supplied per kilogram of mixture, Vitamins: A 10000 IU, D<sub>3</sub> 2000 IU, E 70 mg, K<sub>1</sub> 1.5 mg, B<sub>1</sub> 1.5 mg, B<sub>2</sub> 4 mg, B<sub>6</sub> 3 mg, B<sub>12</sub> 25 µg, pantothenic acid 10 mg, folic acid 2 mg, niacin 20 mg, biotin 0.1 mg, choline chloride 300 mg; Minerals: Zn 100 mg, Mn 40 mg, Cu 20 mg, Fe 100mg, J 1.2 mg, Co 0.6 mg, Se 0.3 mg, Ca 3.20, 3.15, 3.10 g for 0, 1050, 1500 EPU xylanase kg<sup>-1</sup> respectively.

<sup>b</sup> Calculated from ingredients (NRC, 1998).

### On-farm trial

A total of 300 growing-finishing pigs, the offspring of Naima sows and Duroc boars, were divided into three equal groups. Each dietary treatment consisted of five pens (10 males and 10 females per pen). The animals were fattened from average body weight of ca. 30 kg to ca. 110 kg, at two stages: grower (30–60 kg BW) and finisher (60–110 kg BW).

The pigs were fed *ad libitum* complete pelleted diets with a high content of ground triticale and barley. The content of triticale, the major source of NSPs, was 70.00% and 73.37% in grower and finisher diets, respectively. The composition and nutritional value of diets, and nutrient content are presented in Tables 1 and 2, respectively. The diets for experimental group animals were supplemented with endo-1,4-beta-xylanase as micro-pellets, in the amount of 0.175 kg and 0.250 kg t<sup>-1</sup> complete diet. The minimum xylanase activity was 1050 and 1500 EPU kg<sup>-1</sup> feed.

Enzyme activity per g of the supplement was 6200 EPU. The results obtained were verified statistically by one-way ANOVA, and the significance of differences between groups was determined by Duncan's test, using Statistica 7 software.

Table 2. Chemical composition of diets (g kg<sup>-1</sup>)

Ingredients	Growing pigs			Finishing pigs		
	Xylanase <sup>a</sup> EPU kg <sup>-1</sup>			Xylanase <sup>a</sup> EPU kg <sup>-1</sup>		
	0	1050	1500	0	1050	1500
Dry matter	896.7	893.3	887.5	896.5	882.4	879.0
Crude ash	54.1	48.0	51.4	45.1	40.4	43.7
Crude protein	176.8	179.5	177.2	137.6	139.6	133.5
Crude fat	21.9	21.4	20.2	28.1	31.6	31.0
Crude fibre	33.3	32.2	34.2	37.6	34.7	37.7
N-free extractives	610.6	612.2	604.5	648.1	636.1	633.1
Organic matter	842.6	845.3	836.1	851.4	842.0	835.3

<sup>a</sup> content: 6200 EPU endo-1,4-beta-xylanase g<sup>-1</sup>.

## Results

An analysis of the apparent fecal digestibility of nutrients revealed that endo-1,4-beta-xylanase had a highly significant effect on crude fat digestibility, regardless of the enzyme activity level (Table 3). In comparison with the control diet, the increase in crude fat digestibility reached 36% (1050 EPU kg<sup>-1</sup>) and 43% (1500 EPU kg<sup>-1</sup>).

Table 3. Fecal nutrient digestibility (%) of diets

Item	Xylanase, EPU kg <sup>-1</sup>			SEM	P
	0	1050	1500		
Dry matter	81.50 b	82.80	83.17 a	0.289	0.038
Crude protein	77.03 b	78.73 a	78.89 a	0.339	0.039
Crude fat	45.29 B	61.57 A	64.90 A	2.514	0.006
Crude fibre	32.55 b	36.24	37.86 a	0.912	0.043
N-free extractives	89.78	90.53	90.41	0.280	0.516
Organic matter	83.42 Bb	84.91 a	85.17 A	0.266	0.009
Gross energy	80.97	82.04	82.29	0.268	0.097

a, b – P≤0.05.

A, B – P≤0.01.

Endo-1,4-beta-xylanase significantly (P≤0.05) improved total protein digestibility, but an increase in enzyme activity from 1050 to 1500 EPU kg<sup>-1</sup> feed was not followed by an increase in protein digestibility.

Endo-1,4-beta-xylanase at the higher activity level significantly improved the apparent digestibility of dry matter and crude fibre. The difference between the control diet and the diet supplemented with xylanase at 1500 EPU per kg feed reached 2% for dry matter and 16% for crude fibre.

The increase in organic matter digestibility was determined by the enzyme supplementation level (1050 vs. 1500 EPU kg<sup>-1</sup>), and the difference relative to the control diet was significant (1.8%) and highly significant (2%). This could result from low variation (SEM 0.266) in organic matter digestibility within dietary treatments.

No significant differences were found between groups with respect to the apparent digestibility of N-free extractives.

Surprisingly, the mono-enzyme preparation added to diets with a high content of ground triticale and barley had no significant influence on energy digestibility. The difference between the treatment and control groups was as low as 1.3%–1.6%.

The fattening performance parameters of pigs are presented in Table 4. The overall health status of animals and feed intake were satisfactory in all groups throughout the experiment. The average daily gains noted in the control group were highly significantly ( $P \leq 0.01$ ) lower than in experimental groups (1500 EPU/kg), and their values varied depending on the enzyme activity level. After 37 days, the average body weight of pigs reached 61.3 kg and 61.7 kg in experimental groups, and 60.9 kg in the control group. At the second stage of fattening, average daily gains were 3% higher in experimental group animals, compared with control group pigs. There were no significant differences in feed intake and feed conversion efficiency between groups, regardless of pig age. Enzyme supplementation was less effective in the second stage of fattening (Table 4).

Table 4. Effect of the supplemental enzyme on the performance responses in growing pigs

Item	Xylanase, EPU/kg			SEM	P	Differences
	0	1050	1500			
n	5	5	5			
Initial weight (kg)	31.1	30.9	31.0	0.16	0.878	NS
0–37 days						
ADG (g)	805 B	821	831 A	4.26	0.025	0.01
ADFI (kg)	2.03	1.99	1.98	0.02	0.463	NS
FCR (kg/kg)	2.53	2.42	2.39	0.03	0.069	NS
38–87 days						
ADG	949	978	977	6.20	0.099	NS
ADFI	2.92	2.97	2.97	0.03	0.773	NS
FCR	3.08	3.04	3.04	0.03	0.800	NS
0–87 days						
Final weight (kg)	107.4 B	109.2 A	109.6 A	0.34	0.007	0.01
ADG	887 B	910 A	914 A	4.36	0.010	0.01
ADFI	2.54	2.55	2.55	0.02	0.987	NS
FCR	2.86	2.80	2.78	0.02	0.290	NS

ADG (g) – average daily gain; ADFI (kg) – average daily feed intake; FCR – feed conversion ratio.

a, b –  $P \leq 0.05$ .

A, B –  $P \leq 0.01$ .

After 87 days, the final body weights of pigs fed xylanase-supplemented diets were 1.6% and 2.1% higher in comparison with the control group. The observed differences were statistically significant ( $P \leq 0.01$ ). A similar trend was noted with respect to average daily gains. The feed conversion ratio was comparable in both experimental groups, and it was 2.1% and 2.8% lower, respectively, than in the control group.

## Discussion

Not all research studies indicate a beneficial influence of xylanase on apparent or true nutrient digestibility and an increase in energy concentrations as a result of mono-enzyme dietary supplementation. As demonstrated by Dusel et al. (2006) and Nortey et al. (2007 a), xylanase exerts a greater effect when added to diets with a higher content of NSPs, including arabinoxylans degraded by endo-xylanase (Goesaert et al., 2002). Arabinoxylans are present in large amounts in wheat and triticale grain, which is characterized by a relatively high energy value. Hence, most studies focus on diets based on wheat and wheat by-products. The mechanism of action of NSP-hydrolyzing enzymes involves partial hydrolysis of soluble and non-soluble NSPs, including those contained in cell walls. Enhanced enzymatic degradation of NSPs decreases digesta viscosity and passage rate, contributing to digesta retention and homogeneity, and reducing endogenous nitrogen secretion in the small intestine. According to Preston et al. (2001), a decrease in the apparent digestibility of CP and AA is correlated with increased endogenous nitrogen secretion in the presence of NSPs. Yin et al. (2000) noted a negative correlation between the apparent ileal digestibility of DM, E, CP and AA and NSPs concentrations which also determine the rate of fermentation processes, VFA concentrations and the pH of digesta. In a study by Vahjen et al. (2007), the addition of a mono-enzyme preparation to pig diets had no effect on the pH of gastric digesta, but it significantly reduced *Lactobacillus* spp. counts in the stomach and jejunum. Barrera et al. (2004), Kim et al. (2005) and Vahjen et al. (2007) reported a close correlation between digesta viscosity and the digestibility and utilization of nutrients, including AA, and energy, especially in poultry. However, research results remain inconclusive also in this area. An increase in the apparent and intestinal digestibility of nutrients and energy was observed in some studies (Omogbenigun et al., 2004; Diebold et al., 2005; Dusel et al., 2006; Moehn et al., 2007; Nortey et al., 2007 a, b; Sterk et al., 2007). In our study, crude fat showed the strongest response to the use of enzyme supplementation. Improvements in *in vivo* and *in vitro* digestibility of crude fat were described by Sterk et al. (2007) and Tapingkae et al. (2008). Experiments performed on fistulated pigs revealed a significant increase in the digestibility of amino acids such as glutamic acid, glycine, isoleucine and tyrosine, and a trend towards increased digestibility of aspartic acid, alanine, cysteine and leucine (Vahjen et al., 2007). In a study by Lindberg et al. (2007), xylanase added to diets with a high wheat grain content had no effect on the digestibility of OM, CP and NDF, determined in the entire gastrointestinal tract.

Attempts have also been made to use the synergistic effect of xylanase and phytase. As demonstrated by Kim et al. (2008), the response to the applied combination is determined by a variety of factors, such as diet substrates, the age and physiological status of animals, and the adaptation of the gastrointestinal tract to the physicochemical properties of digesta. According to some authors, the effectiveness of digestive enzyme supplements may be reduced by the natural microflora and endogenous plant enzymes, or both.

Differences have been found in digestibility coefficients, depending on the method adopted (ileal vs. fecal digestibility). Diebold et al. (2004) reported a positive effect of xylanase, applied alone or in combination with phospholipase, only on ileal nutrient digestibility, whereas no differences were observed in fecal digestibility, which could be related to various anabolic and catabolic activities of the microflora in the large intestine.

In the present study, the addition of endo-1,4-beta-xylanase at both activity levels to complete diets with a high triticale content was more effective in younger pigs, as evidenced by a significant increase ( $P \leq 0.01$ ) in their daily gains. A particularly noticeable improvement in the growth performance of pigs at the beginning of fattening could be due to the higher availability of nutrients contained in ground cereals resulting from enhanced enzymatic degradation of NSPs.

A similar trend was observed by Frankiewicz et al. (2003) in growing-finishing pigs fed diets with a high rye content, supplemented with the multi-enzyme preparation BELFEED. An analysis of our results for the entire feeding period indicates that xylanase supplementation had a beneficial influence on the daily gains and final body weights of pigs. An improvement in daily gains and feed conversion efficiency was also noted by Frankiewicz et al. (2003), Barrera et al. (2004) and Tapingkae et al. (2008). In an experiment by Nortey et al. (2007 b), supplemental xylanase improved feed conversion, but it had no effect on daily gains. On the other hand, Kim et al. (2005) reported that the growth performance of piglets was not affected by xylanase, applied alone or in combination with phosphatase. Some research findings suggest that the efficacy of xylanase supplementation is determined by the qualitative composition of carbohydrates in cereal grains. There is a positive correlation between the NSPs content of cereal grains and enzyme effectiveness. Thus, xylanase should exert more profound effects on digestion and utilization of nutrients and energy when added to diets with high concentrations of NSPs, including arabinoxylans.

It may be concluded that endo-1,4-beta-xylanase, at activity levels of 1050 and 1500 EPU  $\text{kg}^{-1}$ , added to complete diets with a high content of triticale (70%) and barley (6.8%), significantly ( $P \leq 0.05$ ) increased the apparent digestibility of total protein and highly significantly ( $P \leq 0.01$ ) that of crude fat. No significant differences in the digestibility of N-free extractives and energy were found between groups. The increased activity of xylanase (1500 EPU  $\text{kg}^{-1}$  feed) contributed to an increase in the digestibility of dry matter, crude fibre ( $P \leq 0.05$ ) and organic matter ( $P \leq 0.01$ ), compared with the control group. Higher ( $P \leq 0.01$ ) daily gains and average final body weights were noted in pigs fed diets supplemented with the mono-enzyme preparation. The feed conversion ratio (FCR) tended to improve in pigs receiving xylanase-supplemented diets, but the observed differences were statistically non-significant.

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**Wpływ suplementacji diet z udziałem zbóż ksyłanazą na pozorną strawność kałową i wskaźniki  
tuczu świń**

STRESZCZENIE

Obserwacją objęto 300 rosnących tuczników (30 kg–110 kg) pochodzących z kojarzenia loch linii Naima z knurami rasy Duroc, które podzielono na 3 grupy żywieniowe (5 kojców w grupie, 10 lozek i 10 wieprzków w kojcu). Określono wskaźniki tuczu świń oraz pozorną strawność składników pokarmowych i energii mieszanek grower metodą bilansową prostą. Żywienie tuczników zróżnicowano dodatkiem endo – 1,4- $\beta$ -ksylanazy o aktywności 6200 EPU w 1 g preparatu, który wprowadzono do mieszanek pełnoporcjowych z wysokim udziałem pszenżyta (70–73%) i jęczmienia (6,8–12%). Minimalna aktywność ksyłanazy w 1 kg mieszanki odpowiadała 1050 i 1500 EPU. Zastosowanie w żywieniu tuczników ksyłanazy o poziomie aktywności 1050 i 1500 EPU  $\text{kg}^{-1}$  mieszanki pełnoporcjowej podwyższyło istotnie ( $P \leq 0,05$ ) współczynnik strawności pozornej białka ogólnego oraz wysoko-istotnie ( $P \leq 0,01$ ) tłuszczu surowego. Nie stwierdzono różnic w strawności związków bezazotowych wyciągowych i energii. Podwyższona aktywność enzymatyczna ksyłanazy do 1500 EPU  $\text{kg}^{-1}$  wpłynęła na wzrost strawności suchej masy, włókna surowego ( $P \leq 0,05$ ) i substancji organicznej ( $P \leq 0,01$ ) w stosunku do żywienia kontrolnego. Wyższe ( $P \leq 0,01$ ) dobowe przyrosty i średnią masę ciała przy zakończeniu doświadczenia osiągnęły tuczniaki żywione mieszankami z dodatkiem preparatu monoenzymatycznego. Tendencje do poprawy wskaźnika wykorzystania paszy w wyniku suplementacji żywienia ksyłanazą nie znalazły potwierdzenia statystycznego.