## The effect of arabinoxylanase and protease supplementation on nutritional value of diets containing wheat bran or rice bran in growing pig<sup>\*</sup>

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## ABSTRACT

One digestibility and one performance experiments were conducted to investigate the effects of a feed enzyme of arabinoxylanase and arabinoxylanase plus protease on digestibility of nutrients, nitrogen (N) flow and performance of the growing pigs fed a maize-wheat bran-based diet or a maize-rice bran-based diet. In the digestion experiment, six Landrace  $\times$  Large White  $\times$  Chinese Black male pigs of approximately 20 kg initial body weight, fitted with a simple T-cannula at the terminal ileum, were used to study dietary nutrient digestibility and nitrogen flow of the experimental diets. The six experimental diets were prepared in this study: maize-wheat bran diet, consists of maize, wheat bran, soyabean, of rapeseed meal, without feed enzyme supplementation (MWB); MWB supplemented with arabinoxylanase (A); MWB supplemented with arabinoxylanase plus protease (AP); maize-rice bran diet, consists of maize, rice bran, soyabean, rapeseed meal, without feed enzyme supplementation (MRB); MRB supplemented with arabinoxylanase (A); MRB supplemented with arabinoxylanase plus protease (AP). The MWB diet contained: %: soluble nonstrach polysaccharide (NSP) 0.8, insoluble NSP 19, crude protein 22, and 18.6 MJ/kg gross energy (GE), while the MRB diet contained, %, respectively: 0.5, 22, 21, and 18.9 MJ/kg GE. In the performance experiment, lasting 40 days, 60 pigs (30 gilts and 30 castrates) with body weight of  $20.5\pm0.76$  kg were randomly allocated to the six dietary treatments with a completely randomized design (5 gilts and 5 castrates per treatment). Feed and water were provided *ad libitum*.

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The results show there were no significant differences in average daily gain (ADG) (P=0.09), feed gain ratio (F/G; P=0.09), ileal digestibility of nutrients (P=0.09) and N flow (P>0.08) between the MWB and MRB diet, although the crude fibre level in the MRB diet had higher crude fibre content (9.7 g/kg) than that the MWB diet. The dry matter intake (DMI) of the MWB diet was increased (P<0.05) by the enzyme A supplementation. The ADG and F/G of the pigs fed the MWB diet were improved (P<0.05) by either arabinoxylanase or arabinoxylanase plus protease supplementation. The ileal endogenous N excretion determined with enzymatic hydrolysed casein diet in this experiment was half higher than that calculated from the N-free diet. The ileal digestibility of soluble NSP fractions of the MWB and MRB diets were significantly (P<0.05) increased by either enzyme arabinoxylanase or arabinoxylanase or arabinoxylanase or arabinoxylanase or enzyme effect on insoluble NSP fraction digestibility. The results of this study suggest that NSP degrading enzyme do mainly reduce the negative nutritional effect of the soluble NSP in the small intestine of growing pigs.

KEY WORDS: pigs, enzymes, rice bran, wheat bran, digestion, performance

## INTRODUCTION

Rice bran and wheat bran are both widely used in pig diets, especially in Asia, but digestibility and animal performance experiments have shown that inclusion of either of the bran reduces feed intake (Chen, 1981), decreases ileal and overall digestibility (Chen 1979; Graham et al., 1986), and results in poor growth rates (Hanrahan and O'Grady, 1970; Chen, 1981). Recent studies on the cereal shows that plant cell walls contain primarily complex carbohydrates referred to as non-starch polysaccharides (NSP), which can negatively affect nutrient utilization (Englyst and Hudson, 1996). The NSP constituents of the endosperm cell walls of wheat and barley are mainly arabinoxylans and barley also contains high levels of β-glucans (Yin et al., 2000a,b,c; 2001a,b). Many studies have investigated ways of removing these polymers, which encapsulate the desired nutrients in the endosperm. However, feed processing techniques such as extrusion cooking, gamma irradiation, micronization and flaking did not significantly improve nutrient utilization (McClean, 1993).

Xylanase enzyme, in combination with ß-glucanase has been reported to improve hulless barley apparent ileal digestibility of nutrients (Yin et al., 2000a; 2001a,b) and growth performance of young pigs (Baidoo et al., 1998). However, the effects of feed enzyme on nutrient utilization and nitrogen flow through the terminal ileum and excretion in faeces in growing pigs fed rice or wheat bran based diet were seldom reported in the literature. The objects of this study were to determine the effects of carbohydrase and protease supplementation of the maize-rice bran and maize-wheat bran-based diets on apparent and true digestibility of nutrients, nitrogen flow and performance of growing pigs.

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## MATERIAL AND METHODS

## Diets

One butch of wheat bran and one butch of rice bran were used in this experiment. Six diets (Table 1) were prepared: maize-wheat bran diet without feed enzyme supplementation (MWB); MWB supplemented with arabinoxylanase (HuaFan Feed Company, Zhao Qing, Guangzhou, China) 1 g/kg diet (MWB+A); maize-wheat bran-base diet supplemented with arabinoxylanase (1 g/kg diet) plus protease (HuaFan Feed Company, Zhao Qing, Guangzhou, China) 0.2 g/kg diet (MWB+AP); MWB without feed enzyme supplementation (MRB); MRB supplemented with arabinoxylanase, 1 g/kg diet (MRB+A); MRB supplemented with arabinoxylanase (1 g/kg diet) plus protease (1 g/kg diet) plus protease (1 g/kg diet) plus protease (2 g/kg diet) (MRB+AP).

TABLE 1

Composition of experimental diets (maize-wheat bran based diet (MBW), that diet supplemented with arabinoxylanase (MWB+A) or with arabinoxylanase + protease (MWB +AP) and maize-rice bran diet (MRB), that diet supplemented with arabinoxylanase (MWB+A) and arabinoxylanase + protease (MWB +AP), g/kg

Diet	MWB-	MWB	MWB	MRB-	MRB+A	MRB
Diet	IVI W D-	+A	+AP	WIKD-	MKD+A	+AP
Maize	490.0	489.0	488.8	490.0	489.0	488.8
Wheat bran	262.0	262.0	262.0	-	-	-
Soyabean meal	208.7	208.7	208.7	208.7	208.7	208.7
Rapeseed oil	10.5	10.5	10.5	10.5	10.5	10.5
Rice bran	-	-	-	262.0	262.0	262.0
L-lysine HCl	1.70	1.70	1.70	1.70	1.70	1.70
Salt	3.5	3.5	3.5	3.5	3.5	3.5
Limestone	8.6	8.6	8.6	8.6	8.6	8.6
Dicalcium phosphate	12.5	12.5	12.5	12.5	12.5	12.5
Mineral/vitamins1	2.5	2.5	2.5	2.5	2.5	2.5
Arabinoxylanase	-	1	1	-	1	1
Protease	-	-	0.2	-	-	0.2

<sup>1</sup> supplying per kg diet: vit. A, 8000 IU; vit. D<sub>3</sub>, 2000 IU; mg: vit. E, 40; vit., K 1; vit. B<sub>2</sub>, 2; vit. B<sub>12</sub>, 0.012; Cu, 15 (from CuSO4.5H<sub>2</sub>O); Zn, 100 (from ZnO); Fe, 100 (from FeSO<sub>4</sub>.H<sub>2</sub>O); Mn, 20 (from MnO); IO<sub>3</sub> (from Ca (IO<sub>3</sub>)<sub>3</sub>); Se, 0.3 (from Na,SeO<sub>3</sub>)

## Experiment 1. Digestion experiment

Animals and procedures. Six Large White  $\times$  Landrace  $\times$  Chinese Black barrows with an average initial body weight (BW) 21.0±0.9 kg, were surgically fitted with a simple T-cannula at the distal ileum according to the procedure described by Sauer et al. (1983). The cannulas were modified according to de Lange et al. (1989).

After surgery, the pigs were housed individually in stainless steel metabolic crates in a temperature-controlled barn (20 to 22°C). During a 13-d recovery period, the pigs were fed a commercial starter diet containing 18% of protein.

The experiment was designed and conducted according to a  $6 \times 6$  Latin square design. After recovering from surgery, the pigs were offered one of the 5 diets (Table 1, diet MRB + AP was excluded) at 08:00 and 18:00 daily as a wet mash. The sixth diet was of the enzyme hydrolysed casein (EHC) diet which was used for determination of the ileal endogenous nitrogen (N) and amino acid (AA) flow. Chromic oxide was incorporated into all of the diets as an indigestible marker. The EHC diet was formulated with g/kg of: enzyme hydrolysed casein 150, (MW>5000Da, New Zealand Pharmaceuticals Ltd. Palmerston North, New Zealand), rapeseed oil 35, cellulose 50, sucrose 70, maize starch 629 and chromic oxide 2, and vitamin and mineral mixture 64 (for composition see Table 1). Pigs were fed at 2.6 times of their maintenance energy requirement (NRC, 1998) based on their BW at the beginning of each experimental period. Water was added to each meal in a 2:1 ratio. Additional water was available from a nipple drinker.

The experimental period lasted 14 days each. Pigs were allowed to adapt to their respective experimental diets for 7 d and then 3 days of faeces collection. On the  $11^{\text{th}}$  day, ileal digesta samples were collected continuously for 24 h using plastic bags (length, 16 cm; i.d., 10 cm) attached to the cannula. The bag contained approximately 10 ml of a formic acid solution (10%, v/v) to minimize further microbial activity. The bag was removed and replaced as soon as it was partially filled with digesta. Digesta were immediately frozen at -20°C following collection.

## Experiment 2. Performance experiment

Sixty Landrace × Large White × Chinese Black pigs (30 gilts and 30 barrows) with body weight of  $20.5\pm0.76$  kg were randomly allocated to the six dietary treatments (Table 1) with a completely randomised design (5 gilts and 5 barrows per treatment). The pigs were housed individually in floor pens ( $1.2 \text{ m} \times 1.3 \text{ m}$ ) in an environmentally controlled room and at an environmental temperature of at least 18°C. The pens were fitted with flooring made of plastic covered woven iron and stainless steel single-space feeders and nipple drinker; pen partitioning were made of metal bars. Feed and water were provided *ad libitum*.

Individual pig weights and pen feed intake were determined weekly. If feed was dry (not contaminated) the weight was recorded directly. If contamination had occurred the feed was dried and weighed for correction of feed intake. The experimental period lasted 40 days.

## Chemical analysis

After the conclusion of the animal trial, faeces and digesta were freeze-dried, pooled for each within animal and period, ground through a 1.00-mm mesh screen, and mixed before analyses.

For the digesta collected from the EHC diet were thawed and pooled for each pig over the each collection periods. A sub-sample of approximately 100 g was taken from each digesta pool, and centrifuged at 7000 g for 11 min. The supernatant fractions were than ultra filtered using Centriprep YM-10 (Amicon-Millipore, Beyerly, MA, USA; Cat. No. 4305, 10000 Da MM cut-off) according to the manufacture's instructions. The precipitate from the centrifugation step was added to the retentate from the ultra filtration, and the material freeze-dried and finely ground.

Dry matter (DM), gross energy (GE), crude protein (CP, N  $\times$  6.25, macro-Kjeldahl), crude fibre and crude fat contents were determined according to AOAC (1990). Samples were also analysed for chromic oxide (Fenton and Fenton, 1979). Amino acid (AA) contents were determined by ion exchange chromatography following hydrolysis in 6N hydrochloric acid at 110°C for 24 h. Methionine and cysteine were determined as methionine sulphone and cysteic acid after oxidation with performic acid. Soluble and insoluble NSP sugars and uronic acids in diets were determined by gas chromatography (Pye Unicam 304) according to Englyst and Cummings (1984).

## Calculations and statistical analyses

The apparent digestibility of energy, crude protein and AA were calculated as:

$$D_{\rm D}$$
 (%) =100 -  $[I_{\rm D} \times A_{\rm F})/(I_{\rm F} \times A_{\rm D})] \times 100$ 

where  $D_D$  is the apparent digestibility of a nutrient in the diet;  $I_D$  is the marker concentration in the diet;  $A_F$  is the nutrient concentration in faeces or ileal digesta;  $I_F$  is the marker concentration in faeces or ileal digesta; and  $A_D$  is the nutrient concentration in the diet. All variables were expressed in percentages.

The ileal true digestibility (TID) of CP and AA were calculated as:

ITD (%) = 
$$100 \times (1 - [(10 \times AA_{dief}) - [(10 \times AA_{dief}) \times (AA_{AID}/100)] - AA_{E})/(10 \times AA_{dief})]$$

where  $AA_E$  is the gut endogenous AA or N losses expressed as g/kg dry matter intake determined with the EHC or protein free diet reported by CVB (1998) and the  $AA_F$  was calculated as:

 $AA_{E}$  = concentration of compound in digesta × diet Cr concentration/digesta Cr concentration.

## DIETS SUPPLEMENTED WITH ENZYME FOR PIGS

The results of the growth performance were analysed as a completely randomised design, with a  $2 \times 3$  factorial treatment, in which animals were randomly assigned to the MWB or MRB diet, with or without enzyme A or enzyme B supplementation. Differences between means were compared using the Student-Neuman-Keuls (SNK) method at a significance level of P<0.05. Data from the digestion trial were first subjected to analysis of variance (ANOVA). Sources of variation were diets (n=6), periods (n=6) and pigs (n=6). Where appropriate, treatment means were compared using the Student-Newman Keuls' multiple range tests. The ANOVA, comparisons of means, and regression analyses were carried out using the General Linear Model Procedure of SAS Institute, Inc. (1988).

## RESULTS

The proximate analysis of a number of chemical constituents of the wheat bran and rice bran diets, including AA is shown in Table 2. The CP and most AA were slightly higher for the wheat bran diet. Conversely, the crude fibre and insoluble NSP were higher for rice bran diet, although the soluble NSP were lower for the rice bran diet.

The effect of dietary treatments on the pig performance is shown in Table 3. Pigs fed the MWB diet supplemented with either the arabinoxylanase (MWB+A) had a higher feed intake (P<0.05), higher daily gains (P<0.05) and better feed utilization (P<0.05) than pigs fed unsupplemented diet. Additional supplementation of protease (diet MWB+AP) did not improve daily gains or feed utilization, but lowered the feed intake to the MWB diet level. Enzyme treatment did not improved (P>0.05) the performance of pigs fed the MRB diets and there were no differences (P>0.05) in performance between pigs fed the MWB diet and MRB without or enzyme supplemented diets.

The effects of dietary treatments on the total fresh faeces excretion, total and ileal endogenous N excretion is shown in Table 4. Enzyme supplementation reduced (P<0.05) the fresh faeces and total N flow (P<0.05). The endogenous N flow, calculated according the Centraal Veevoeder Bureau (1998) data based on N-free diet, was less than half of endogenous N determined in our experiment using EHC diet.

The digestibility of DM, GE, CP, NSP and AA are given in Tables 5 and 6. Pigs fed the MWB diet supplemented with either arabinoxylanase or arabinoxylanase plus protease had a higher (P<0.05) apparent ileal digestibility of DM, GE, CP and apparent and true ileal digestibility of arginine, histidine, isoleucine, lysine and threonine than the pigs fed the other diets. Enzyme supplementation increased (P<0.05) the ileal digestibility of soluble NSP fractions, however, there were no enzyme effects (P>0.05) on digestibility of insoluble NSP. The

Item	1	Diet		
nem	wheat bran	rice bran		
Dry mater	867	858		
Gross energy, MJ/kg	18.6	18.9		
Crude fibre	49.4	61.8		
Crude fat	54.2	52.4		
Crude protein	221.5	213.3		
Amino acids				
arginine	7.7	7.6		
cystine	3.8	3.6		
histidine	7.5	7.7		
isoleucine	11.6	11.5		
leucine	20.5	19.8		
lysine	12.1	12.1		
methionine	3.2	3.3		
phenylalanine	12.8	12.5		
threonine	7.5	7.2		
valine	9.6	9.2		
Soluble NSP				
rhamnose	0.2	0.1		
fucose	0.2	0.1		
arabinose	1.8	1.2		
xylose	1.3	1.1		
mannose	0.3	0.3		
galactose	2.1	1.2		
glucose	1.8	1.0		
total	7.8	5.1		
Insoluble NSP				
rhamnose	0.5	0.6		
fucose	2.8	3.5		
arabinose	50.0	81.6		
xylose	93.0	104.8		
mannose	1.6	1.8		
galactose	4.1	4.7		
glucose	41.4	47.0		
total	193.5	220.7		

Proximate analysis of maize wheat bran and maize rice bran diets and contents of essential amino acids and non-starch polysaccharide (NSP) contents, g/kg DM

TABLE 2

Performance of pigs (10 animal per treatment, 53 d.f) fed the maize-wheat bran-based diet (MWB), that diet supplemented with arabinoxylanase (MWB+A) or with arabinoxylanase + protease (MWB + AP) and maize-rice bran diet (MRB), that diet supplemented with arabinoxylanase (MWB+A) and arabinoxylanase + protease (MWB + AP)

	1	<b>`</b>								
Enzyme	MW	/B		MRB			SEM		P=	
Indices	-	+A	+AP	-	+A	+AP	bran	enzyme	bran	enzyme
Weight, kg										
initial	19.5	20.5	21.0	21.9	20.3	20.0	0.74	0.91	0.20	0.18
final	48.8	53.9	53.5	50.8	50.3	49.4	1.13	1.20	0.09	0.07
DM intake, kg/d	1.400 <sup>b</sup>	1.498ª	1.399 <sup>b</sup>	1.498 <sup>b</sup>	1.488 <sup>b</sup>	1.423 <sup>b</sup>	0.042	0.033	0.10	0.06
Gain, g/day	733 <sup>b</sup>	835 <sup>a</sup>	813 <sup>a</sup>	723 <sup>b</sup>	750 <sup>b</sup>	735 <sup>b</sup>	22.9	23.0	0.09	0.07
Feed DM gain ratio	1.91ª	1.79 <sup>b</sup>	1.72 <sup>b</sup>	2.07ª	1.98ª	1.94 <sup>b</sup>	0.039	0.040	0.09	0.06

<sup>a,b</sup> means in the same row with different superscripts differ at P<0.05

#### TABLE 4

TABLE 3

Total fresh faeces flow, g/kg dry matter intake, (DMI), and total and ileal endogenous nitrogen (N) flows at the distal ileum of the pigs fed fed the maize-wheat bran-based diet (MWB), that diet supplemented with arabinoxylanase (MWB+A) or with arabinoxylanase + protease (MWB +AP) and maize-rice bran diet (MRB), that diet supplemented with arabinoxylanase (MWB+A) and arabinoxylanase + protease (MWB +AP)

Indices	MWB	MWB	MWB	MRB	MRB	SEM	
Indices	IVI VV D	+A	+AP	MIND	+AP		
Total fresh faeces flow, g/kg DM	1031ª	983 <sup>b</sup>	988 <sup>b</sup>	1053ª	1000 <sup>b</sup>	8.3	
Total N flow, g/kg DM	8.9ª	7.1 <sup>b</sup>	7.2 <sup>b</sup>	9.2ª	8.0 <sup>a b</sup>	0.21	
Ileal endogenous N flow, g/kg DM1	4.4	4.4	4.4	4.4	4.4	0.01	
Ileal endogenous N flow, g/kg DMI <sup>2</sup>	2.0	2.0	2.0	2.0	2.0	0.01	

<sup>1</sup> calculated from the enzyme hydrolysed casein diet (EHC)

<sup>2</sup> calculated from the nitrogen free diet, according to CVB (1998)

<sup>a,b</sup> means in the same row with difference superscripts differ at P<0.05

digestibility of DM, GE, CP and AA were not improved (P<0.05) by addition of arabinoxylanase to the MRB diet. There were no significant differences in total digestibility between the unsupplemented an supplemented MWB and MRB diets.

Value of DE at the ileal (IDE) and total tract (TDE) levels and true ileal digestible CP and essential AA contents are presented in Table 7. Pigs fed the MWB diet supplemented with either the enzymes A or AP had a higher (P<0.05) IDE, CP, arginine, histidine, isoleucine, lysine and threonine than the pigs fed other diets. There were no difference (P<0.05) in DE and true ileal digestible CP and AA contents among the other diets.

Apparent ileal and total digestibility (%) of dry matter, gross energy, crude protein and non-starch polysaccharides (NSP) in pigs fed the maize-wheat bran-based diet (MWB), that diet supplemented with arabinoxylanase (MWB+A) or with arabinoxylanase + protease (MWB +AP) and maize-rice bran diet (MRB), that diet supplemented with arabinoxylanase (MWB+A) and arabinoxylanase + protease (MWB +AP)

protease (MWB +AP)						
Indicies	MWB	MWB+A	MWB+AP	MRB	MRB+A	SEM
Ileal digestibility						
dry matter	61.0 <sup>b</sup>	66.8 <sup>a</sup>	67.0 <sup>a</sup>	59.9 <sup>b</sup>	62.3 <sup>ab</sup>	3.52
energy (GE)	61.8 <sup>b</sup>	66.5 <sup>a</sup>	67.0 <sup>a</sup>	60.9 <sup>b</sup>	63.2 <sup>ab</sup>	2.63
crude protein	69.2 <sup>b</sup>	74.3ª	75.2ª	68.2 <sup>b</sup>	71.8 <sup>ab</sup>	3.61
soluble NSP						
rhamnose	16.3 <sup>b</sup>	24.8 <sup>a</sup>	25.0ª	17.9 <sup>b</sup>	23.8ª	3.03
fucose	14.1 <sup>b</sup>	19.1ª	20.3ª	15.9 <sup>b</sup>	21.7ª	2.90
arabinose	20.3 <sup>b</sup>	26.3ª	25.9ª	19.8 <sup>b</sup>	27.8ª	3.35
xylose	15.9 <sup>b</sup>	20.9ª	21.5ª	16.6 <sup>b</sup>	22.3ª	3.03
mannose	14.8 <sup>b</sup>	20.9ª	22.3ª	15.9 <sup>b</sup>	25.9ª	3.90
galactose	17.5 <sup>b</sup>	23.4ª	22.6ª	16.1 <sup>b</sup>	24.4ª	2.93
glucose	22.0 <sup>b</sup>	28.1ª	29.4ª	23.9 <sup>b</sup>	30.9 <sup>a</sup>	3.00
total	17.2 <sup>b</sup>	23.0ª	23.9ª	18.5 <sup>b</sup>	25.5ª	3.52
insoluble NSP						
rhamnose	11.8	13.4	12.8	13.6	14.4	2.99
fucose	10.9	12.6	12.5	12.2	13.3	3.05
arabinose	15.5	17.0	16.3	16.5	19.8	2.09
xylose	13.3	11.5	14.9	13.0	15.4	2.98
mannose	9.9	10.3	11.8	12.4	14.1	4.34
galactose	13.3	12.2	14.5	13.5	12.0	2.22
glucose	13.4	12.5	12.8	13.3	14.0	3.00
total	12.4	12.3	13.4	13.7	14.6	2.98
Overall digestibility						
dry matter	73.2	75.8	76.0	73.2	75.9	3.43
energy	75.0	78.2	77.3	74.0	74.6	2.59
crude protein	80.8	81.4	82.0	81.2	81.9	3.61
soluble NSP						
rhamnose	54.5	58.6	54.8	57.3	58.3	3.30
fucose	56.6	58.8	57.9	55.9	58.8	2.98
arabinose	60.2	64.5	63.2	59.8	62.2	3.21
xylose	56.3	57.2	56.8	57.3	59.9	3.39
mannose	58.9	59.9	58.7	60.3	62.3	4.20
galactose	61.2	62.3	61.8	62.6	63.9	3.89
glucose	65.9	64.3	66.8	64.8	63.0	3.00
total	59.1	61.2	62.3	59.9	61.2	3.23

continued on the next page

TABLE 5

						TABLE 5
						continued
Indices	MWB	MWB+A	MWB+AP	MRB	MRB+A	SEM
insoluble NSP						
rhamnose	30.2	32.6	29.8	31.8	33.7	3.56
fucose	26.5	26.9	27.9	28.9	30.2	3.98
arabinose	31.6	33.0	32.8	34.0	35.0	4.03
xylose	30.6	31.6	30.2	29.9	30.5	3.89
mannose	28.8	30.6	31.0	30.5	32.9	3.00
galactose	29.9	30.2	32.5	30.0	32.5	3.26
glucose	30.3	32.6	31.8	29.7	32.6	3.80
total	30.0	31.1	31.0	30.8	32.6	3.89

 $^{a,b}$  means in the same row with difference superscripts differ at P<0.05

#### TABLE 6

Ileal apparent and true digestibility (%) of the essential amino acids in pigs fed the maize-wheat branbased diet (MWB), that diet supplemented with arabinoxylanase (MWB+A) or with arabinoxylanase + protease (MWB +AP) and maize-rice bran diet (MRB), that diet supplemented with arabinoxylanase (MWB+A) and arabinoxylanase + protease (MWB +AP)

Indices	MWB	MWB+A	MWB+AP	MRB	MRB+A	SEM
Apparent digestibility						
arginine	75.0 <sup>b</sup>	80.3ª	81.5 <sup>a</sup>	73.3 <sup>b</sup>	75.5 <sup>b</sup>	0.59
cystine	73.1	75.5	74.3	72.4	74.4	0.68
histidine	70.2 <sup>b</sup>	75.3ª	75.5ª	69.3 <sup>b</sup>	71.6 <sup>b</sup>	0.70
isoleucine	80.4 <sup>b</sup>	88.0 <sup>a</sup>	87.4ª	79.5 <sup>b</sup>	81.6 <sup>b</sup>	0.52
leucine	81.2	83.2	84.5	80.4	81.2	0.61
lysine	64.2 <sup>b</sup>	69.3ª	68.8ª	63.6 <sup>b</sup>	64.7 <sup>b</sup>	0.80
methionine	77.0	80.5	79.5	78.5	80.6	0.70
phenylalanine	80.1	82.3	83.4	79.0	80.8	0.66
threonine	65.1 <sup>b</sup>	69.5ª	69.6ª	64.1 <sup>b</sup>	67.7 <sup>b</sup>	0.52
valine	70.3	73.6	74.0	68.4	72.5	0.97
True digestibility <sup>1</sup>						
crude protein	87.0 <sup>b</sup>	92.3ª	93.1ª	86.4 <sup>b</sup>	87.8 <sup>b</sup>	1.0
arginine	90.0 <sup>b</sup>	95.3ª	96.1ª	89.0 <sup>b</sup>	90.5 <sup>b</sup>	0.9
cystine	88.1	90.5	89.9	87.8	90.2	0.8
histidine	84.2 <sup>b</sup>	90.3a	90.9ª	83.9 <sup>b</sup>	86.3 <sup>b</sup>	0.8
isoleucine	89.0 <sup>b</sup>	97.9ª	99.5ª	89.5 <sup>b</sup>	93.3 <sup>b</sup>	0.7
leucine	96.3	98.3	98.9	96.9	98.0	0.8
lysine	87.3 <sup>b</sup>	91.8ª	91.3ª	86.6 <sup>b</sup>	89.1 <sup>ab</sup>	1.0
methionine	83.2	86.5	85.4	84.2	86.3	0.9
phenylalanine	97.0	99.2	99.9	96.6	97.8	0.8
threonine	90.3 <sup>b</sup>	95.4ª	95.3ª	89.3 <sup>b</sup>	94.0 <sup>ab</sup>	1.1
valine	94.8	97.0	98.2	90.6	92.9	0.9

<sup>1</sup> calculated from the enzyme hydrolysed casein based diet method

 $^{a,b}$  means in the same row with difference superscripts differ at P<0.05

#### TABLE 7

Value of digestible energy at ileal (IDE, MJ/kg) and total tract level (TDE, MJ/kg) and ileal true digestible of crude protein and the essential amino acids contents (g/kg dry matter ) of the maize-wheat bran-based diet (MWB), that diet supplemented with arabinoxylanase (MWB+A) or with arabinoxylanase + protease (MWB + AP) and maize-rice bran diet (MRB), that diet supplemented with arabinoxylanas (MWB+A) and arabinoxylanase + protease (MWB + AP)

Indices	MWB	MWB+A	MWB+AP	MRB	MRB+A	SEM
Digestible energy						
ileal	11.5 <sup>b</sup>	125ª	12.5ª	11.5 <sup>b</sup>	11.9 <sup>b</sup>	0.03
total tract	13.9	14.5	14.4	13.9	14.0	0.04
Ileal true digestible						
crude protein	19.30 <sup>b</sup>	20.35 ª	20.62 <sup>a</sup>	18.43 <sup>b</sup>	18.70 <sup>b</sup>	0.04
arginine	6.95 <sup>b</sup>	7.36ª	7.43ª	6.73 <sup>b</sup>	6.85 <sup>b</sup>	0.05
cystine	3.35	3.44	3.43	3.17	3.25	0.06
histidine	6.31 <sup>b</sup>	6.77 <sup>a</sup>	6.81ª	6.45 <sup>b</sup>	6.93 <sup>b</sup>	0.05
isoleucine	10.37 <sup>b</sup>	11.4 <sup>a</sup>	11.60 <sup>a</sup>	10.33 <sup>b</sup>	10.76 <sup>b</sup>	0.05
leucine	19.77	20.18	20.30	19.20	19.42	0.06
lysine	10.57 <sup>b</sup>	11.11ª	11.06 <sup>a</sup>	10.50 <sup>b</sup>	10.80 <sup>b</sup>	0.04
methionine	2.69	2.79	2.76	2.75	2.81	0.07
phenylalanine	12.42	12.70	12.79	12.05	12.20	0.06
threonine	6.77 <sup>b</sup>	7.15 <sup>a</sup>	7.13 <sup>a</sup>	6.45 <sup>b</sup>	6.79 <sup>b</sup>	0.04
valine	9.08	9.29	9.40	8.34	8.55	0.09

<sup>a,b</sup> means in the same row with difference superscripts differ at P<0.05

The effect of enzyme supplementation on hindgut fermentation of nutrients is shown in Table 8. The hindgut fermentation of CP and soluble NSP, expressed either by subtracting overall from ileal digestibility or as a proportion of undigested material reaching the hindgut, were reduced (P<0.05) by enzyme supplementation, but the hindgut fermentation of insoluble NSP was not affected by enzyme treatment and there was no difference (P<0.05) in hindgut fermentation between the MWB diet and the MRB diet.

## DISCUSSION

Although the crude fibre content for the MRB diet was 24% higher than for the MWB diet, ADG, average daily feed intake (ADFI), F/G, DE and ileal true digestible AA for the MRB diet were similar (P=0.08-0.09) to those for the MWB diet. This response may be due to an inadequate number of replicates being conducted. Chen (1981) found that there were no differences in feed to gain or DE to gain ratio between rice bran- and wheat bran diets, when both were added at a rate of 20% into a maize-soyabean meal-based diet for growing-finishing pigs. Yin et al. (1993) reported that

#### TABLE 8

Hindgut disappearance (HF, %) of dry matter, gross energy, crude protein and non-starch polysaccharides (NSP) in pigs fed the maize-wheat bran-based diet (MWB), that diet supplemented with arabinoxylanase (MWB+A) or with arabinoxylanase + protease (MWB + AP) and maize-rice bran diet (MRB), that diet supplemented with arabinoxylanase (MWB+A) and arabinoxylanase + protease (MWB + AP)

Indices	MWB	MWB+AA	MWB+AP	MRB	MRB+A	SEM
Hindgut disappearance <sup>1</sup>						
energy	13.2ª	9.0 <sup>b</sup>	9.0 <sup>b</sup>	13.1ª	11.4a	2.55
crude protein	11.6ª	7.1 <sup>b</sup>	6.8 <sup>b</sup>	13.0ª	9.1 <sup>b</sup>	1.06
soluble NSP						
rhamnose	38.2ª	33.8 <sup>b</sup>	29.8 <sup>b</sup>	39.4ª	34.5 <sup>b</sup>	5.63
fucose	42.5 <sup>b</sup>	39.7 <sup>b</sup>	37.6 <sup>b</sup>	40.0 <sup>b</sup>	37.1 <sup>b</sup>	4.89
arabinose	39.9ª	38.2ª	37.3ª	40.0 <sup>a</sup>	34.4 <sup>b</sup>	6.94
xylose	40.4 <sup>a</sup>	36.3 <sup>b</sup>	35.3 <sup>b</sup>	40.7 <sup>b</sup>	37.6	3.98
mannose	44.1ª	39.0 <sup>b</sup>	36.4 <sup>b</sup>	44.4 <sup>a</sup>	36.4 <sup>b</sup>	6.56
galactose	43.7 <sup>a</sup>	38.9 <sup>b</sup>	39.2 <sup>b</sup>	46.5ª	38.0 <sup>b</sup>	5.98
glucose	43.9ª	36.2 <sup>b</sup>	37.4 <sup>b</sup>	40.9 <sup>a</sup>	32.1 <sup>b</sup>	6.89
total	41.9 <sup>a</sup>	38.2 <sup>ab</sup>	38.4 <sup>ab</sup>	41.4ª	35.7 <sup>b</sup>	5.84
insoluble NSP						
rhamnose	18.4	19.2	17.0	18.2	19.3	5.85
fucose	15.6	14.3	15.4	16.7	16.9	6.12
arabinose	16.1	16.0	16.5	17.5	15.2	4.89
xylose	17.3	20.1	15.3	16.9	15.1	7.45
mannose	18.9	20.3	20.6	19.2	21.3	6.00
galactose	16.6	18.0	18.2	16.5	18.2	5.89
glucose	16.9	20.1	19.0	16.4	18.8	6.02
total	17.6	18.8	17.6	17.1	18.0	5.84
Hindgut disappearance <sup>2</sup>						
energy	34.5ª	26.8 <sup>b</sup>	27.2 <sup>b</sup>	33.5ª	30.1 <sup>ab</sup>	3.89
crude protein	37.7 <sup>a</sup>	27.6 <sup>b</sup>	27.4 <sup>ь</sup>	40.9 <sup>a</sup>	35.1 <sup>b</sup>	2.89
soluble NSP						
rhamnose	45.6 <sup>a</sup>	44.9 <sup>ab</sup>	39.7 <sup>b</sup>	48.0ª	45.2ª	5.02
fucose	49.5	49.0	47.1	47.8	47.0	6.03
arabinose	50.0	51.8	50.3	50.0	47.6	5.55
xylose	48.0	45.9	45.0	48.8	48.3	6.08
mannose	51.8	49.3	46.8	52.8	49.1	5.55
galactose	53.0	50.8	50.6	61.1	50.2	6.23
glucose	56.3	50.3	52.9	58.7	46.4	5.08
total	50.6	49.6	50.4	50.8	47.9	4.98

continued on the next page

						continued
Indices	MWB	MWB+AA	MWB+AP	MRB	MRB+A	SEM
insoluble NSP						
rhamnose	20.9	22.2	23.8	21.1	22.5	5.85
fucose	17.5	16.3	17.6	19.0	18.9	6.51
arabinose	19.0	19.3	20.8	19.8	18.9	5.55
xylose	19.9	22.7	23.6	17.5	21.8	6.08
mannose	21.0	22.3	23.4	21.9	22.7	6.66
galactose	19.1	20.5	21.3	19.1	20.7	5.45
glucose	19.5	23.0	24.1	18.9	19.9	6.48
total	20.0	21.6	20.3	20.8	21.0	5.43

<sup>a,b</sup> means in the same row with difference superscripts differ (P<0.05)

<sup>1</sup> fermentation expressed as the difference between overall and ileal digestibility

<sup>2</sup> fermentation expressed as a proportion of undigested material reaching hindgut

(=Ferm 1 /100 - ileal digestibility)

the nutritional value of rice bran mainly depends on the rice milling procedure. The rice bran used in this trial comes from the rice fine milling process. In this procedure, the outside ring of rice hull was at first dehulled and taken away and the bran come from the second rice milling. The rice bran from the fine milling showed a higher ileal digestibility of crude protein (73%) and total amino acids (74%) than rice bran from rough rice milling (56 and 64%, respectively).

However, both rice bran and wheat bran increase gut fill and also influence other physiological actions such as transit time, nutrient absorption and digestive secretions (Chen, 1979; Graham et al., 1986; Knudsen and Hansen, 1991). Kass et al. (1980) reported that a level of 400 g/kg lucerne meal, resulting in a concentration of 311 g/kg NDF in the diet for growing pigs, significantly affected the daily gain and feed conversion efficiency compared with the control diet (228 g/kg of NDF). The average daily gain was reduced from 700 to 520 g and the feed conversion efficiency increased from 3.23 to 4.91.

In view of the results reported in the literature (Taverner and Campbell, 1988; Van Lunen and Schulze, 1996; Baidoo et al., 1998; Yin et al., 2000a,b), it was expected that inclusion of enzymes would increase the DE and available AA for the MWB-based diet or MRB-based diet. Since the mechanism of such improvement seems to involve breakdown of the cell wall non-starch polysaccharides (NSP) this would have possible benefits in terms of reducing viscosity, as seen in poultry (Bedford and Classen, 1992; Campbell and Bedford, 1992) and to a lesser extent in pigs (Inborr et al., 1993). Van Lunen and Schulze (1996) found a positive effect of xylanase addition on growth rate and F/G, with an improvement of 9.2 and 5.3%, respectively, regardless of the level of wheat and maize inclusion in diets for weaned pigs. In the present study, enzyme supplementation only improved the performance

TABLE 8

for the pigs fed the wheat bran, which had higher soluble NSP fractions than the rice bran. Similar to the performance data, the digestibility of nutrients was improved by the enzyme supplementation only with the wheat bran diet. However, enzyme supplementation improved the soluble NSP fraction digestibility for wheat and rice bran diets, although there were no effects on digestibility of insoluble NSP fractions. These results demonstrated that the enzymes used in this study can only breakdown the soluble NSP fractions of the bran as reported by Marquardt et al. (1994). The reason for the no enzyme effect on the performance and digestion in pigs fed the MRB diet may be that there is less scope for enzyme action, since the total soluble NSP in the MRB diet was much lower than that of the MWB diet (4.38 vs 6.76 g/ kg). This indicated that the response of enzyme mixture supplementation to wheat bran diet appears to result largely from the functions of arabinoxylanase, rather than protease.

However, it is clear from this study that arabinoxylanase addition has reduced faeces and N excretion in faeces and shifted part of digestion from the hindgut to the small intestine, which results in less environmental pollutions and less nutrients available for bacterial fermentation in the hindgut. Undigested soluble fibre may exacerbate such diseases as non-specific colitis (Taylor, 1999) and, more recently, Pluske et al. (1996) have shown that soluble fibre may provoke *Serpulina hyodysenteriae*. Hence, controlling hindgut microbial activity, particularly through nutrimental means, may be of increasing relevance as the use of in-feed antibiotics is more and more constrained.

Recent studies suggest that endogenous gut N losses are higher than previously estimated and that the differences in apparent digestibility between feedstuffs are attributed to difference in endogenous gut N losses rather than to differences in true digestibility (Moughan and Rutherfurd, 1990; Marty et al., 1994). Traditionally, the endogenous loss of N and AA has been determined following protein free alimentation. This method can be criticised, however, due to the physiologically abnormal nature of the protein free state. There are at least two major potential problems with this approach. First, the rate of whole body protein metabolism will decrease when animals are fed a protein free diet (Moughan and Rutherfurd, 1990) and this may lead to a lowered secretion of proteinaceous material into the gut. In addition to this direct effect of the protein-deplete metabolic state, the absence of stimulation from dietary peptides or protein, *per se*, may lead to a reduced secretion of protein into the gut. The present study with the EHC method confirmed these conclusion and therefore it would be useful to determine ileal endogenous AA follow with the EHC method for correcting the apparent ileal digestibility of AA to true ones.

In summary, it can be concluded that the rice and wheat bran used in this study have the similar feeding values for growing pigs. The soluble NSP in the wheat and rice bran diet can be partly hydrolysed in the gastro-intestinal tract of pigs by arabinoxylanase supplementation of feeds.

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### STRESZCZENIE

# Wpływ dodatku arabinoksylanazy i proteazy na wartość pokarmową diet zawierających otręby pszenne lub ryżowe w żywieniu rosnących świń

Przeprowadzono dwa doświadczenia, jedno strawnościowe i jedno wzrostowe, celem zbadania wpływu dodatku enzymów paszowych: arabinoksylanazy lub arabinoksylanazy z proteazą, na strawność składników pokarmowych, przepływ azotu oraz wyniki produkcyjne rosnących świń. Doświadczenie strawnościowe w układzie kwadratu łacińskiego 6×6, przeprowadzono na 6 knurkach mieszańcach (landrasa × wielka biała × chińska czarna), o początkowej m.c. około 20 kg, z kaniulami typu T w końcu jelita biodrowego; oznaczono w nim strawność i przepływ azotu. Zastosowano 6 następujących diet: 1. kukurydza + otręby pszenne (MWB); 2. MWB plus arabinoksylanaza (A); 3. MWB plus arabinoksylanaza i proteaza (AP); 4. kukurydza + otręby ryżowe (MRB); 5. MRB + arabinoksylanaza A; 6. MRB + arabinoksylanaza i proteaza (AP). Dieta MWB zawierała rozpuszczalne nieskrobiowe polisacharydy (NSP), 0,8%, nierozpuszczalne NSP, 19%, białka ogólnego 22%, oraz 18,6 MJ/kg energii brutto (EB), dawka MRB - odpowiednio: 0,5; 22; 21 oraz 18,9 MJ EB. W doświadczeniu wzrostowym 30 maciorek i 30 wieprzków, o początkowej masie ciała 20,5  $\pm$ 0,76 kg, podzielono losowo na 6 grup (po 5 maciorek i 5 wieprzków w każdej) i żywiono do woli takimi samymi dietami jak w doświadczeniu strawnościowym.

Nie stwierdzono istotnych różnic w średnich dziennych przyrostach (P=0,09), wykorzystaniu paszy (P=0,09), jelitowej strawności składników pokarmowych (P=0,09) i przepływie azotu (P>0,08) u świń otrzymujących diety MWB i MRB, chociaż zawartość włókna surowego w diecie MRB była większa o 9,7 g/kg niż w diecie MWB. Pobranie s.m. diety MWB zwiększyło się (P<0,05) po uzupełnieniu jej enzymem A. Dodatek do diety MWB zarówno arabinoksylanazy jak i arabinoksylanazy z proteazą poprawił średnie dzienne przyrosty i wykorzystanie paszy (P<0,05). Wydzielanie N endogennego, oznaczone w jelicie biodrowym przy skarmianiu diety z enzymatycznie hydrolizowaną kazeiną, było o połowę większe niż obliczone przy zastosowaniu diety bezbiałkowej. Jelitowa strawność rozpuszczalnych NSP diet MWB i MRB była istotnie większa (P<0,05) po uzupełnieniu dawek enzymami: arabinoksylanazą oraz arabinoksylanazą i proteazą. Nie stwierdzono wpływu dodatku enzymów na strawność nierozpuszczalnych NSP.

Otrzymane wyniki sugerują, że enzymy rozkładające NSP zmniejszają głównie negatywny antyodżywczy wpływ rozpuszczalnych NSP w jelicie cienkim rosnących świń.