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# Microbial activity in the gut of piglets: II. Effect of fibre source and enzyme supplementation ☆

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**Abstract**

Twenty four Duroc × Landrace male piglets, aged 21 days, were assigned to 1 of 4 experimental diets. Diets 1 and 2 contained 150 g $kg^{-1}$ wheat bran and diets 3 and 4 contained 90 g $kg^{-1}$ maize cobs as the major fibre source. All diets contained 480 g $kg^{-1}$ wheat and 200 g $kg^{-1}$ soybean meal. Diets 2 and 4 were supplemented with the following enzyme complex: 800 U/kg cellulase, 1800 U/kg glucanase and 2600 U/kg xylanase.

The replacement of wheat bran by maize cobs increased the acetic ($P<0.05$) and decreased the butyric acid production ($P<0.05$) in the cecum. Piglets fed diets with maize cobs had lower ($P<0.05$) levels of butyric acid in the colon than those fed wheat bran.

The xylanolytic, pectinolytic and cellulolytic enzyme activities were higher ($P<0.05$) in the cecum and colon of piglets fed the wheat bran based diets. The supplementation of the diet with the enzyme complex did not significantly affect the levels of short chain fatty acids formed in the small intestine and there was a non significant increase of the levels of acetic, propionic and butyric acids in the cecum and colon of piglets ($P<0.10$). No interactions were present between fibre sources and enzyme addition.

The results suggest that the enzyme supplementation of the diet did not bring significant benefits to the animals and that when maize cobs replaces wheat bran in diets it negatively affects butyric acid production and fibre-degrading enzyme activity in the hindgut of piglets.


*Keywords:* Piglets; Fibre; Carbohydrases; Intestinal fermentations

## 1. Introduction

According to Bauer et al. (2001) the fermentability of the different carbohydrate ingredients in the pig diet controls their ability to steer the GI microflora toward greater stability and to improve health conditions of the host. The major end products of these fermentations are lactic acid in the stomach and small intestine and short chain fatty acids (SCFA) in the large intestine (Bach Knudsen et al., 1991).

The physical properties of non-starch polysaccharides (NSP) not only affect the amount but also the proportion of SCFA synthesized, with soluble fibres being more efficiently converted to butyric acid, which improves the epithelial cell proliferation in the small and large intestine (Lizardo et al., 1997). In pig studies the

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Table 1
Composition of the experimental diets (g $kg^{-1}$) and chemical composition (g $kg^{-1}$)

| | Diet | | | |
|---|---|---|---|---|
| | 1 | 2 | 3 | 4 |
| Wheat | 484 | 484 | 481 | 480 |
| Wheat bran | 150 | 150 | – | – |
| Maize cobs | – | – | 90 | 90 |
| Soybean meal | 200 | 200 | 200 | 200 |
| Fish meal | 60 | 60 | 60 | 60 |
| Soybean oil | 70 | 70 | 70 | 70 |
| L-Lysine | 4 | 4 | 5 | 5 |
| DL-Methionine | 1 | 1 | 2 | 2 |
| L-Threonine | 1 | 1 | 2 | 2 |
| L-Tryptophan | – | – | 0.5 | 0.5 |
| Calcium carbonate | 10 | 10 | 10 | 10 |
| Dicalcium phosphate | 5 | 5 | 5 | 5 |
| Salt | 5 | 5 | 5 | 5 |
| Premix | 10 | 10 | 10 | 10 |
| Enzyme complex [1] | – | 0.15 | – | 0.15 |
| *Composition analysis* | | | | |
| DM | 907 | 902 | 913 | 911 |
| Crude protein | 181 | 184 | 178 | 178 |
| Gross energy (MJ $kg^{-1}$) | 17.9 | 17.9 | 18.0 | 18.0 |
| NDF | 134 | 128 | 134 | 130 |
| ADF | 38 | 39 | 55 | 56 |

(1) Contained: 800 U/kg of endo-1,4-β-cellulase, 1800 U/kg of endo-1,3(4)-β-D-glucanase and 2600 U/kg of endo-1,4-β-xylanase.

supplementation with cell wall degrading enzymes did not demonstrate consistent improvements in growth performances and digestibility. According to Diebold et al. (2004) the consumption of diets supplemented with xylanases increased the ileal digestibility of DM but no significant differences were found on digestibility and SCFA at the faecal level. However, Yin et al. (2001) reported an increase in the NSP digestibility at the ileal and faecal levels, due to the supplementation of a wheat middling based diet with xylanase. The response to carbohydrases supplementations is supposed greater with young piglets and with diets rich in fibre. Wheat bran is a traditional source of fibre in piglet diets, but maize cobs may be also utilized. Maize cobs are rich in intrinsic xylo-oligosacharides, mostly as arabinoxylans, that may have a prebiotic effect in the animal. The use of cell wall degrading enzymes may destroy the arabinoxylans present in maize cobs, releasing the endogenous xylo-oligosacharides with consequent improvements on the intestinal fermentative activity.

The aim of the present study is to determine the intestinal fermentative activity in piglets fed a maize cobs or a wheat bran diet and to determine the effect of supplementing these diets with an enzyme complex containing xylanase, glucanase and cellulase.

## 2. Materials and methods

Twenty four Duroc × Landrace male piglets, aged 21 days, were assigned to experimental diets in a per feeding basis. Diets were arranged on a 2 × 2 factorial design. Diets 1 and 2 contained wheat bran and diets 3 and 4 contained maize cobs as the major fibre source (Table 1). Diets 2 and 4 were supplemented with the following enzyme complex: 800 U/kg cellulase, 1800 U/kg glucanase and 2600 U/kg xylanase. The NDF levels were 133.6, 127.6, 134.1 and 130.2 g $kg^{-1}$ DM and the ADF levels were 38.4, 39.1, 55.4 and 55.9 g $kg^{-1}$ DM for diets 1, 2, 3 and 4, respectively. At the end of the experiment, the 42 day-old pigs were slaughtered, the

Table 2
Effect of fibre source and enzyme supplementation on the short chain fatty acids levels (mmol $l^{-1}$) and total enzymatic activity[1] in the gastrointestinal contents of the piglets

| | Fibre | | Enzyme | | F | E | F × E | RSD[2] |
|---|---|---|---|---|---|---|---|---|
| | Wheat bran | Maize cob | – | + | | | | |
| *Small Intestine* | | | | | | | | |
| C2 | 19.6 | 16.6 | 18.6 | 17.7 | NS | NS | 0.206 | 11.4 |
| C3 | 3.9 | 2.7 | 3.7 | 3.0 | NS | NS | 0.559 | 2.1 |
| C4 | 2.7 | 2.0 | 2.5 | 2.2 | NS | NS | 0.245 | 1.6 |
| Total | 26.3 | 21.3 | 24.7 | 22.8 | NS | NS | 0.237 | 14.8 |
| *Cecum* | | | | | | | | |
| C2 | 40.0 | 48.4 | 42.1 | 46.3 | * | NS | 0.773 | 9.2 |
| C3 | 16.2 | 17.5 | 16.3 | 17.4 | NS | NS | 0.335 | 2.9 |
| C4 | 13.1 | 10.3 | 11.2 | 12.2 | * | NS | 0.296 | 3.0 |
| Total | 69.3 | 76.2 | 69.6 | 75.9 | NS | NS | 0.536 | 14.0 |
| *Colon* | | | | | | | | |
| C2 | 48.8 | 48.9 | 45.1 | 52.5 | NS | • | 0.113 | 9.1 |
| C3 | 14.5 | 14.7 | 13.4 | 15.7 | NS | • | 0.075 | 3.2 |
| C4 | 15.9 | 13.0 | 13.9 | 15.0 | * | NS | 0.050 | 3.3 |
| Total | 79.2 | 76.6 | 72.4 | 83.3 | NS | • | 0.061 | 14.2 |
| *Cecum* | | | | | | | | |
| Xylanase | 38.9 | 22.0 | 26.7 | 34.3 | ** | NS | 0.873 | 15.4 |
| Pectinase | 50.3 | 30.7 | 38.1 | 42.9 | * | NS | 0.324 | 19.9 |
| Cellulase | 16.8 | 9.0 | 11.0 | 14.8 | ** | NS | 0.072 | 6.4 |
| *Colon* | | | | | | | | |
| Xylanase | 49.3 | 25.6 | 39.9 | 35.3 | ** | NS | 0.366 | 18.2 |
| Pectinase | 49.1 | 34.0 | 48.3 | 34.7 | NS | NS | 0.109 | 21.6 |
| Cellulase | 16.0 | 7.0 | 12.1 | 11.0 | ** | NS | 0.291 | 4.6 |

(1) mg of sugars released by g of dry matter contents.
(2) F: Fibre, E: enzymes, F × E: interaction fibre source x enzyme addition $^{**}P<0.01$, $^{*}P<0.05$, $\bullet P<0.10$, NS: Not significant, RSD: Residual standard deviation.

contents of the small intestine, cecum and colon were removed and frozen at −20 °C. The SCFA were quantified by gas chromatography and xylanase, cellulase and pectinase activities by spectrophotometry according to the methodologies described by Freire et al. (2003). Data were subject to analysis of variance according to the GLM procedure (SAS, 1991).

## 3. Results

Neither the fibre source nor the addition of the enzyme complex affected the levels (mmol $l^{-1}$) of SCFA that were formed in the small intestine (Table 2). The replacement of wheat bran by maize cobs in the diets increased the acetic acid content by 17% ($P<0.05$) but decreased the butyric acid content by 21% ($P<0.05$) in the cecum. The xylanolytic ($P<0.01$), pectinolytic ($P<0.05$) and cellulolytic ($P<0.01$) activities were higher in the cecum of the piglets fed wheat bran than in the cecum of the piglets fed maize cobs by 43%, 39% and 46%, respectively. The inclusion of the enzyme complex to the diets did not significantly increase ($P>0.05$) the production of SCFA and the xylanolytic, pectinolytic and cellulolytic activities in the cecum. Piglets fed diets with maize cobs as the major fibre source had lower ($P<0.05$) levels of butyric acid in the colon than those fed diets with wheat bran. All other SCFA levels analysed in the colon were not different in piglets fed either diet. As seen previously in the cecum, the replacement of wheat bran by maize cobs in the diet decreased the xylanolytic and cellulolytic activity in the colon by 50% ($P<0.01$). The supplementation of the piglet diet with the enzyme complex tended to increase the levels of acetic acid, propionic acid and total SCFA in the colon ($P<0.10$). With regards to enzyme activity and SCFA production in the hindgut no interactions were present between fibre sources and enzyme addition.

## 4. Discussion

The higher butyrate production in the cecum and colon of piglets fed the wheat bran diets, may be due to the fermentation of the arabinoxylans present in the hemicelluloses of wheat bran. A similar result was found by Bach Knudsen et al. (1991) with oat fibre fractions. Butyrate is important for the normal metabolism, structure and function of the epithelial cells of the large intestine. Therefore, the increased production of butyrate in piglets fed diets containing wheat bran may indicate that this fibre source can be more advantageous for the young pig's intestinal development. The increased levels of acetic acid in the cecum of piglets receiving the maize cobs diets can be caused by the high level of cellulose in these diets, as acetate is produced by the microbial fermentation of cellulose. The supplementation of diets with the enzyme complex had no effect on the SCFA in the cecum and only a numerical increase ($P>0.05$) was found in the colon. These results suggest that the enzyme supplementation did not improve the availability of the fibre sources to the microbial fermentation, in agreement with Diebold et al. (2004), who found no beneficial effect of a xylanase or an enzyme complex on the concentration of SCFA in the faeces of piglets.

The lower microbial enzyme activity in the cecum and in the colon of piglets receiving the maize cobs diets may be due to a high lignin level of the maize cobs, which in turn decreases the access of the microbial enzymes to the substrates. According to Bauer et al. (2001) fibre sources rich in lignin produce less SCFA when incubated in vitro, showing low capacity to be fermented by the microflora collected from the piglet faeces.

In conclusion, wheat bran may be more advantageous in terms of SCFA production and fibre-degrading enzyme activity than maize cobs in the intestine of the young pig and no benefits were found with the enzyme supplementation.



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