

## Short Communication

# Effects of feeding rice and the degree of starch gelatinisation of rice on nutrient digestibility and ileal morphology of young pigs

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We evaluated the influence of the main cereal of the diet and the degree of heat processing of rice on apparent ileal and total tract digestibility of nutrients and ileum morphology of pigs at 37 d of age. Control pigs were fed a complex diet that contained 500 g/kg cooked and flaked maize with a degree of starch gelatinisation (SG) of 840 g/kg. Experimental groups received the same complex diet in which maize was substituted (w/w) by rice with three different degrees of SG; 110, 520 and 760 g/kg that corresponded to raw rice and cooked rice processed under two different set of conditions. The digestibility of dietary components, except for nitrogen, was greater for the cooked rice – than for the raw rice – and the maize diet ( $P < 0.001$ ). Pigs fed cooked rice had higher villus height to crypt depth ratio ( $P < 0.001$ ) and greater percentage of zigzag-oriented villi and lesser percentage of tongue-oriented villi ( $P < 0.01$ ) than pigs fed other diets. Compared with feeding maize, feeding rice improved dietary component digestibility and ileal morphology in piglets. Mild cooking of rice (SG = 520 g/kg) enhanced diet digestibility and ileum morphology. However, processing the rice to increase SG to 760 g/kg did not increase nutrient digestibility further and in fact impaired ileal morphology.

### Rice: Starch gelatinisation: Heat processing: Nutrient digestibility: Ileum morphology: Piglets

Weaning is associated with changes in the structure of the mucosa of the gastrointestinal tract, which impair the ability to digest and absorb nutrients and predispose the young pig to diarrhoea. Starch is the main source of energy of diets and it is almost completely digested in the small intestine. However, starch degradation depends on the structure and size of the granules and, therefore, the type of cereal plays a role on the digestive and metabolic adaptation to weaning. Rice feeding protects pigs against bacterial infections<sup>(1,2)</sup> and improves nutrient digestibility, feed intake and performance with respect to maize feeding<sup>(3,4)</sup>. Heat processing (HP) of cereals increases starch gelatinisation (SG), improving nutrient availability and pig performance<sup>(5,6)</sup>. However, an excess of heat might increase the proportion of resistant starch (RS), reducing nutrient digestibility and piglet growth<sup>(7)</sup>. The present study investigates the influence of the main cereal of the diet and HP of rice on nutrient digestibility and ileal morphology of young pigs.

### Materials and methods

#### Pig husbandry

The experimental procedures used were approved by the Animal Welfare Committee of the University of Madrid<sup>(8)</sup>. Twenty-eight cross-bred piglets weaned at  $21 \pm 2$  d of age

were used. Piglets were housed in individual flat-deck pens (1.1 m × 1.1 m) with free access to feed and water throughout the experiment (21–37 d of age). Room temperature was maintained at  $32 \pm 1^\circ\text{C}$  for the first week and was reduced by approximately  $2^\circ\text{C}$  per week until it reached  $28^\circ\text{C}$ .

#### Diets and experimental design

A batch of broken rice (*Oryza sativa* L., Japonica variety; 800 g/kg Senia and 200 g/kg Tainato cultivars) was obtained from a commercial supplier (Essasa, Valladolid, Spain) and divided into three portions. The first portion was ground through a hammer mill (2.5 mm screen) and used as such (SG = 110 g/kg). The second portion was steam cooked (30 % moisture) in a hydrothermal reactor provided with four bodies (Amandus Kahl, Reinbek, Germany) for 60 min (15 min at  $100^\circ\text{C}$ , 10 min at  $105^\circ\text{C}$  and then settled for 35 min). The third portion was steam cooked (40 % moisture) for 90 min (15 min at  $100^\circ\text{C}$ , 10 min at  $120^\circ\text{C}$  and then settled for 65 min) and flaked through riffled rolls. The degree of SG of these two portions was 520 and 760 g/kg, respectively. The yellow maize (*Zea mays* L.) was steam cooked (30 % moisture) for 60 min (15 min at  $90^\circ\text{C}$ , 10 min at  $117^\circ\text{C}$  and then settled for 35 min). All HP cereals were air-dried at  $150^\circ\text{C}$  for 15 min.

**Abbreviations:** CD, crypt depth; HP, heat processing; RS, resistant starch; SG, starch gelatinisation; VH, villus height.

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The experimental design was completely randomised with four diets and seven individual pigs per diet. There was a control diet based on dried whey, fishmeal, soyabean meal and 500 g/kg cooked and flaked maize and three extra diets in which the maize was substituted (w/w) by rice that is either raw, cooked or cooked and flaked. Celite (acid-washed diatomaceous earth; Ceca, St-Bautizile, France) was added at the rate of 6.0 g/kg to all diets. The composition and the nutritive value of cereals and diets have been reported elsewhere<sup>(7)</sup>.

#### Measurements and post-mortem procedures

Pigs received their experimental diets from 21 and 37 d of age. The last 3 days' representative faecal samples were collected daily by rectal massage from each pig, pooled, frozen ( $-20^{\circ}\text{C}$ ) and stored until chemical analyses. At 37 d of age, the piglets were deprived of food for 14 h, fed *ad libitum* for 2 h and slaughtered<sup>(8)</sup>. Digesta (5 g) was collected from the segment extending from the caecum to 80 cm cranial to the caecum (ileal region), frozen ( $-20^{\circ}\text{C}$ ) and stored until chemical analyses. The data collected were used to determine the apparent total tract and the apparent ileal digestibility of nutrients.

A 5 cm sample from the middle section of the ileum was excised from each pig and placed into a 100 g/kg buffered neutral formaldehyde solution (pH 7.2–7.4). Paraffin-embedded samples were sliced to approximately 5  $\mu\text{m}$ , stained with haematoxylin and eosin and the villus height (VH) and width and crypt depth (CD) and width were measured. The area available for absorption was determined in triplicate cross-sections in all pigs as described by Hampson<sup>(9)</sup>. The height of fifteen vertically oriented villi per replicate with their associated crypts and widths were considered for calculating the average values. In addition, differences in villi orientation and shape were determined comparing the dimensions of the top and the base of the villus. Villi were classified as tongue, finger, leaf and zigzag shaped as described by Van Leeuwen *et al.*<sup>(10)</sup> Histological measurements were conducted under a light microscope (Olympus, B  $\times$  40; Olympus Optical Co., Hamburg, Germany) and images were digitally captured

by computer-assisted image analysis with 40 $\times$  combined magnification (10 $\times$  eyepiece and 4.0 $\times$  objective).

#### Laboratory analysis

Before analysis, ileal and faecal samples were thawed overnight, homogenised, dried ( $60^{\circ}\text{C}$  for 72 h) and ground (1 mm screen; Retsch, Stuttgart, Germany). Ingredients, diets, faeces and ileal contents were analysed for moisture by the oven-drying method, ash by muffle furnace, and crude protein with a Leco nitrogen analyser (FP-528; Leco Corp., St Joseph, MI, USA) as described by AOAC International<sup>(11)</sup>. The gross energy of diets, digesta and faeces was determined with an adiabatic bomb calorimeter (Parr 1356; Parr Instrument Company, Moline, IL, USA) and the content of acid-insoluble ash by the method of Vogtmann *et al.*<sup>(12)</sup>. Starch content was measured according to the method described by Vicente *et al.*<sup>(7)</sup>. The degree of SG of cereals was determined by enzymatic hydrolysis as described by Medel *et al.*<sup>(13)</sup> and the RS by using the Megazyme assay kit (Megazyme International Ireland Ltd, Wicklow, Ireland). The results were expressed as grams of gelatinised starch or RS/kg starch.

#### Statistical analysis

Data were analysed as a completely randomised design with the type of diet as main effect, using the GLM procedure of SAS<sup>(14)</sup>. The Bonferroni test was used to make mean pairwise comparisons.

#### Results

The analysed chemical composition of cereals and diets did not differ from expected values (data not shown). Cooking decreased the RS content of rice from 18.3 to 14.5 g/kg, whereas cooking and flaking increased RS to 19.5 g/kg. The use of cooked rice instead of raw rice or cooked and flaked maize increased the digestibility of all nutrients except crude protein that was not affected (Table 1). Increasing the

**Table 1.** Effect of cereal and heat processing of rice on apparent ileal digestibility (AID) and apparent total tract digestibility (ATTD) (g/kg) of dietary components of pigs at 37 d of age

	Maize Cooked–flaked	Rice			Pooled SEM†	P
		Raw	Cooked	Cooked–flaked		
<b>AID</b>						
DM	871 <sup>b</sup>	872 <sup>b</sup>	886 <sup>a</sup>	881 <sup>a</sup>	2.8	***
OM	868 <sup>c</sup>	875 <sup>b,c</sup>	886 <sup>a</sup>	882 <sup>a,b</sup>	2.7	***
GE	852 <sup>c</sup>	859 <sup>b,c</sup>	871 <sup>a</sup>	864 <sup>a,b</sup>	2.8	***
Starch	973 <sup>c</sup>	983 <sup>b</sup>	994 <sup>a</sup>	987 <sup>a,b</sup>	2.9	***
CP	789	792	794	794	12.1	NS
<b>ATTD</b>						
DM	888 <sup>b</sup>	889 <sup>b</sup>	904 <sup>a</sup>	899 <sup>a</sup>	2.2	***
OM	885 <sup>c</sup>	893 <sup>b,c</sup>	905 <sup>a</sup>	900 <sup>a,b</sup>	2.9	***
GE	869 <sup>c</sup>	877 <sup>b</sup>	885 <sup>a</sup>	882 <sup>a,b</sup>	2.7	***
CP	801	812	817	816	12.5	NS

OM, organic matter; GE, gross energy; CP, crude protein.

<sup>a,b,c</sup> Mean values within a row with unlike superscript letters were significantly different ( $p < 0.05$ ).

\*\*\*  $P < 0.001$ .

† Seven pens of one pig each per treatment.

**Table 2.** Effect of cereal and heat processing of rice on ileal morphology and villus score at 37 d of age

	Maize Cooked–flaked	Rice			Pooled SEM†	P
		Raw	Cooked	Cooked–flaked		
<b>Villus</b>						
Height (µm)	336	402	413	339	30.0	‡
Width (µm)	121	115	106	118	4.7	NS
Area (µm <sup>2</sup> x10)	48.8 <sup>b</sup>	54.2 <sup>a</sup>	56.3 <sup>a</sup>	50.1 <sup>b</sup>	0.63	***
<b>Crypt</b>						
Depth (µm)	152 <sup>a</sup>	143 <sup>a</sup>	110 <sup>b</sup>	148 <sup>a</sup>	9.5	*
Width (µm)	21.3 <sup>a</sup>	20.0 <sup>a</sup>	15.4 <sup>b</sup>	20.8 <sup>a</sup>	0.35	***
Area (µm <sup>2</sup> x10)	4.2	3.9	2.7	3.1	0.75	NS
VH:CD ratio	2.2 <sup>d</sup>	2.9 <sup>b</sup>	4.0 <sup>a</sup>	2.4 <sup>c</sup>	0.29	***
<b>Villus score (%)§</b>						
Tongue	39.2 <sup>a</sup>	28.4 <sup>b</sup>	15.4 <sup>d</sup>	24.2 <sup>c</sup>	4.21	**
Finger	12.2	17.5	25.6	19.6	3.92	‡
Leaf	37.8	34.8	26.8	36.3	4.62	NS
Zigzag	10.6 <sup>c</sup>	19.3 <sup>b</sup>	32.2 <sup>a</sup>	19.5 <sup>b</sup>	3.24	***

VH, villus height; CD, crypt depth.

<sup>a,b,c,d</sup> Mean values within a row with unlike superscript letters were significantly different ( $p < 0.05$ ).

\* $P < 0.05$ , \*\* $P < 0.01$ , \*\*\* $P < 0.001$ .

† Seven pens of one pig each per treatment.

‡  $P < 0.10$ .

§ Percentage of ileum area with presence of tongue-, finger-, leaf- and zigzag-shaped villi.

degree of SG of rice by cooking and flaking did not improve any further the digestibility of dietary components.

Pigs fed raw or cooked rice showed higher ( $P < 0.001$ ) villus area available for nutrient absorption than pigs fed cooked and flaked maize or rice (Table 2). Also, the VH:CD ratio was increased with rice feeding ( $P < 0.001$ ). Cooked and flaked rice reduced the VH:CD ratio as compared to cooked rice ( $P < 0.01$ ). Piglets fed cooked rice had a greater area of the mucosa occupied with zigzag-oriented villis and a lesser area occupied with tongue-shaped-oriented villis ( $P < 0.01$ ) than piglets fed maize, with piglets fed raw rice and cooked and flaked rice being intermediate.

## Discussion

Starch is the main constituent of cereals and, therefore, differences in nutrient digestibility between rice and maize are probably related to differences in starch composition and structure. Starch consists of a mixture of amylose and amylopectin, and it is believed that starch digestibility is inversely related to the amylose content and granule size<sup>(15)</sup>. Therefore, rice starch should be less resistant to enzymatic hydrolysis and more digestible in pigs than maize starch.

HP modifies the chemical structure of the cereal and might improve its nutritive value by facilitating enzymatic degradation. Several authors<sup>(16,17)</sup> have demonstrated that HP improved nutrient digestibility in pigs fed wheat or barley diets. However, the information available on the effects of HP of rice is scarce. In the present trial, HP of rice improved digestibility of most dietary components. By contrast, Li *et al.*<sup>(18)</sup> observed that extrusion did not affect digestibility of most nutrients of rice and in fact reduced apparent ileal digestibility of starch, suggesting that extrusion damaged the structure of components of the rice-based diet. Probably the conditions of the heating process applied in this experiment were higher than required, reducing the availability of rice starch.

Morphological characteristics of the mucosa may provide additional information on the functional status of the digestive

tract. Low feed intake after weaning is likely to result in damaged gut architecture, with deepening of the crypts, shortening of the villi and less area available for nutrient absorption. It has been demonstrated that pigs fed cooked rice ate more feed than pigs fed cooked maize<sup>(3,4)</sup>. The VH:CD ratio, indicator of functional maturity of the enterocytes<sup>(9)</sup>, was increased with cooked rice suggesting an increase in the proportion of mature cells capable of absorbing nutrients. In this regard, Pluske *et al.*<sup>(19)</sup> found a positive correlation between feed intake and VH and, consequently, with post-weaning body-weight gain. By contrast, feeding cooked and flaked rice increased CD and reduced villi area and VH:CD ratio as compared to feeding cooked rice, findings that are often associated with reduced mucosal enzyme activity<sup>(20)</sup>. Therefore, cooking and flaking the rice not only increased SG with respect to cooking rice but also impaired mucosa structure which in turn might reduce nutrient absorption.

The orientation and shape of the villi add information to evaluate properly the functionality of the mucosa. For example, it is known that weaning changes the morphology of the villi from finger-shaped- to tongue-, leaf- and zigzag-oriented villi. Feeding rice tended to increase the percentage of zigzag-oriented villi, whereas the area with tongue-shaped villi was decreased. Therefore, there is a morphological response to feeding rice independent of the effects observed on intestinal characteristics such as villus and crypt dimensions. When the rice was cooked and flaked, the percentage of the area occupied with tongue- and leaf-shaped villi, characteristic of impaired mucosal structure, increased. Also, the area with zigzag-oriented villi decreased, which might impair nutrient digestibility and piglet performance.

## Conclusions

Feeding rice instead of maize increased nutrient digestibility and improved the structure of the ileal mucosa of pigs. The application of a moderate amount of heat to rice improved SG and nutrient digestibility. A further increase in the severity of HP to maximise the degree of SG of rice did not improve further

nutrient digestibility and, in fact, compromised mucosa health at mid-ileum. Therefore, SG is not a good measure of the quality of the process applied to improve digestibility of rice.

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