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# **ABSTRACT:**

The effects of using extruded rice product instead of dried whey on growth performance, CATTD, blood profiles, faecal shedding of Lactobacillus and Escherichia coli (the microbial counts of the digesta were expressed as log10 colony forming units per gram), and faecal scores of weanling pigs were studied in a group of 120 pigs that were 21 days old and had an average initial body weight (BW) of  $6.52 \pm 0.22$  (SE) kg. Each of the four dietary treatments was administered to five pigs in a pen, and there were six copies of each treatment. A diet consisting of 20% dry whey, 20% soybeans, and maize was given to the pigs in the control group. Each group followed the same diet as the other, with the exception that 3%, 6%, and 9% extruded rice was used in lieu of dry whey. No changes were noted in the average daily growth, average daily feed intake, or gain/feed ratio over the six weeks of the trial. Day 14 CATTD was reduced (P < 0.05) in pigs given 6% and 9% extruded rice compared to pigs given the control diet. Pigs in the experimental group had a higher blood creatinine content on Day 14 compared to the control group, when given 9% extruded rice (P < 0.05). The stool E. coli levels on Day 14 were lower in the pigs given diets that included 6% and 9% extruded rice compared to the pigs given the control diet (P < 0.05). This study's findings suggest that weanling pigs fed extruded rice had lower faecal E. coli levels and improved growth performance.

Keywords: microbiota; digestibility; weanling pigs; extruded rice

## INTRODUCTION:

Environmental, nutritional, and disease stressors often induce intestinal barrier dysfunction, digestive disorders, and impaired growth performance (Kim et al. 2012; Yan et al. 2012; Zhao et al. 2012). Several studies have shown that dried whey can improve performance and CATTD of pigs weaned at three to four weeks of age due to the lactose fraction of dried whey (Owsley et al. 1986; Cera et al. 1988; Mahan 1992). However, because of the high cost of dried whey, it would be preferable to find a substitute for it. Rice is one of the most important food crops worldwide (Vandeputte and Delcour 2004). Previous research indicates that feeding cooked rice might protect young pigs against diarrhoea, increase CATTD and

average daily gain (ADG) (Mathews et al. 1999; Pluske et al. 2003; Mateos et al. 2006; Vicente et al. 2008). Theoretically, the extrusion process for cereals could improve ani- mal performance by increased CTTAD of DM, N, GE and growth performance (Miller 1990; Amornthewaphat and Attamangkune 2008). However, Hongtrakul et al. (1998) reported that extrusion of broken rice had no effects on growth performance of weanling pigs. Moreover, the mechanism of the effect of extruded rice on the performance of weanling pigs was not clearly evalu- ated. Rice grain is characterised by its high starch content and low level of non-starch polysaccharide. Also, starch encapsulation is lower for rice and it has a smaller size of starch granules, lower





amylose content and less lipid-amylose complexes. A wide standardisation for *in vitro* starch digestion meth- ods is therefore of crucial importance to provide valuable laboratory tools for rapid assessment of the nutritional value of starch-based feed grains (Giuberti et al. 2014). Therefore, rice starch is ex- pected to be more susceptible to enzymatic action. This study was conducted to evaluate the effects of extruded rice product as a replacement for dried whey in weanling pigs.

Table 1. Ingredient composition and nutrient content of diets (as-fed basis)

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### MATERIAL AND METHODS

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## Vol-12 Issue-02 Aug 2023

**Preparation of extruded rice**. Extruded rice (broken rice) was obtained from a local market (Cheonan, Korea). The composition and amino acid composition are presented in Table 1. **Experimental animal diet and experimental design**. All pigs used in this trial were handled inaccordance with the guidelines set forth by the Animal Care and Use Committee of Dankook University, South Korea. A total of 120 crossbred weanling pigs [(Landrace × Yorkshire) × Duroc] with an initial body weight (BW) of  $6.52 \pm 0.22$ (SEM) kg were used in a 42 days feeding experiment. The pigs were weaned at 21 days and then selected by weight, and were allocated to one of

Item		Phase 1	(0-14 days)			Phase 2	(15-42 day	s)
	CON	NR3	NR6	NR9	CON	NR3	NR6	NR9
Ingredients (%)								
Corn	15.5	15.5	15.5	15.5	15.5	15.5	15.5	15.5
Extruded corn	25.2	25.36	25.47	25.4	27.88	28.15	28.26	28.41
Extruded soybean meal	26.65	26.45	26.25	26.1	24.1	23.8	23.68	23.48
Extruded oats	3	3	3	3	3	3	3	3
Fish meal	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00
Whey powder	20.00	17.00	14.00	11.00	20.00	17.00	14.00	11.00
Rice by-product	_	3.00	6.00	9.00	_	3.00	6.00	9.00
Soybean oil	4.24	4.24	4.32	4.52	4.49	4.24	4.24	4.24
Dicalcium phosphate	0.52	0.42	0.31	0.21	0.45	0.35	0.25	0.17
Limestone	0.57	0.68	0.79	0.88	0.49	0.59	0.68	0.79
Sodium chloride	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Choline chloride	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
dL-methionine,98%	0.07	0.07	0.07	0.07	0.08	0.08	0.08	0.08
L-lysine·HC	0.25	0.28	0.29	0.32	0.26	0.29	0.31	0.33
Vitamin premix <sup>1</sup>	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Mineral premix <sup>2</sup>	0.15	0.15	0.15	0.20	0.20	0.15	0.15	0.15
ME, Mcal/kg	3.40	3.40	3.40	3.40	3.38	3.38	3.38	3.38
CP (%)	21.00	21.00	21.00	21.00	20.00	20.00	20.00	20.00
Lysine (%)	1.35	1.35	1.35	1.34	1.30	1.30	1.30	1.29
Ca (%)	0.80	0.80	0.80	0.80	0.75	0.75	0.75	0.75
Total P (%)	0.65	0.65	0.65	0.65	0.63	0.64	0.63	0.63
Met + Cys (%)	0.74	0.74	0.74	0.74	0.72	0.72	0.72	0.72
СР	21.13	21.09	21.30	21.25	19.99	20.05	20.15	20.15
Ca	0.81	0.79	0.83	0.80	0.74	0.76	0.74	0.75



CON = 20% dried whey; NR3 = 17% dried whey, 3% extruded rice; NR6 = 14% dried whey, 6% extruded rice; NR9 = 11% dried whey, 9% extruded rice

<sup>1</sup>provided per kilogram of diet: vitamin A, 11 025 IU; vitamin D<sub>3</sub>, 1103 IU; vitamin E, 44 IU; vitamin K<sub>3</sub>, 4.4 mg; thiamin, 4 mg; riboflavin, 8.8 mg; vitamin B<sub>12</sub>, 33  $\mu$ g; niacin, 50 mg; pantothenic acid, 29 mg; choline, 166 mg

<sup>2</sup>provided per kilogram of diet: Zn (as  $ZnSO_4$ ), 85 mg; Mn (MnO<sub>2</sub>), 8 mg; Cu (as  $CuSO_4 \cdot 5 H_2O$ ), 12 mg; I (as KI), 0.28 mg; and Se (as Na<sub>2</sub>SeO<sub>3</sub>  $\cdot 5 H_2O$ ), 0.15 mg

Table 2. The nutritional composition of the rice by-prod- uct (as-fed basis)

Composition	(%)
Crude Protein	17.02
Crude Fat	8.03
Crude Ash	6.89
Ca 0.20	
P 1.74	
Lysine 0.75	
Methionine	0.30
Threonine	0.60
Isoleucine	0.44
Arginine	0.78

the four treatments using a completely randomised block design. All treatment groups had a similar ratio of males and females. The experimental treat-resentative faecal samples were collected by rectal massage from at least three piglets from each pen, pooled and frozen (-20 °C), and stored until analy- sis. Diet and faecal samples were dried in an oven (60 °C; 24 h for diet and 48 h for faeces) and then homogenised with a laboratory grinder (0.5 mm screen for diet and 1.0 mm screen for faeces) before analysis. Dietary and faecal dry matter ments were as follows: (1) CON, 20% dried (2) NR3, 17% dried whey, 3% extruded rice; (3) NR6,

14% dried whey, 6% extruded rice; (4) NR9, 11% dried whey, 9% extruded rice (as-fed basis; Table 2). The extruded rice was accompanied at a through- put of 510 kg/h and an average exit temperature of 120 °C in an Insta-Pro<sup>TM</sup> (Triple F, IA, USA) dry- extruder); the rice had had increased ADG, G/F and apparent digestibility of CP and energy compared to corn. The pigs were housed in an environmen- tally controlled nursery room. The stainless steel pens were 0.5  $\times$  0.6  $\times$  2.0 m and had a slatted plas- tic floor. Each pen was provided with a stainless steel feeder and one nipple drinker that allowed for *ad libitum* access to feed and water through(DM), crude protein (CP) and gross energy (GE) content were analysed according to AOAC guidelines (1997). Chromium content was measured with UV ab- sorption spectrophotometry (Shimadzu, UV-1201, Japan). The CATTD of DM, CP, and GE was de- termined using the following indicator method of Sauer and de Lange (1992):  $ADF = 1-[(Cr_2O_3D \times NF)/(Cr_2O_3F \times ND)]$ where:

Cr O D = chromic oxide concentration in the assay diet (g/kg)

whey; 2 3

out the experiment. Ventilation was provided by a mechanical system, and lighting was automatically regulated to provide 12 h of artificial light per day. The ambient temperature within the room was ap- proximately 30 °C, and it was decreased by 2 °C each week until it reached 26 °C. The care and use protocol was approved by the Animal Care and Use Committee of Dankook University. Sampling and measurements. The pigs were weighed individually at the beginning of the experi- ment and at Days 14 and 42 and ADG was calculat- ed per replicate. Pigs were given food to avoid feed wastage, and average daily feed intake (ADFI) and gain/feed ratio (G/F) were calculated. Two grams chromium oxide

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 $(Cr_2O_2)/kg$  were added to the diets as an

indigestible marker from Day 7 to Day 14 and from Day 35 to Day 42. At 14 and 42 days of age, **rep-NF** = concentration of a nutrient in the faeces (g/kg) $Cr_2O_2F$  = concentration of chromic oxide in the faeces (g/kg) ND = concentration of a nutrient in the assay diet (g/kg) At the beginning of the experiment, two pigs were selected at random from each pen (n = 48) and blood samples were collected via jugular venipuncture. The same pigs were bled again on Day 14 and Day 42. At each collection time, the blood samples were col-lected into a nonheparinised vacutainer (Becton Dickinson Vacutainer Systems, Franklin Lakes, NJ) to enable evaluation of creatinine, blood urea nitrogen (BUN), total protein, and albumin. The serum samples were then centrifuged  $(2000 \times g)$  for 30 min at 4 °C, after which creatinine, BUN, total protein, and albumin levels were determined using an auto- matic biochemistry analyser (HITACHI 747, Japan). On Days 0, 7, 14 and 21, faecal samples were col-lected from the rectum with sterile rubber gloves and place in sterile plastic tubes with lids. The samples were stored in a freezer at -20 °C until analysis for Lactobacillus and Escherichia coli (the microbial counts of digesta were expressed as log<sub>10</sub> colony forming units per gram). In vitro survival of Lactobacillus and E. coli was determined accord- ing to the methods of Mikkelsen et al. (2003) with certain modifications. In brief, before enumera- tion, frozen faecal samples were incubated at 4 °C for 10 h. Thereafter, 1 g of digesta was taken from each sample and serially diluted 10-fold with ster-ile

digesta was taken from each sample and serially diluted 10-fold with ster- ile physiological saline, resulting in dilutions ranging from 10-1 to 10-8 for enumeration. *E. coli* was Vol-12 Issue-02 Aug 2023



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# Vol-12 Issue-02 Aug 2023

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Table 3. Effect of extruded rice product on growth performance in weanling pigs

Items	CON	NR3	NR6	NR9	SEM	P-value
Day 0 to 14						
Average daily gain (g)	254	252	252	241	7	0.65
Average daily feed intake (g)	458	423	406	404	18	0.32
Gain/feed	0.555	0.596	0.621	0.597	0.024	0.85
Day 14 to 42						
Average daily gain (g)	526	524	538	544	12	0.58
Average daily feed intake (g)	865	828	845	903	39	0.08
Gain/feed	0.608	0.633	0.637	0.602	0.024	0.12
Day 0 to 42						
Average daily gain (g)	436	433	443	443	9	0.24
Average daily feed intake (g)	729	693	698	737	28	0.09
Gain/feed	0.598	0.625	0.635	0.601	0.020	0.15

CON = 20% dried whey; NR3 = 17% dried whey, 3% extruded rice; NR6 = 14% dried whey, 6% extruded rice; cultivated on MacConkey agar. *Lactobacillus* was cultured on MRS agar. Each dilution was deter- mined in triplicate and the result was the average of three replicated experiments. *Lactobacillus* was inoculated into Hungate roll tubes, and *E. coli* was grown on plates. All tubes and plates were incu- bated at 37 °C for 36 h. The microbial enumerations of digesta were expressed as  $\log_{10}$  colony-forming units per gram. Bacteria were enumerated by a vi- sual count of colonies using the best replicate set from dilutions that resulted in 30 to 300 colonies per plate or tube.

Diarrhoea incidence (DI) was estimated by pen as the number of days in which pigs showed clinical signs of diarrhoea symptoms as a proportion of the total number of days of the trial. The evaluationstandard was as follows: 1 = hard, dry pellet, 2 = firm, formed faeces, 3 = soft, moist faeces that re- tains shape, 4 = soft, unformed faeces that assume the shape of the container, and 5 = watery liquid that can be poured (Hu et al. 2012).

**Statistical analyses**. Data were subjected to ANOVA, with dietary treatment as the classifica- tion factor, using the GLM procedure (SAS Inst., Inc., Cary, NC, USA). The experimental unit was the pen. Before carrying out statistical analysis of the microbial counts, logarithmic conversion of the data was

NR9 = 11% dried whey, 9% extruded rice; SEM = standard error mean

performed. Differences among all treat- ments were separated by Duncan's multiple range test. Variability in data was expressed as the pooled standard error (SE) and a probability level of P <

0.05 was considered statistically significant.

Items	CON	NR3	NR6	NR9	SEM	P-value	
Day 14							
Dry matter	0.812 <sup>a</sup>	0.807 <sup>ab</sup>	0.797 <sup>ab</sup>	$0.788^{b}$	0.5	0.04	
Crude protein	$0.788^{a}$	0.765 <sup>ab</sup>	0.754 <sup>ab</sup>	0.723 <sup>b</sup>	0.9	0.03	
Gross energy	0.881ª	0.852 <sup>ab</sup>	0.848 <sup>ab</sup>	0.802 <sup>b</sup>	1.2	0.03	
Day 42							
Dry matter	0.831	0.841	0.843	0.851	2.2	0.09	
Crude protein	0.803	0.821	0.815	0.824	1.4	0.23	
Gross energy	0.900	0.912	0.920	0.917	1.0	0.31	

Table 4. Effect of extruded rice products as alternative to dried whey on CATTD of DM, CP and weanling pigs

CON = 20% dried whey; NR3 = 17% dried whey, 3% extruded rice; NR6 = 14% dried whey, 6% extruded rice; NR9 = 11% dried whey, 9% extruded rice; SEM = standard error mean

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

RESULTS

## Growth performance and CATTD of DM, CP and GE

ADG, ADFI, and G/F were not affected by the dietary extruded rice level during any of the periods of the experiment (Table 3). Effects of extruded rice on the CATTD of DM, CP, and GE are presented in Table 4. On Day 14, pigs in the NR9 group had a lower (P < 0.05) CTTAD of DM, CP, and GE than pigs in the CON group. However, no differences were observed on Day 42.

## Blood characteristics

Total protein, albumin, blood urea nitrogen con- centrations were not influenced by dietary treat- ment throughout the entire experimental period, (Table 5). However, on Day 14, the creatinine concentration of pigs in the NR9 group was higher (P < 0.05) than that of pigs in the CON group.

# Faecal shedding of Lactobacillus and E. coli,

and incidence of diarrhoea

*Lactobacillus* and *E. coli* in faeces of pigs receiv- ing each treatment are shown in Table 6. Pigs fed6% and 9% extruded rice had lower (P < 0.05) *E. coli* counts than pigs that received the CON treatment on Day 14. No differences (P > 0.05) between groups were observed in *Lactobacillus* and *E. coli* counts on Day 42. No mortality occurred during the trial. Pigs fed extruded rice had lower diarrhoea inci- dence than pigs fed the CON diet, but the results were not statistically significant.

### DISCUSSION

In the current study, growth performance was not affected by extruded rice, which was similar to the results reported by Gomez and Valdivieso (1983). However, Alcantara et al. (1989) reported that pigs fed 20% rough rice in place of corn had increased ADG, gain/feed ratio (G/F), and appar- ent digestibility of CP and energy. The difference in results might be due to the components used as the substitute. Alcantara et al. (1989) used ex- truded rice to replace corn, while we used ex- truded rice to replace dried whey. In our study, the CTTAD of DM, CP, and GE in pigs of the NR9 group were lower than in pigs of the CON group on Day 14. However, on Day 42 and in the overall experiment, no differences were observed. This indicates that the lower CTTAD for DM, CP, and Table 5. Effect of extruded rice products as alternative to dried whey on blood biochemical profiles in weanling pigs

Items (mg/dl)	CON	NR3	NR6	NR9	SEM	P-value	
Total protein							
Day 0	5.02	5.17	5.33	5.05	0.17	0.23	
Day 14	4.92	4.82	5.12	4.80	0.23	0.16	
Day 42	5.48	5.73	6.02	5.57	0.27	0.35	

Albumin						
Day 0	3.45	3.40	3.33	3.33	0.10	0.15
Day 14	3.00	2.80	2.93	2.82	0.08	0.45
Day 42	3.48	3.30	3.50	3.33	0.10	0.13
Creatinine						
Day 0	1.13 <sup>b</sup>	1.22 <sup>ab</sup>	1.18 <sup>ab</sup>	1.23 <sup>a</sup>	0.03	0.04
Day 14	1.00	1.02	0.97	1.00	0.03	0.52
Day 42	1.37	1.22	1.20	1.22	0.58	0.36
Blood urea nitrogen						
Day 0	20.47	16.03	14.75	14.73	1.87	0.08
Day 14	9.63	9.23	10.52	10.02	0.94	0.14
Day 42	11.63	12.42	11.05	13.12	1.17	0.35

 $\overline{\text{CON}} = 20\%$  dried whey; NR3 = 17% dried whey, 3% extruded rice; NR6 = 14% dried whey, 6% extruded rice; NR9 = 11% dried whey, 9% extruded rice; SEM = standard error mean

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

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Table 6. Effect of extruded rice products as alternative to dried whey on faecal microorganisms in weanling pigs

Items (log <sub>10</sub> cfu/g)	NR3	NR6	NR9	SEM	<i>P</i> -value
Day 14	C 20ab	< 10h	c 02h	0.00	0.04
E. COll	6.29 <sup>ab</sup>	<b>6.</b> 10 <sup>8</sup>	6.025	0.06	0.04
Lactobacillus	6.88	7.02	6.95	0.22	0.35
Day 42					
E. coli	7.78	8.20	8.12	0.35	018
Lactobacillus	9.45	9.81	9.77	0.47	0.12

 $\overline{\text{CON}} = 20\%$  dried whey; NR3 = 17% dried whey, 3% extruded rice; NR6 = 14% dried whey, 6% extruded rice; NR9 = 11% dried whey, 9% extruded rice; SEM = standard error mean

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

GE on Day 14 might be caused by the adaptation of the weanling pigs to the diets. Prior to weaning, piglets depend on the sow's milk, which contains a high protein concentration. Hence, the weanling pigs need time to adapt to the diets. Rice grain is characterised by its high starch content and low level of non-starch polysaccharides (Choct 2002). Also, starch encapsulation is lower for rice and it has a smaller size of starch granules, lower amylose content and less lipid-amylose complex- es. Therefore, rice starch is expected to be more susceptible to enzymatic action (Giuberti et al. 2014). Compared with dried whey, rice has a lower protein concentration. The heat processing of rice did not seem to affect the growth performance in the present study.

The lower incidence of diarrhoea documented here might be due to fewer viable *E. coli* in the intestine. Significant reductions in the coliform bacteria count and the incidence of diarrhoea have been described when non-starch polysaccharides are included in low fibre diets (Mateos et al. 2006). Increasing the dietary nonstarch polysaccharides may reinforce the dietrelated microbiota in the hindgut by increasing carbohydrate fermentation, instead of protein (Williams et al. 2001). In con- clusion, weanling pigs fed extruded rice in place of dried whey did not show altered growth performance. The extruded rice can lower viable *E. coli* counts in the intestine and decrease the incidence of diarrhoea. We conclude that extruded rice can be used as a substitute for dried whey in the wean- ling pig diet. Thus, we conclude that extruded rice is an ingredient of choice in pre-starter diets for weaning pigs. Also, its inclusion at moder- ate levels reduces the incidence of diarrhoea in weaning diets.

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