Effect of Dietary Fiber Source on the Growth Performance and Intestinal Microflora in Piglets

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ABSTRACT

Growth performance and the levels and enzymatic activity of intestinal lactobacilli were studied in weaned piglets fed for 4 weeks with basal diets containing different sources of dietary fiber. These included:

1. 50% corn and 50% rice bran (T1)
2. 100% rice bran (T2)
3. 50% corn and 50% sawdust (T3)
4. 100% sawdust (T4), and
5. 100% corn used as control (T5).

Most groups showed similar body weight (BW) gain as the control (T5) group, whereas T4 animals showed significantly lower ($P < 0.05$) BW gain than T5, which was related to the lowest feed intake in T4. While the blood chemistry profiles were similar in most groups, T4 animals had a higher (statistically non significant) serum levels of blood urea nitrogen, aspartate aminotransferase and ala¬nine aminotransferase. T2 and T3 groups had the highest ($P < 0.05$) number of anaerobic bacteria in the feces and intestines compared to T5, while no differences were observed between T4 and T5. Initial screening for pH and bile acid resistance resulted in 23 strains of intestinal lactic acid bacteria, mostly from T3 and T4 groups, of which 61% produced one or more dietary enzymes, including protease, cellulase, phytase, and α- amylase. Although high level of sawdust in piglet diets negatively affects growth and health performance, if mixed with other sources of fiber, it may

Table 1. Chemical composition of the five dietary treatments in piglets

Ingredients	Supplementation levels (% of diet)				
	T1	T2	T3	T4	T5
Corn	29.26	0	29.26	0	58.52
soybean oil meal 45%	30.3	30.3	30.3	30.3	30.3
Raw-rice bran	29.26	58.52	0	0	0
Sawdust	0	0	29.26	58.52	0
Ground limestone	0.62	0.62	0.62	0.62	0.62
Dicalcium phosphate	1.6	1.6	1.6	1.6	1.6
Salt	0.3	0.3	0.3	0.3	0.3
Animal fat	4	4	4	4	4
Molasses	4	4	4	4	4
Lysine-98%	0.2	0.2	0.2	0.2	0.2
Methionin-90%	0.13	0.13	0.13	0.13	0.13
Cholin chloride	0.05	0.05	0.05	0.05	0.05
Sugar	0.03	0.03	0.03	0.03	0.03
Vitamin-mix	0.15	0.15	0.15	0.15	0.15
Mineral-mix	0.1	0.1	0.1	0.1	0.1
Total	**100**	**100**	**100**	**100**	**100**

offer beneficial and cheaper alternative source of dietary fiber that ensures acceptable growth performance and promotes intestinal microflora with beneficial digestive enzymes.

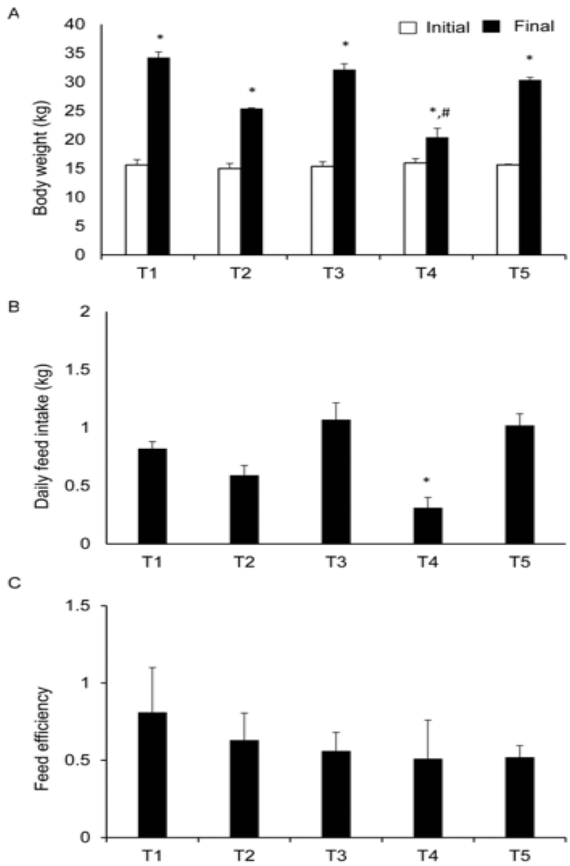
INTRODUCTION

Dietary fiber in swine diets plays several important roles. Maintaining normal physiological state needs a minimum level of dietary fiber in the digestive canal.[15] Fibrous material, primarily cellulose, is degraded to volatile fatty acids by microbial enzymes in the intestine, and these volatile fatty acids absorbed by the animal may contribute up to 30% of the maintenance energy needs of pigs.[2,3,14] Furthermore, cellulolytic bacteria are increased in the large intestines of pigs fed high fiber diets1 and dietary fibers can also have prebiotic effects in pigs due to interactions with the gut micro-environment and gut-associated immune system.[5]

Despite several benefits of dietary fiber, the inclusion of high levels of fiber in the diet can also lead to a reduction in feed intake, impaired growth performance, and higher risks of intestinal disease, especially in immature piglets that lack digestive enzymes and microflora to utilize digestive fibers.[10] However, the effects of dietary fiber on productive and health performance of piglets are dependent on the fiber properties, and may differ considerably between fiber sources.[5]

Several sources of dietary fiber have been used in the global swine production. These include common sources such as corn and soybean meal, cereals such as wheat, and byproducts, such as rice bran and distiller's grains. However, as the demand for cereal grains for human use increases, the availability of the conventional fibrous feeds for swine will most likely be limited, placing an economic burden on swine producers, which will in turn force them to utilize alternative and non-traditional animal feeds. The objective of this study was to evalu-

Figure 1. *Body weight gain (A), feed intake (B) and feed efficiency (C) of piglets fed for 4 weeks with different experimental diets. T1, 50% corn and 50% rice bran; T2, 100% rice bran; T3, 50% corn and 50% sawdust; T4, 100% sawdust; T5, 100% corn. For Body weight:* significant difference (P< 0.05) form initial weight, # significant difference from control (T5) (P< 0.05). For daily feed intake: *significant difference from control (T5) (P< 0.05).*



ate the effect of diets containing different sources of dietary fiber on the growth and health performance, as well as on the levels and enzymatic activity of intestinal lactobacilli, in piglets.

MATERIALS AND METHODS

Animals and Dietary Treatments

Weaned piglets (Landrace × Large White × Duroc) were provided by Doongji farm (Gunwi, Korea). Animals were acclimated for 2 weeks before the start of the feed trials, during which they were fed to 3% of body weight twice a day with a fish diet without antibiotics. In a 4-week study, 20 piglets (15.5 ± 1.5 kg) were randomly assigned to one of five dietary treatments. All animals were housed in clean rooms within pens (2 m × 0.8 m) with plasticcovers and an expanded plastic floor. The five dietary treatments presented in Table 1 contain:

1. corn 50% plus rice bran 50% (T1)
2. rice bran 100% (T2)
3. corn 50% plus sawdust 50% (T3)
4. sawdust 100% (T4)
5. corn 100% (T5 used as control)

Experimental diets were mixed by fodder mixer with regular fodder (KC Feed, Yeongcheon, Korea). All animals were fed the experimental feeds described above for 4 weeks. Water was provided ad libitum by nipple-waterers.

The composition of the five experimental diets is presented in Table 1. These diets provided 19% crude protein, 6.6% Ether extract, 3.4 Kcal/kg metabolizable energy, 0.7% calcium, 0.6% phosphorous, 1.1% lysine, 0.4% methionine, 16,000 IU vitamin A, and 66 IU vitamin E. The chemical composition of experimental diets, determined by Scientec Lab Center (Daejeon, Korea), is given in Table 2.

Feed Consumption and Body Weight Gain

An initial body weight was taken at the beginning of the experiment, with subsequent pig body weights and feed disappearance measurements obtained once per week until the end of the experiment period. Body weights and feed intake were used to determine the weight gain, daily feed intake, and feed efficiency.

Table 2. Determined chemical composition of the fiber sources (g/kg as fed basis)

Dry matter	736.4	773.1	763.1	768.8	737
Crude protein	58.5	51.8	32.9	19.3	62.8
Crude fat	48.1	32.5	13.8	8.6	36.6
Crude fiber	31.8	32.2	110.8	129.6	28.1
Crude ash	58.9	54.1	21.6	16.2	45.9

Blood Biochemistry

To determine if feeding different sources of dietary fiber was associated with abnormal biochemistry profile, blood samples were drawn from piglets in each group, and serum biochemistry analyses were performed as described previously.[8] Briefly, animals were starved for 3 h, and blood samples were collected from the jugular vein into vacutainer tubes (BD, USA). Serum was extracted by centrifugation for 10 min at 2000 × g and analyzed by using an autoanalyzer (Shimadzu CL-7200, Shimadzu Co., Japan), according to the manufacturer's instructions. The blood parameters measured included albumin (ALB), alkaline phosphatase (ALKP), alanine aminotransferase (ALT), amylase (AMYL), BUN (blood urea nitrogen), calcium (Ca), cholesterol (CHOL), creatinine (CREA), glucose (GLU), phosphorus (P), total bilirubin (TBIL), total protein (TP), and globulin (GLOB).

Quantification of Fecal and Intestinal Bacteria

A previously reported method[4] was followed for collection and processing of fecal and intestinal samples for quantification of total anaerobic bacteria. Briefly, fecal samples were collected by using cotton swabs and immediately sealed up anaerobically. The upper duodenum and lower colon of slaughtered animals were ligated aseptically and transported in bags of dry ice. Each fecal and intestinal sample weighing about 1 g was serially diluted. A 0.1 ml of the appropriate dilutions was placed on the surface of blood agar (Komed, Sungnam, Korea). After anaerobic cultivation at 37°C for 24-48 h, the number of colonies in each sample was counted.

Selection of Lactobacilli Producing Dietary Enzymes

A two-step selection method described in our previous study [4] was followed to isolate lactobacilli producing dietary enzymes, including, α-amylase, lipase, phytase, and protease. Briefly, the first step involved screening for acid and bile tolerance of the intestinal and fecal isolates from pigs under the five dietary treatments. Then, a total of 23 strains isolated in the first screening test were examined for their potential in producing the dietary enzymes, following reported methods[4] and using screening media and dyes presented in Table 3.

Statistical Analysis

Table 3. Screening media and dyes used for the detection of various enzyme activities

Enzyme	Substrate (basal medium : MRS agar)	Dye	Identification
Protease	2% (w/v) skim milk		Clear zone
Cellulase (CMCase)	1% (w/v) Carboxymethyl cellulose salt	0.2% (w/v) congo red sol.	Clear zone
Cellulase	1% (w/v) Methyl cellulose	0.85% Nacl	Clear zone
Phytase	1% (w/v) Ca-phytate		Clear zone
α-Amylase	1% (w/v) soluble starch		Clear zone

Table 4. *Enzyme activities of intestinal lactobacilli of piglets*

Strain No.	Substrate (Enzyme activity at 37°C)				
	Skim milk (protease)	Methyl cellulose (cellulase)	Corn starch (α-amylase)	Ca-phytate (phytase)	CMC (Carboxymethyl cellulose)
T1-1	-	-	-	-	-
T1-2	-	-	-	-	-
T1-3	-	-	-	-	-
T2-1	-	-	-	-	-
T2-2	-	-	-	-	-
T2-3	-	-	-	-	-
T3-1	-	-	-	+	-
T3-2	-	-	+	+	+
T3-3	+	-	+	+	+
T3-4	-	-	+	+	+
T3-5	+	-	-	+	+
T3-6	-	-	+	+	+
T3-7	++	-	+	+	+
T3-8	+	-	+	+	-
T4-1	-	-	-	-	-
T4-2	-	-	+	+	+
T4-3	-	-	-	+	-
T4-4	-	-	-	+	-
T4-5	++	-	-	+	-
T4-6	-	-	-	+	-
T4-7	-	-	-	+	+
T5-1	-	-	-	-	-
T5-2	-	-	-	-	-

Results are expressed as the mean and standard error of the mean (SEM) of each treatment group. Analysis of variance (ANOVA) and the Duncan's multiple range tests were used to determine significant differences among different treatments. All statistics were performed using SAS (SAS 9.3 Inst. Inc.).

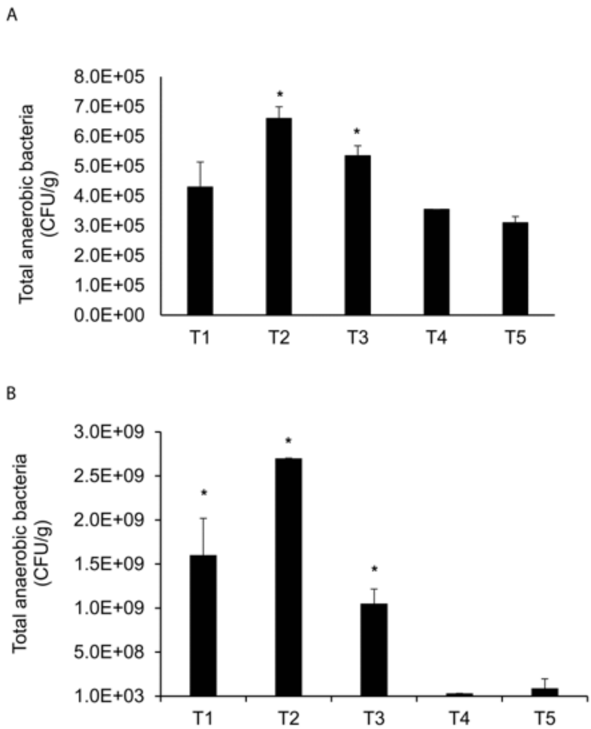
RESULTS AND DISCUSSION

Growth Performance

Body weight (BW) gain, daily feed intake, and feed efficiency of piglets during the 4-wk experimental period are presented in Figure 1. The average body weight (BW) of piglets in each experimental group was about 15 kg at the start of the experiment. At the end of the 4-wk period, T1 (corn 50%, rice bran 50%) and T3 (corn 50%, sawdust 50%) groups showed similar increases in BW as T5 (corn 100%) group, whereas T4 (sawdust 100%) group showed significantly lower ($P < 0.05$) BW gain than T5 group. T2 (rice bran 100%) group also had lower (but not significant) BW gain compared to T5 group. Similar BW gain pattern in the T1, T3, and T5 groups was corroborated by the findings that they had similar daily feed intake (Figure 1B). Also, the lowest BW gain in T4 group was related to the lowest feed consumption observed in this group. No differences were observed in the feed efficiency of the experimental groups (Figure 1C).

These findings suggest that growth performance is hampered by feeding piglets with 100 % sawdust as a source of dietary fiber. This is expected because of the high proportion of indigestible fiber fed to this group. However, it is interesting to note that piglets fed on 50% sawdust (T3) performed similarly as those on conventional sources of dietary fiber, suggesting these piglets might have developed some level of adaptation through time to sawdust. In agreement with these findings, previous reports have shown that high fiber diet in piglets suppresses weight gain only during the first few days and piglets adapt to the high fiber diet on continuous feeding (Anugwa, 1989)[6]

Figure 2. *Total number of anaerobic bacteria in the feces (A) and intestine (B) of piglets fed for 4 weeks with different experimental diets. T1, 50% corn and 50% rice bran; T2, 100% rice bran; T3, 50% corn and 50% sawdust; T4, 100% sawdust; T5, 100% corn. *significant difference (P< 0.05) from control (T5)*



Blood Biochemistry

No statistically significant differences were found in the biochemistry profile of piglets on different experimental diets. However, the concentrations of blood urea nitrogen (BUN), as well as aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were greater (non-significant) in T4 group compared to the control (T5) animals (data not shown), suggesting that keeping piglets for long under 100% sawdust as a sole source of dietary fiber may somehow affect the normal functioning of their organs, such as the liver.

Fecal and Intestinal Bacteria

The total number of anaerobic bacteria in piglets fed different sources of dietary fiber is shown in Figure 2. In both feces (Figure 2A) and intestines (Figure 2B) of the piglets, the total number of anaerobic bacteria was higher in T2 and T3 groups compared to the control (T5) group, while no differences were observed between T4 and T1 group. Cellulolytic bacteria represent significant proportion of the culturable flora when high-fiber diets are fed.12 Studies demonstrated that feeding pigs with high-fiber diets increased the number of fibrolytic (xylanolytic and cellulolytic) microorganisms and their activity in the large intestine depending on the duration of high-fiber feeding and on the source of dietary fiber.[6,11] Prolonged feeding of dietary fiber increased the number of fibrolytic bacteria in pigs,[11] and while diets containing 40 and 96% alfalfa meal increased the number of cellulolytic bacteria by 200%, no increase of cellulollytic bacteria was observed in pigs fed 20% corn-cob diet.[12] Accordingly, feeding piglets with 100% rice bran in T2 or 50% sawdust in T3 increased the number of anaerobic bacteria in the intestines and feces, whereas increasing the level of sawdust to 100% in T4 had a suppressing effect on the number of anaerobic intestinal flora.

Selection of lactobacilli Producing Dietary Enzymes

Many feed ingredients to swine have high concentration of fiber. However, the utilization of high fiber diets is dependent on microbial fermentation in the gastrointestinal tracts of swine. Studies have shown that the number of these beneficial microbes in the intestines increases upon feeding animals with non-starch polysaccharides.[7,14] Furthermore, pigs fed high-fiber diets will adapt to digest non-starch polysaccharides within 3-5 weeks.[6]

In the current study, we screened for lactic acid bacteria in the intestines of piglets fed for 4 weeks with different sources and levels of dietary fiber. Out of the 23 bacteria isolated in the second screening, 61% produce one or more dietary enzymes, including, protease, cellulase, phytase, and α- amylase (Table 5). Interestingly, most of these strains were isolated from piglets fed with 50-100% of sawdust as a source of dietary fiber. Despite the poor performance of piglets on 100% sawdust (T4), the high proportion of strains with beneficial digestive enzymes marks the possibility of using controlled level of sawdust in swine diets to develop potential probiotic candidates that can be used in the swine industry. Furthermore, the similar growth and health performance of piglets fed with 50% sawdust (T3) with other conventional sources of dietary fiber, such as corn and rice bran (T1 and T2), suggests the possibility of achieving acceptable performance in piglets with the addition of non-conventional and relatively cheaper source of dietary fiber.

CONCLUSION

Taken together, the results of the current study have shown that piglets fed different sources of dietary fiber respond differently in terms growth and health performance and the levels and activities of anaerobic intestinal microflora. A high level of sawdust in piglet diets negatively affects the feed intake, growth, and health performance of piglets. However, when sawdust is mixed with other sources of dietary fiber, it is beneficial to maintain acceptable growth performance in piglets at a cheaper cost. More importantly, it helps to promote intestinal microflora with beneficial digestive enzymes that enhance further adaptation of animals to high level of dietary fiber.

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