Effects of Dietary *Bacillus*-based Probiotic on Growth Performance, Nutrients Digestibility, Blood Characteristics and Fecal Noxious Gas Content in Finishing Pigs

Y. J. Chen, B. J. Min, J. H. Cho, O. S. Kwon, K. S. Son, H. J. Kim and I. H. Kim*
Department of Animal Resource & Science, Dankook University, #29 Anseodong, Cheonan, Choognam, 330-714, Korea

ABSTRACT : This study was conducted to evaluate the effects of supplementation with *bacillus*-based probiotic (*Bacillus subtilis*, 1.0×10^7 CFU/g; *Bacillus coagulans*, 2.0×10^6 CFU/g and *Lactobacillus acidophilus*, 5.0×10^6 CFU/g) on finishing pigs growth performance, nutrients digestibility, blood characteristics and fecal noxious gas content and to determine the optimal addition level of this probiotic preparation. A total of forty eight pigs with an initial body weight (BW) of 90.60±2.94 kg were allotted to three dietary treatments (four pigs per pen with four pens per treatment) according to a randomized complete block design. Dietary treatment included: 1) CON (basal diet); 2) BP1 (basal diet+*bacillus*-based probiotic 0.1%) and 3) BP2 (basal diet+*bacillus*-based probiotic 0.2%). The experiment lasted 6 weeks. Through the entire experimental period, ADG was improved by 11% ($p<0.05$) in pigs fed diets supplemented with 0.2% *bacillus*-based probiotic compared to pigs fed the basal diet. ADFI and gain/feed were not affected by the treatments ($p>0.05$). Supplementation of *bacillus*-based probiotic did not affect either DM and N digestibilities or blood characteristics ($p>0.05$) of pigs. Fecal ammonia nitrogen (NH_3-N) measured at the end of experiment was reduced ($p<0.05$) when pigs were fed the diet with 0.2% *bacillus*-based probiotic. Fecal butyric acid concentration also decreased significantly ($p<0.05$) whereas acetic acid and propionic acid concentrations were not affected ($p>0.05$) when pigs were fed diets with added *bacillus*-based probiotic. In conclusion, dietary supplementation of *bacillus*-based probiotic can increase growth performance and decrease fecal noxious gas content concentration. ***(Asian-Aust. J. Anim. Sci. 2006. Vol 19, No. 4 : 587-592)***

Key Words : Probiotics, Digestibility, Blood Characteristics, Fecal Noxious Gas, Finishing Pigs

INTRODUCTION

The intestinal microflora plays a crucial role for both human and animal. During the last few decades, probiotics which also called DFM (Direct-Fed Microbials) have been demonstrated to be useful in maintaining intestinal ecosystem and improving animal health. Recent concerns regarding the use of probiotics suggested its primary action modes were: competition for receptors on the gut mucosa and nutrients with pathogens (competitive exclusion), production of antibacterial substances and stimulation of intestinal immune responses (Vandenbergh, 1993; Dunne et al., 1999; Adlerberth et al., 2000; Wenk, 2000). Although those mechanisms have been well documented by many authors, the efficacy of probiotic preparations in practice is highly inconsistent. Some authors reported the improvement of growth performance (Nousiainen and Setala, 1993), nutrient digestibility (Maxwell et al., 1983), immune system (Fernandes and Shahani, 1990; Malin et al., 1996) and reduction of fecal noxious gas emission (Hong et al., 2002) by the dietary supplementation of probiotics. However, there were also some reverse results obtained in practical feeding trials of growing-finishing pigs (Kornegay and Risley, 1996).

* Corresponding Author: I. H. Kim. Tel: +82-41-550-3652, Fax: +82-41-553-1618, E-mail: inhokim@dankook.ac.kr
Received July 5, 2005; Accepted November 10, 2005

Various results from previous studies may due to several aspects such as age of animals, strain of bacteria and addition level (Bomba et al., 2002). This indicates that it is necessary to evaluate different probiotics preparations due to its dubious efficacy. In our early studies, results showed that the nutrients digestibility and fecal noxious gas emission were affected by the addition lactic acid bacteria (Chen et al., 2005a, b). We hypothesized that nutrients digestibility and the hindgut microflora can be also influenced by dietary supplementation of probiotics which contain *bacillus* species bacteria, consequently decreased the fecal NH_3-N and VFA concentrations. Therefore, the objectives of current study were to investigate the effects of a *bacillus*-based probiotic preparation on growth performance, nutrients digestibility, blood characteristics and fecal noxious gas content in finishing pigs and determine the optimal addition level of this probiotic preparation.

MATERIALS AND METHODS

Experimental design, animals and diets

A total of 48 [(Landrace×Yorkshire)×Duroc] pigs with an initial BW of 90.60±2.94 kg were used to evaluate the effects of *bacillus*-based probiotic (*Bacillus subtilis*, 1.0×10^7 CFU/g; *Bacillus coagulans*, 2.0×10^6 CFU/g and *Lactobacillus acidophilus*, 5.0×10^6 CFU/g) supplementation

Table 1. Formula and chemical composition of diets in finishing pigs (as-fed basis)[1]

Ingredients (%)	CON[2]	BP1[2]	BP2[2]
Corn	67.45	67.35	67.25
Soybean meal	18.14	18.14	18.14
Rice bran	5.00	5.00	5.00
Molasses	5.00	5.00	5.00
Animal fat	2.00	2.00	2.00
Dicalcium phosphate	1.12	1.12	1.12
Calcium carbonate	0.68	0.68	0.68
L-lysine (78%)	0.20	0.20	0.20
Salt	0.15	0.15	0.15
Vitamin premix[3]	0.05	0.05	0.05
Mineral premix[4]	0.15	0.15	0.15
Choline chloride (60%)	0.04	0.04	0.04
L-threonine (98%)	0.02	0.02	0.02
Bacillus-based probiotic	-	0.10	0.20
Chemical composition[5]			
Digestible energy (kcal/kg)	3,365	3,365	3,365
Crude protein (%)	14.80	14.80	14.80
Lysine (%)	0.89	0.89	0.89
Calcium (%)	0.74	0.74	0.74
Phosphorus (%)	0.54	0.54	0.54

[1] Forty eight pigs with an average initial BW of 90.60±2.94 kg.
[2] Abbreviations: CON, control diet; BP1, control diet+0.1% *Bacillus*-based probiotic and BP2, control diet+0.2% *Bacillus*-based probiotic.
[3] Provided per kg of complete diet: 4,000 IU of vitamin A; 800 IU of vitamin D_3; 17 IU of vitamin E; 2 mg of vitamin K; 4 mg of vitamin B_2; 1 mg of vitamin B_6; 16μg of vitamin B_{12}; 11 mg of pantothenic acid; 20 mg of niacin and 0.02 mg of biotin.
[4] Provided per kg of complete diet: 220 mg of Cu; 175 mg of Fe; 191 mg of Zn; 89 mg of Mn; 0.3 mg of I; 0.5 mg of Co and 0.3 mg of Se.
[5] Calculated values.

on growth performance, nutrients digestibility, blood characteristics and fecal noxious gas content in finishing pigs. The experimental period lasted 6 weeks. Pigs were allotted on the basis of initial BW to 3 dietary treatments, each treatment was assigned to 4 replicate pens with 4 pigs per pen. Dietary treatments were as follows (Table 1): 1) CON (basal diet); 2) BP1 (basal diet+*bacillus*-based probiotic 0.1%) and 3) BP2 (basal diet+*bacillus*-based probiotic 0.2%). Diets were formulated to meet or exceed NRC (1998) requirements for all the nutrients regardless of treatment and fed in meal form. Pigs were housed in a double curtain-sided facility. Pens measured 1.80×1.80 m with concrete slats in all of the pens. Feed and water were provided on an *ad libitum* basis throughout all the experimental period.

Sampling and measurements

BW and feed intake were measured at the end of experiment to calculate ADG, ADFI and gain/feed using initial BW as a covariate. Pigs were fed diets containing chromic oxide (Cr_2O_3) for 7 days prior to the collection period. At the end of experiment, fecal grab samples were collected from 2 pigs in each pen for two consecutive days. Then samples were stored in refrigerator at -20°C until analysis. Before chemical analysis, fecal samples were dried at 70°C for 72 h and finely ground to pass through a 1 mm screen. All the fecal samples, along with feed samples, were analyzed for DM and N according to the AOAC procedures (AOAC, 1995). Chromium was analyzed by UV absorption spectrophotometry (Shimadzu, UV-1201, Japan). N was measured using a Leco NS 2000 Nitrogen Analyzer (LECO Corporation, St. Joseph, MI).

At the initiation of experiment, two pigs were randomly chosen from each pen and bled via jugular venipuncture to obtain blood samples for determining WBC, RBC and lymphocyte. Same pigs were bled again at the end of experiment. Blood samples were collected into K_3EDTA vacuum tube (Becton Dickinson Vacutainer Systems, Franklin Lakes, NJ) and analyzed by the automatic blood analyzer (ADVIA 120, Bayer, USA).

Fecal grab samples were also collected for analyzing NH_3-N and VFA concentrations at the end of experiment. NH_3-N concentration was determined according to the method of Chaney and Marbach (1962). The VFA measured in this experiment included acetic acid, propionic acid and butyric acid. Analysis method of VFA was as follow: 2 g of fecal samples were diluted with 8 ml of distilled water and added two drops of concentrated HCl. Then samples were mixed and centrifuged at 17,400×g for 10 min at 4°C. The supernatant was filtered and pipetted in to 2-ml gas chromatography vials. The VFA concentrations were analyzed by gas chromatography (Hewlett Packard 6890 Plus, USA) according to the method of Otto et al. (2003).

Statistical analyses

In this experiment, all statistical analyses were performed as a randomized complete block design using GLM procedures of SAS (1996). Pen was considered as the experimental unit and replication was served as a random effect. The model included the effects of block (replication) and treatment. Variability in the data is expressed as standard error (SE) of the mean and the chosen level of significance was 5%.

RESULTS AND DISCUSSION

Growth performance

The results of ADG, ADFI and Gain/feed are shown in Table 2. ADG in BP1 treatment tended to increase without significant difference ($p>0.05$). When Pigs fed 0.2% probiotic-supplemented diets (BP2), ADG was significant higher compared to CON treatment ($p<0.05$). ADFI and gain/feed were not affected among the treatments ($p<0.05$). Current results are in agreement with Jonsson and Conway (1992), who reported dietary addition of *bacillus* species improved growth performance and health of pigs.

Table 2. Effects of *Bacillus*-based probiotic on growth performance in finishing pigs[1]

Items	CON[2]	BP1[2]	BP2[2]	SE[3]
ADG (g)	717[b]	755[ab]	799[a]	16
ADFI (g)	2,441	2,480	2,588	80
Gain/feed	0.294	0.305	0.309	0.022

[1] Forty eight pigs with an average initial BW of 90.60±2.94 kg.
[2] Abbreviations: CON, control diet; BP1, control diet+0.1% *Bacillus*-based probiotic and BP2, control diet+0.2% *Bacillus*-based probiotic.
[3] Pooled standard error.
[a, b] means in the same row with different superscripts differ ($p<0.05$).

Table 3. Effects *Bacillus*-based probiotic on nutrients digestibility in finishing pigs[1]

Items (%)	CON[2]	BP1[2]	BP2[2]	SE[3]
DM	69.48	70.28	70.63	1.74
N	63.64	63.69	66.11	2.38

[1] Forty eight pigs with an average initial BW of 90.60±2.94 kg.
[2] CON: control diet; BP1: control diet+0.1% *Bacillus*-based probiotic and BP2: control diet+0.2% *Bacillus*-based probiotic.
[3] Pooled Standard error.

Alexopoulos et al. (2004) observed significant improvement when finishing pigs fed diet included probiotic (*Bacillus licheniformis* and *Bacillus subtilis*). Our previously study, conducted by Chen et al. (2005a), also observed an improvement when growing pig fed diets supplemented complex probiotic (*Lactobacillus acidophilus*, *Saccharomyces cerevisiae* and *Bacillus subtilis*). However, Kornegay et al. (1990) reported that there was no effect on growth performance by the supplementation of *Lactobacillus acidophilus* in finishing pigs.

Unlike the diverse results obtained from growing and finishing pigs experiments, many studies of probiotics conducted in nursery pigs found positive effects when diets added probiotic preparations (Lessard and Brissom, 1987; Park et al., 2001). These results indicated that age of pigs is a considerable fact relate to the probiotics efficacy. Stavric and Kornegay (1995) suggested that probiotics are more effective in animals during microflora development or when microflora stability is impaired. They also reported that the improvement of growth performance will often be marginal during optimal rearing and feeding conditions. Data from our experiment showed that the ADG in control group was only 717 g, this value was not as higher as values obtained from some other studies. Therefore, it was reasonable that the response of current probiotic presented more effective in our experiment.

Nutrients digestibility

Table 3 shows the effects of *bacillus*-based probiotic on nutrients digestibility in finishing pigs. Digestibility of DM was not affected by the addition of probiotic ($p>0.05$). N digestibility was increased slightly in BP2 treatment compared to CON treatment, however, there was no significant difference ($p>0.05$).

Table 4. Effects of *Bacillus*-based probiotic on blood components in finishing pigs[1]

Items	CON[2]	BP1[2]	BP2[2]	SE[3]
RBC ($\times10^6$/mm^3)				
0 day	6.24	6.29	6.06	0.15
35 days	6.28	6.68	6.63	0.28
Difference	0.04	0.40	0.57	0.35
WBC ($\times10^3$/mm^3)				
0 day	15.11	14.54	14.18	0.87
35 days	15.81	14.62	14.69	0.61
Difference	0.70	0.08	0.51	0.81
Lymphocyte (%)				
0 day	49.00	43.80	46.20	7.05
35 days	63.00	58.80	65.00	4.95
Difference	14.00	15.00	18.80	8.66

[1] Forty eight pigs with an average initial BW of 90.60±2.94 kg.
[2] CON: control diet; BP1: control diet+0.1% *Bacillus*-based probiotic and BP2: control diet+0.2% *Bacillus*-based probiotic.
[3] Pooled standard error.

Obtained data suggested that digestibilities of DM and N were unaffected by the supplementation of probiotic. Results from Kim et al. (1993) observed no effects on digestibility when finishing pigs fed diets included probiotic. Similarly, Kornegay and Risley (1996), who used two *bacillus* products (Biomate2B®, *Bacillus subtilis, Bacillus licheniformis* and Pelletmate Livestock®, *Bacillus subtilis, Bacillus licheniformis, Bacillus pumilus*) in finishing pigs diets did not find any influences on nutrients (DM, NDF, ADF, ash and N) digestibilities. In the study which conducted by Shon et al. (2005), no improvement of DM digestibility were observed when both growing and finishing pigs fed diets included *Lactobacillus reuteri*-based probiotic (*Lactibacillus reuteri, Lactobacillus salivarius, Lactobacillus plantarum and Saccharomyces cerevisiae*). Their study also suggested that N digestibility was not affected in growing pigs while was improved in finishing pigs. In nursery pigs, Xuan et al. (2001) found nutrients digestibility was not affected by the addition of a probiotics complex (*Bacillus sp.* and *Saccharomyces cerevisiae*). On the contrary, positive effects on nutrients digestibility were reported by some other authors (Maxwell et al., 1983; Kil et al., 2004; Lim et al., 2004). According to Jonsson and Conway (1992)'s study, they suggested that *Bacillus spp.* are not normal components of indigenous intestinal microflora, so that those bacteria are hard to colonize in the digestive tract. Inconsistent results on nutrients digestibility might be due to this reason.

Blood characteristics

Effects of *bacillus*-based probiotic supplementation on blood characteristics in finishing pigs are present in Table 4. Obtained data show that there were no influences on RBC, WBC and lymphocyte when diets added *bacillus*-based probiotic. The capacity of probiotics to influence the

Table 5. Effects of *Bacillus*-based probiotic on fecal NH_3-N and VFA concentrations in finishing pigs[1]

Items	CON[2]	BP1[2]	BP2[2]	SE[3]
NH_3-N (ppm)	685[a]	650[ab]	568[b]	28
Volatile fatty acids (ppm)				
Acetic acid	373	332	357	38
Propionic acid	458	360	451	35
Butyric acid	238[a]	175[b]	180[b]	14

[1] Forty eight pigs with an average initial BW of 90.60±2.94 kg.
[2] Abbreviations: CON, control diet; BP1, control diet+0.1% *Bacillus*-based probiotic and BP2, control diet+0.2% *Bacillus*-based probiotic.
[3] Pooled standard error.
[a, b] means in the same row with different superscripts differ ($p<0.05$).

immune system is one of the more recent developments in this field (Bloksma et al., 1979). Some beneficial effects of probiotics on animal's immune system have been proposed by many authors. Perdigon et al. (1991) suggested *Lactobacillus casei* have immunoadjuvant activity. Takahashi et al. (1998) and Vitini et al. (2000) reported that *Bifidobacterium longum* and other lactic acid bacteria can increase the total amount of intestinal IgA. However, Kluber et al. (1985) reported no effect of *Streptococcus faecium* on the cell-mediated immune response in weaning pigs. Similar results also observed by Apgar et al. (1993). Current results are in agreement with those reports. Likewise, our previous studies also observed no effects of single or complex probiotic preparations on blood characteristics (WBC, RBC and lymphocyte) in growing and finishing pigs (Chen et al., 2005a, b). These inconsistent responses to probiotics supplementation in growing and finishing pig diets may be due to varying age and health status of the experimental animals used. The effects of probiotics proposed to be more effective during young age and under stress conditions such as weaning or dietary changes for piglets (Jonsson and Conway, 1992). Also, our experimental period only lasted 6 weeks, which might be too short to induce any changes in finishing pigs.

Fecal noxious gas content

Fecal NH_3-N was decreased significantly ($p<0.05$) when pigs fed diets supplemented with 0.2% *bacillus*-based probiotic compared to pigs fed basal diets (Table 5). Acetic acid and propionic acid were not affected by the addition of *bacillus*-based probiotic. Butyric acid was decreased significantly when pigs fed diets including *bacillus*-based probiotic than those of pigs fed basal diets ($p<0.05$).

Fecal noxious gas emission such as NH_3-N, H_2S and VFA has become on of the major air pollution in modern concentrative pig production (van Breemen et al., 1982; Slanina, 1994). High concentrations of ammonia or H_2S can cause hazard effects to humans and animals (Drummond et al., 1980; Crook et al., 1991; Busse, 1993). Ferket et al. (2002) reviewed the nutritional strategies to reduce negative emissions from nonruminants. They suggested that the ultimately fecal noxious gas emission of animals is related to nutrients utilization and intestinal microflora ecosystem. In our study, NH_3-N was decreased about 20% (685 vs. 568 ppm) by dietary addition of 0.2% probiotic. This is in agreement with Hong et al. (2004), who reported a reduction in NH_3-N when finishing pigs fed diets added complex probiotic (*Phichia anomala* ST, *Galactomyces geotrichum* SR59 and *Thiobacillus sp.*). Ji and Kim (2002) observed the ammonia production of pigs was decreased 21% by the addition of complex probiotics (*Bacillus sp.*, *Aspergillus oryzae*, and *Lactobacillus acidophilus*). However, digestibility of N was not increased in present results. Therefore, the reduction of fecal NH_3-N may not result from the nutrients digestibility but the alternation of intestinal microflora or some other reasons of pigs.

Production of certain VFA is also the result of anaerobic microbial fermentation of soluble carbohydrates (Argenzio and Southworth, 1974; Ferket et al., 2002). Mackie et al. (1998) also suggested that VFA originate partly by anaerobic bacteria in the gastrointestinal tract and feces. Our study observed fecal butyric acid decreased while acetic acid and propionic acid without influence when *bacillus*-based probiotic added to diet. O'Neill and Phillips (1992) suggested the proportion of individual VFA to total VFA concentration associate with odor offensiveness. The manure odor quality mainly contributes from long-chain VFA (Spoelstra, 1980). Thus, the decrease in the proportion of long-chain or brancked-chanined VFA such as butyrate, isobutyrate and valerate has the potential to reduce odor emission (Ferket et al., 2002).

IMPLICATIONS

This study indicates that dietary supplementation of *bacillus*-based probiotic preparation at the level of 0.2% is effective in improving growth performance and reducing fecal NH_3-N and butyric acid concentrations in finishing pigs. Therefore, we propose that current probiotic preparation can replace some double-side effect additives such antibiotics or growth promoters etc. Possible mechanisms need to be further investigated in order to finely explain how this *bacillus*-based probiotic exert its beneficial effects.

REFERENCES

Adlerberth, I., M. Cerquetti, I. Poilane, A. Wold and A. Collignon. 2000. Mechanisms of colonisation and colonisation resistance of the digestive tract. Microb. Ecol. Health Dis. 11:223-239.

Alexopoulos, C., I. E. Georgoulakis, A. Tzivara, C. S. Kyriakis, A. Govaris and S. C. Kyriakis. 2004. J. Vet. Med. Physiol. Pathol. Clin. Med. 51:306.

AOAC. 1995. Official method of analysis. 16th Edition. Association of Official Analytical Chemists, Washington, DC.

Apgar, G. A., E. T. Kornegay, M. D. Lindemann and C. M. Wood. 1993. The effect of feeding various levels of *Bifidobacteriurn globosurn A* on the performance, gastrointestinal measurements, and immunity of weanling pigs and on the performance and carcass measurements of growing-finishing pigs. J. Anim. Sci. 71:2173-2179.

Argenzio, R. A. and M. Southworth. 1974. Sites of organic acid production and absorption in gastrointestinal tract of the pig. Am. J. Physiol. 228:454-460.

Bloksma, N., E. De Heer, M. Van Dijk and M. Willers. 1979. Adjuvanticity of *Lactobacilli*. I. Differential effects of viable and killed bacteria. Clin. Exp. Immunol. 37:367-375.

Bomba, A., R. Nemcova, S. Gancarcıkova, R. Herich, P. Guba and D. Mudronova. 2002. Improvement of the probiotic effect of micro-organisms by their combination with maltodextrins, fructo-oligosaccharides and polyunsaturated fatty acids. Br. Journal of Nutrition. 88 (Suppl.)1:95-99.

Busse, F. W. 1993. Comparison measurements of the house climate in swine stables with and without respiratory diseases or cannibalism. In: Livestock Environment (Ed. E. Collins and C. Boon). Fourth International Symposium, University of Warwick, Coventry, England. ASAE, St. Joseph, MI. pp. 904-908.

Chaney, A. L. and E. P. Marbach. 1962. Modified regents for determination of urea and ammonia. Clin. Chem. 8:131.

Chen, Y. J., K. S. Son, B. J. Min, J. H. Cho, O. S. Kwon and I. H. Kim. 2005a. Effects of dietary probiotic on growth performance, nutrients digestibility, blood characteristics and fecal noxious gas content in growing pigs. Asian-Aust. J. Anim. Sci. 18:1464-1468.

Chen, Y. J., K. S. Son, B. J. Min, J. H. Cho, O. S. Kwon and I. H. Kim. 2005b. Effects of dietary probiotic on growth performance, nutrients digestibility, blood characteristics and fecal noxious gas content in growing pigs. Asian-Aust. J. Anim. Sci. 18(10):1464-1468.

Crook, B., J. F. Robertson, S. A. T. Glass, E. M. Botheroyd, J. Lacey and M. D. Topping. 1991. Airborne dust, ammonia, microorganisms and antigens in pig confinement houses and the respiratory health of exposed farm-workers. Am. Indust. Hygiene Assoc. J. 52:271-279.

Drummond, J. G., S. E. Curtis, J. Simon and H. W. Norton. 1980. Effects of aerial ammonia on growth and health of young pigs. J. Anim. Sci. 50:1085-1091.

Dunne, C., L. Murphy, S. Flynn, L. O'Mahony, S. O'Halloran, M. Feeney, D. Morrissey, G. Thornton, G. Fitzgerald, C. Daly, B. Kiely, E. M. M. Quigley, G. C. O'Sullivan, F. Shanahan and J. Kevin. 1999. Probiotics: from myth to reality. Demonstration of functionality in animal models of disease and in human clinical trials. Antonie van Leeuwenhoek 76:279-292.

Ferket, P. R., E. van Heugten, T. A. T. G. van Kempen and R. Angel. 2002. Nutritional strategies to reduce environmental emissions from nonruminants. J. Anim. Sci. 80 (E. Suppl. 2):E168-E182.

Fernandes, C. F. and K. M. Shahani. 1990. Anticarcinogenic and immunological properties of dietary lactobacilli. J. Food Prot. 53:704-710.

Hong, J. W., I. H. Kim, O. S. Kwon, J. H. Kim, B. J. Min and W. B. Lee. 2002. Effects of dietary probiotics supplementation on growth performance and fecal gas emission in nursing and finishing pigs. J. Anim. Sci. Technol. (Kor.) 44:305-314.

Ji, F. and S. W. Kim. 2002. Reducing odor in swine production: Effect of enzymes and probiotics on ammonia production. J. Anim. Sci. Vol. 80 (Suppl. 1).

Jonsson, E. and P. Conway. 1992. Probiotics for pigs. In: (Ed. R. Fuller) Probiotics: The Scientific Basis. Chapman and Hall, London. pp. 260-316.

Kil, D. Y., S. J. Lim, J. Z. Tian, B. G. Kim, K. S. Kim and Y. Y. Kim. 2004. Effect of continuous feeding of probiotics on growth performance, nutrient digestibility, blood urea nitrogen and immune responses in pigs. J. Anim. Sci. Technol. (Kor.) 46:39-48.

Kim, I. H., J. D. Hancock, R. H. Hines and C. R. Risley. 1993. Effects of cellulase and bacterial feed additives on the nutritional value of sorghum grain for finishing pigs. Kansas Agric. Exp. Sta. Rep. Prog. No. 695:144.

Kluber, E. F., D. S. Pollmann and F. Blecha. 1985. Effect of feeding *Streptococcus faecium* to artificially reared pigs on growth, hematology and cell-mediated immunity. Nutr. Rep. Int. 32:57.

Kornegay, E. T. and C. R. Risley. 1996. Nutrient digestibilities of a corn-soybean meal diet as influenced by *Bacillus* products fed to finishing swine. J. Anim. Sci. 74:799-805.

Kornegay, E. T., C. M. Wood, G. G. Ball and C. R. Risley. 1990. Use of *Lactobacillus acidophilus* for growing and finishing pigs. VA Polytech. Inst. State Univ. Anim. Sci. Res. Rep. 9:13.

Lessard, M. and G. J. Brisson. 1987. Effect of a lactobacillus fermentation product on growth, immune response and fecal enzyme activity in weaned pigs. Can. J. Anim. Sci. 67:509.

Lim, H. S., B. H. Kim and I. K. Paik. 2004. Effects of Natufermen® supplementation to the diet on the performance of weanling pigs. J. Anim. Sci. Technol. (Kor.) 46:981-988.

Mackie, R. I., P. G. Stroot and V. H. Varel. 1998. Biochemical identification and biological origin of key odor components in livestock waste. J. Anim. Sci. 76:1331-1342.

Malin, M., H.Suomalainen, M. Saxelin and E. Isolauri. 1996. Promotion of IgA immune response in patients with Crohn's disease by oral bacteriotherapy with *Lactobacillus GG*. Annals Nutr. Metab. 40:137-145.

Maxwell, C. V., D. S. Buchanan, F. N. Owens, S. E. Gilliland, W. G. Luce and R. Vencl. 1983. Effect of probiotic supplementation on performance, fecal parameters and digestibility in growing-finishing swine. Oklahoma Agric. Exp. Sta. Anim. Sci. Res. Rep. pp. 114:157.

Nousiainen, J. and J. Setala. 1993. Lactic acid bacteria as animal probiotics. In Lactic Acid Bacteria. (Ed. S. Salminen and A. von Wright). New York, Marcel Dekker. pp. 315-356.

NRC. 1998. Nutrient requirement of pigs. 10th Edition. National Research Council, Academy Press. Washington, DC.

O'Neill, D. H. and V. R. Phillips. 1992. A review of the control of odour nuisance from livestock buildings: Part 3, properties of the odorous substances which have been identified in livestock wastes or in the air around them. J. Agric. Eng. Res. 53:23-50.

Otto, E. R., M. Yokoyama, S. Hengemuehle, R. D. von Bermuth, T. van Kempen and N. L. Trottier. 2003. Ammonia, volatile fatty acids, phenolics, and odor offensiveness in manure from growing pigs fed diets reduced in protein concentration. J. Anim. Sci. 81:1754-1763.

Park, D. Y., H. Namkung and I. K. Paik. 2001. Effects of

supplementary enzymes or probiotics on the performance and ammonia gas production in weanling pigs. J. Anim. Sci. Technol. (Kor.) 43:485-496.

Perdigon, G., S. Alvarez and A. Pesce de Ruiz Holgado. 1991. Immunoadjuvant activity of oral *Lactobacillus casei*: influence of dose on the secretory immune response and protective capacity in intestinal infections. Dairy Res. 58:485-496.

SAS. 1996. SAS user's guide. Release 6.12 edition. SAS Institute. Inc Cary NC. USA.

Shon, K. S., J. W. Hong, O. S. Kwon, B. J. Min, W. B. Lee, I. H. Kim, Y. H. Park and I. S. Lee. 2005. Effects of *Lactobacillus reuteri*-based direct-fed microbial supplementation for growing-finishing pigs. Asian-Aust. J. Anim. Sci. 18:370-374.

Slanina, S. 1994. Forest dieback and ammonia-a typical Dutch problem. Chemistry International. 16:2-3.

Spoelstra, S. F. 1980. Origin of objectionable odorous components in piggery wastes and the possibility of applying indicator components for studying odour development. Agric. Env. 5:241-260.

Stavric, S. and E. T. Kornegay. 1995. Microbial probiotic for pigs and poultry. Biotechnology in Animal Feeds and Animal Feeding. (Ed. R. J. Wallace and A. Chesson). Weinheim:VCH Verlagsgesellschaft mbH. pp. 205-231.

Takahashi, T., E. Nakagawa, T. Nara, T. Yajima and T. Kuwata. 1998. Effects of orally ingested *bifidobacterium longum* on the mucosal IgA response of mice to dietary antigens. Bio. Biotechnol. Biochem. 62:10-15.

van Breemen, N., P. A. Burrough, E. J. Velthorst, H. F. van Dobben, Toke de Wit, T. B. Ridder, H. F. R. Reijnders. 1982. Soil acidification from atmospheric ammonium sulphate in forest canopy throughfall. Nat. 299:548-550.

Vandenbergh, P. A. 1993. Lactic acid bacteria, their metabolic products and interference with microbial growth. FEMS Microbiology Review. 12:221-238.

Vitini, E., S. Alvarez, M. Medina, M. Medici, M. V. de Budeguer and G. Perdigon. 2000. Gut mucosal immunostimulation by lactic acid bacteria. Biocell. 24:223-232.

Wenk, C. 2000. Recent advances in animal feed additives such as metabolic modifiers, antimicrobial agents, probiotics, enzymes and highly available minerals. Asian-Aust. J. Anim. Sci. 13:86-95.

Xuan, Z. N., J. D. Kim, K. N. Heo, H. J. Jung, J. H. Lee, Y. K. Han, Y. Y. Kim and I. H. Han. 2001. Study on the development of a probiotics complex for weaned pigs. Asian-Aust. J. Anim. Sci. 14:1425-1428.