RESEARCH ARTICLE

J Anim Sci Technol 2024;66(4):763-780
https://doi.org/10.5187/jast.2023.e88

JAST
Journal of Animal Science and Technology
pISSN 2672-0191 eISSN 2055-0391

Effects of single and complex probiotics in growing-finishing pigs and swine compost

Kyeongho Jeon[1#], Minho Song[2#], Jihwan Lee[3#], Hanjin Oh[1], Dongcheol Song[1], Seyeon Chang[1], Jaewoo An[1], Hyunah Cho[1], Sehyun Park[1], Hyeunbum Kim[4*], Jinho Cho[1*]

[1]*Department of Animal Science, Chungbuk National University, Cheongju 28644, Korea*
[2]*Department of Animal Science and Biotechnology, Chungnam National University, Daejeon 34134, Korea*
[3]*Department of Poultry Science, University of Georgia (UGA), Athens, GA 30602, USA*
[4]*Department of Animal Resource and Science, Dankook University, Cheonan 31116, Korea*

Received: Jul 19, 2023
Revised: Aug 2, 2023
Accepted: Aug 11, 2023

#These authors contributed equally to this work.

***Corresponding author**
Hyeunbum Kim
Department of Animal Resource and Science, Dankook University, Cheonan 31116, Korea.
Tel: +82-41-550-3652
E-mail: hbkim@dankook.ac.kr

Jinho Cho
Department of Animal Science, Chungbuk National University, Cheongju 28644, Korea.
Tel: +82-43-261-2544
E-mail: jinhcho@chungbuk.ac.kr

ORCID
Kyeongho Jeon
https://orcid.org/0000-0003-2321-3319
Minho Song
https://orcid.org/0000-0002-4515-5212
Jihwan Lee
https://orcid.org/0000-0001-8161-4853

Abstract

This study was conducted to supplement single and complex probiotics to investigate the effect on growing-finishing pigs and compost. In experiment 1, the 64 crossbred ([Landrace × Yorkshire] × Duroc) pigs with an initial body weight of 18.75 ± 0.33 kg and a birth of 63 days were assigned to a completely randomized four treatment groups based on the initial body weight (4 pigs in a pen with 4 replicate pens for each treatment). For 13 weeks, the dietary treatments were provided: 1) Control (CON; basal diet), 2) T1 (CON + 0.2% *Bacillus subtilis*), 3) T2 (CON + 0.2% *Saccharomyces cerevisiae*), 4) T3 (CON + 0.2% *Bacillus subtilis* + 0.2% *Saccharomyces cerevisiae*). In experiment 2, the pig manure was obtained from Chungbuk National University (Cheongju, Korea) swine farm. For 12 weeks, the supplementary treatments were provided: 1) CON, non-additive compost; 2) T1, spray *Bacillus subtilis* 10 g per 3.306 m^2; 3) T2, spray *Bacillus subtilis* 40 g per 3.306 m^2; 4) T3, spray *Saccharomyces cerevisiae* 10 g per 3.306 m^2; 5) T4: spray *Saccharomyces cerevisiae* 40 g per 3.306 m^2; 6) T5, spray *Bacillus subtilis* 5 g + *Saccharomyces cerevisiae* 5 g per 3.306 m^2; 7) T6, spray *Saccharomyces subtilis* 20 g + *S. cerevisiae* 20 g per 3.306 m^2 and there were 6 replicates each treatment. In experiment 1, During the overall experimental period, T3 showed significantly improved ($p < 0.05$) feed conversion ratio and average daily gain compared to other groups. In average maturity score, T3 showed significantly higher ($p < 0.05$) than other groups. Supplementing complex probiotics group improved ($p < 0.05$) H2S emissions and fecal microflora compared to the non-supplementing group. In experiment 2, additive probiotics groups had no effect ($p > 0.05$) on moisture content than the non-additive group at 9 and 12 weeks. T6 showed a significantly improved ($p < 0.05$) average maturity score at all periods and ammonia emissions at 1 week and 4 weeks compared to other groups. In summary, supplementation complex probiotics induced positive effects on both pigs and compost.

Keywords: Compost maturity, Finishing pigs, Growing pigs, Probiotics

Hanjin Oh
https://orcid.org/0000-0002-3396-483X
Dongcheol Song
https://orcid.org/0000-0002-5704-603X
Seyeon Chang
https://orcid.org/0000-0002-5238-2982
Jaewoo An
https://orcid.org/0000-0002-5602-5499
Hyunah Cho
https://orcid.org/0000-0003-3469-6715
Sehyun Park
https://orcid.org/0000-0002-6253-9496
Hyeunbum Kim
https://orcid.org/0000-0003-1366-6090
Jinho Cho
http://orcid.org/0000-0001-7151-0778

Competing interests
No potential conflict of interest relevant to this article was reported.

Funding sources
This study was provided by the Ministry of Education and the National Research Foundation (NRF) of the Korea as part of the "Leaders in Industry-University Cooperation 3.0" Project (Code 20221345356219).

Acknowledgements
Not applicable.

Availability of data and material
All data generated or analyzed during this study are included in this published article.

Authors' contributions
Conceptualization: Song M, Kim H, Cho J.
Data curation: Oh H, Song D, An J.
Formal analysis: Chang S, Park S.
Methodology: Lee J, Oh H, An J.
Software: Chang S, Cho H.
Validation: Oh H, An J, Park S.
Investigation: Cho J.
Writing - original draft: Jeon K, Lee J, Cho J.
Writing - review & editing: Jeon K, Song M, Lee J, Oh H, Song D, Chang S, An J, Cho H, Park S, Kim H, Cho J.

Ethics approval and consent to participate
The experimental procedures received prior approval from the Animal Ethics Committee of Chungbuk National University (CBNUA-1740-22-02).

INTRODUCTION

Various issues have emerged associated with improving livestock manure management (LM) due to the increasing demand for animal-sourced foods [1]. LM generally contains heavy metals (arsenic, copper, and zinc) that can be hazardous to humans and the environment. It also generates various harmful compounds (e.g., volatile fatty acids, alcohols, amines, hydrogen sulfide [H_2S], and ammonia [NH_3]) that can cause an unpleasant odor [2]. In particular, NH_3 is produced at a higher concentration than other odorous gases. It is involved in air pollution as a precursor to secondary ultrafine dust [3].

Composting is the primary method of LM treatment applied on a farm [4]. However, the process of composting can have various adverse effects such as nitrogen loss and increase in greenhouse gas due to rapid degradation of nitrogenous organic matter and the presence of anaerobic space in feedstocks [5]. Previous studies have shown that nitrogen losses during composting can result in the production of NH_3, nitrous oxide, and leachate, which can reduce the agricultural value of composted products, and contribute to increase in greenhouse gas emissions and unpleasant odor [6].

Probiotics have been shown to improve the environment of digestive organ microorganisms by reducing harmful microorganisms in the intestine, resulting in improved nitrogen utilization and reduced nitrogen excretion in pigs [7]. Li and Kim [8] have reported that supplementation of *Saccharomyces cerevisiae* (*S. cerevisiae*) can improve nitrogen digestibility in growing pigs. Other studies have suggested that supplementation including *Bacillus subtilis* (*B. subtilis*) can attenuate NH_3 release by suppressing the urease-producing microorganisms in the gastrointestinal lumen by producing the protein-digesting enzyme such as subtilisin in pigs [8]. Wang et al. [9], have reported that *S. cerevisiae* can reduce the *Methanobrevibacter* spp. known to produce methanogen and methane. According to results of previous studies, supplementation containing *Bacillus* spp. might reduce NH_3 emissions in pigs [8,10].

However, there are few studies on adding complex probiotics to swine diets and manure. Therefore, the objective of this study was to determine effects of single and complex probiotics in growing-finishing pigs and compost on growth performance, odorous gas emissions, blood profiles, and compost maturity.

MATERIALS AND METHODS

Ethics approval and consent to participate

All experimental procedures received prior approval from the Animal Ethics Committee of Chungbuk National University (CBNUA-1740-22-02).

Source of probiotics

The probiotics used in the current study were kindly provided by a commercial company (Garam Co. Ltd., Eumseong, Korea).

Experiment 1

Experimental design, animals, and housing

A total of 64 crossbred growing pigs ([Landrace × Yorkshire] × Duroc) with an average initial body weight (BW) of 18.75 ± 0.33 kg and a birth of 63 days were used for 13 weeks in this study. All pigs were assigned to a completely randomized four treatment groups based on the initial BW. There were 4 pigs in a pen with 4 replicate pens for each treatment. Dietary treatments were as

https://doi.org/10.5187/jast.2023.e88

Table 1. Ingredients and chemical composition of the basal experimental diets (as fed basis)

Items	Grower phase (0−6 w)	Finisher phase (7−13 w)
Ingredients (%)	100	100
Corn	53.479	55.776
Soybean meal	15.660	13.100
Wheat (11%)	3.750	3.750
Rice bran	6.500	6.500
DDGS	11.500	11.500
Limestone	1.270	0.910
Vegetable oil	1.320	1.720
Sugar	4.590	4.870
Poultry oil	0.200	0.200
Salt	0.358	0.376
Choline chloride	0.040	0.040
Lysine sulphate	0.724	0.711
L-Methionine (99%)	0.083	0.077
Tryptophan (98%)	0.049	0.038
Threonine	0.146	0.162
MDCP	0.061	0.000
Emulsifier	0.050	0.050
Vitamin and mineral premix[1)]	0.220	0.220
Calculated values		
Dry matter (%)	86.69	86.45
Protein (%)	15.90	14.89
Fat (%)	5.51	5.75
Fiber (%)	3.83	3.79
Ash (%)	5.19	4.41
Calcium (%)	0.72	0.46
Phosphorus (%)	0.49	0.44
Na (%)	0.20	0.20
Cl (%)	0.35	0.35
NE (kcal/kg)	2,408.70	2,446.50
Analyzed values (g/kg)		
AID Arg	7.40	6.70
AID Ile	4.63	4.26
AID Leu	12.07	11.48
AID Lys	8.90	8.20
AID Met + Cys	5.25	5.00
AID Met	3.17	3.01
AID Thr	5.43	5.25
AID Trp	1.60	1.39
AID Val	5.71	5.28

[1)]Provided per kilogram of complete diet: vitamin A, 11,025 U; vitamin D_3,1,103 U; vitamin E, 44 U; vitamin K, 4.4 mg; riboflavin, 8.3 mg; niacin, 50 mg; thiamine, 4 mg; d-pantothenic, 29 mg; choline, 166 mg; and vitamin B_{12}, 33 μg; Cu (as $CuSO_4 \cdot 5H_2O$), 12 mg; Zn (as $ZnSO_4$), 85 mg; Mn (as MnO_2), 8 mg; I (as KI), 0.28 mg; and selenium (as $Na_2SeO_3 \cdot 5H_2O$), 0.15 mg.

DDGS, Distiller's dried grains with solubles; MDCP, monodicalcium phosphate; NE, Net energy; AID, apparent ileal digestibility; Arg, arginine; Ile, Isoleucine; Leu, leucine; Lys, lysine; Met, methionine; Cys, cystine; Thr, threonine; Trp, tryptophan; Val, valine.

follows: 1) Control (CON; basal diet), 2) T1 (CON + 0.2% *B. subtilis*), 3) T2 (CON + 0.2% *S. cerevisiae*), 4) T3 (CON + 0.2% *B. subtilis* + 0.2% *S. cerevisiae*). The probiotic used in this study such as *B. subtilis* and *S. cerevisiae* contains 2.0×10^{10} CFU kg^{-1} and 3.0×10^{10} CFU kg^{-1}, respectively. All diets were formulated to meet or exceed the NRC [11] requirement (Table 1). The diet was divided into two phases: the grower phase (0–6 weeks) and the finisher phase (7–13 weeks). Each of the pigs had *ad libitum* access to water. A nipple drinker and single-sided stainless steel automated feeder were placed with each pen.

Measurements and sampling

Growth performance

BW was recorded on initial, 6, 9, and 13 weeks to calculate average daily gain (ADG), average daily feed intake (ADFI), and feed conversion ratio (FCR). The ADG was calculated by subtracting the BW of the previous time point from the BW of the current time point and dividing it by the period. ADFI was calculated by subtracting the remaining feed amount from the initial feed amount and dividing it by the period, and FCR was calculated by dividing feed intake by ADG.

Nutrient digestibility

In experiment periods, fresh fecal samples are collected at 6 and 13 weeks using rectal massage after each treatment. Fresh fecal and feed samples were stored in a freezer at −20℃ after collection immediately. The stored fecal samples were dried at 70℃ for 3 days and then crushed on a 1 mm screen at the end of the experiment. Chromic oxide was analyzed immediately after supplementation of 0.2% as an indigestible marker that was added to the pig's diet the apparent total tract digestibility (ATTD) of crude protein (CP), dry matter (DM), and gross energy (GE). Chromium levels were analyzed with ultraviolet absorption spectrometry (UV-1201, Shimadzu, Kyoto, Japan) using a method used by Williams et al. [12]. The procedures utilized for the determination of DM (method 930.15), and CP (method 999.03) were conducted with the methods of AOAC [13], and GE using a bomb calorimeter (Parr 6400 Bomb Calorimeter, Parr Instrument, Moline, IL, USA).

Calculating the ATTD used the following formula:

$$\text{"Digestibility (\%)} = [1 - \{(Nf \times Cd)/(Nd \times Cf)\}] \times 100\text{"}$$

Nf = nutrient concentration in feces (DM %), Nd = nutrient concentration in diet (DM %), Cd = chromium concentration in diet (DM %), and Cf = chromium concentration in feces (DM %).

Blood profiles

At 6 and 13 weeks, blood samples from the anterior vena cava of 4 pigs per treatment. Blood samples were collected into vacuum tubes containing K_3EDTA (Becton, Dickinson and Co., Franklin Lakes, NJ, USA) for complete blood count analysis, and nonheparinized tubes for serum analysis, respectively. After collection, serum samples were centrifuged (3,000×g) for 15 min at 4℃. The white blood cell (WBC), and red blood cell (RBC) levels were determined using an automatic blood analyzer (ADVIA 120®, Bayer Lab, NY, USA). The blood urea nitrogen (BUN), creatinine, and total protein levels were measured using a chemistry analyzer (Cobas C702, Roche, Munich, Germany).

Maturity score of compost

Compost was used by mixing sawdust with manure obtained from pigs. The manure was collected

https://doi.org/10.5187/jast.2023.e88

at the 6 and 13 weeks. Compost was prepared by adding sawdust to swine manure at a ratio of 4:1 (swine manure: sawdust) for adjusting moisture content (MC) at the beginning of the experiment. Each compost was stored in a plastic box with air holes. Each compost was mixed weekly to supply oxygen. Compost maturity was evaluated using a maturity analyzer (CoMMe-100, E&A TECH, Dangjin, Korea), according to maturity analyses specified in the fertilizer quality inspection and sampling standards in Korea. MC was adjusted to be around 50% for all samples before they were analyzed for maturity score according to methods described by Song et al. [14]: score 1, immature (barely progressing in compost maturity); score of 2, initial maturity (an initial state in which maturity progressed); score of 3, the middle of maturity (compost maturity in which a longer stay was required); score of 4, the latter part of maturity (compost was almost mature); score of 5, maturity completion (compost was mature). The samples collected at 6 weeks were ripened for 14 weeks, and the samples collected at 13 weeks were ripened for 13 weeks.

Odorous gas emissions

The feces (150 g) that collected for 2 pigs each treated by a rectal massage at 6 and 13 weeks. The samples were mixed with 150 g of collected feces 100 g of sawdust, and 50 g of urine to analyze gas emissions. About samples were stored in a 4.2 L plastic box at room temperature 26℃ and fermented for 72 h. The plastic boxes with small holes sealed with plaster were used for analyzing fecal NH_3, H_2S, and acetic acid (CH_3COOH) emissions of samples. The samples with plastic boxes are shaken 20 s to break down any crust formation before the measurement. NH_3 concentrations were determined within the scope of 5.0–100.0 ppm (No.3La, detection tube, Gastec, Kanagawa, Japan), H_2S concentrations were determined within scope of 2.0–20.0 ppm (No.4LK, detection tube, Gastec), and CH_3COOH concentrations were determined within the scope of 2.5–10.0 ppm (No.81L, detection tube, Gastec, Kanagawa, Japan).

Fecal microflora

The samples of fresh fecal were collected by rectal massage at 6 and 13 weeks from 4 pigs in each treatment by rectal massage. The samples were immediately packaged in plastic bags and transferred to the laboratory freezer (−20℃) for the duration of the experiment. To count the number of *Lactobacillus* and *Escherichia coli* (*E. coli*), 1 g of samples from each treatment were diluted with 9 mL of 1 % peptone broth (Becton, Dickinson and Co) and homogenized. In 6-fold to 4-fold dilution (1 % peptone solution) samples were used to analyze the viability of *E. coli* on MacConkey agar plates (Difco Laboratories, Detroit, MI, USA) and *Lactobacillus* on de Man, Rogosa, and Sharpe agar plates (Difco Laboratories) respectively. *E. coli* were incubated at 37℃ for 24 h and *Lactobacillus* were incubated for 48 h.

Experiment 2

Experimental Design, Animals, and Housing

This study used swine manure and sawdust as raw materials for composting. Swine manure from CON of experiment 1 was obtained. The manure was collected from all CON replicates and mixed. The compost was prepared with the same method as experiment 1. The compost was matured in a 12 L plastic box and mixed weekly to provide oxygen and ripening at ambient temperature for 12 weeks. There were 6 replicates for each treatment. Experimental treatments were as follows: 1) CON, normal compost without probiotics; 2) T1, spray *B. subtilis* 10 g per 3.306 m^2; 3) T2, spray *B. subtilis* 40 g per 3.306 m^2; 4) T3, spray *S. cerevisiae* 10 g per 3.306 m^2; 5) T4, spray *S. cerevisiae* 40 g per 3.306 m^2; 6) T5, spray *B. subtilis* 5 g + *S. cerevisiae* 5 g per 3.306 m^2; 7) T6, spray *B. subtilis* 20 g + *S. cerevisiae* 20 g per 3.306 m^2. In this study, 2.0×10^{10} CFU kg^{-1} of *B. subtilis* and 3.0×10^{10} CFU

kg^{-1} of *S. cerevisiae* were used.

Measurements and sampling

Moisture content of compost

The collected compost samples were determined before and after drying at 105℃ for 24 h to analyze for MC according to methods and calculating formula described by Singh et al. [15]. Calculating the MC used the following formula:

$$\text{"MC (\%)} = \{(W_w - W_d)/W_w)\} \times 100\text{"}$$

W_w = weight of the sample before drying, W_d = weight of the sample after drying.

Maturity score of compost

The measurement methods are the same as experiment 1.

Odorous gas emissions of compost

Initially, the samples were agitated and collected 300 g on 1, 4, 8, and 12 weeks, respectively. About 300 g samples were stored in a 4.2 L plastic box at room temperature 26℃ and fermented for 72 h. The plastic boxes with small holes sealed with plaster were used for analyzing H_2S, NH_3, and methyl mercaptan (CH_3SH) emissions of samples. The samples with plastic boxes are shaken for 20 s to break down any crust formation before the measurement. H_2S concentrations were determined within scope of 2.0–20.0 ppm (No.4LK, detection tube, Gastec), NH_3 concentrations were determined within scope of 5.0–100.0 ppm (No.3La, detection tube, Gastec), and CH_3SH concentrations were determined within scope of 2.5–70.0 ppm (No.71, detection tube, Gastec).

Statistical analysis

All data excluding compost maturity were analyzed with the PROC General Linear Models procedure of SAS 9.4 software (version 9.4, SAS Institute, Cary, NC, USA). The maturity score was analyzed with a Chi-square test using the FREQ procedure of SAS. The GraphPad Prism 8 software (GraphPad Software, San Diego, CA, USA) was used to visualize the maturity score and MC. Tukey's multiple range test was used as post-hoc test was used to analyze the differences between means and a $p < 0.05$ was considered statistically significant for analysis.

Results

Experiment 1

Growth performance

The effects of supplemental probiotics on growth performance are presented in Table 2. There was no significant difference ($p > 0.05$) in ADG, ADFI and FCR in 0–6 weeks and 6–13 weeks among treatments. In BW, there was no significant difference ($p > 0.05$) at initial, 6 weeks and 9 weeks, respectively. However, T3 showed significantly higher ($p < 0.05$) BW than CON in 13 weeks, significantly higher ($p < 0.05$) ADG than CON and T2 in 9–13 weeks, and significantly lower ($p < 0.05$) FCR than the other groups for 9-13 weeks and the overall experimental period (0–13 weeks), respectively. Additionally, T3 showed significantly higher ($p < 0.001$) ADG than other groups in the overall experimental period.

Nutrient digestibility

The effects of supplemental probiotics on nutrient digestibility are presented in Table 3. There was

https://doi.org/10.5187/jast.2023.e88

Table 2. Effects of single and complex probiotics supplementation on growth performance in growing-finishing pigs (Exp 1)

Items	CON[1)]	T1	T2	T3	SE	*p*-value
BW (kg)						
0 week	18.52	18.81	18.84	18.83	0.974	0.995
6 weeks	51.01	51.73	51.77	52.17	1.237	0.931
9 weeks	70.38	71.44	71.25	72.32	1.387	0.811
13weeks	95.98[b]	98.83[ab]	97.97[ab]	101.29[a]	1.279	0.047
0–6 weeks						
ADG (kg)	0.77	0.78	0.79	0.79	0.010	0.561
ADFI (kg)	1.70	1.72	1.69	1.70	0.033	0.923
FCR (kg/kg)	2.20	2.20	2.16	2.14	0.036	0.569
6–9 weeks						
ADG (kg)	0.92	0.94	0.93	0.96	0.018	0.455
ADFI (kg)	2.41	2.46	2.42	2.46	0.048	0.815
FCR (kg/kg)	2.62	2.63	2.62	2.56	0.047	0.724
9–13 weeks						
ADG (kg)	0.91[b]	0.98[ab]	0.95[b]	1.03[a]	0.026	0.021
ADFI (kg)	2.87	2.84	2.86	2.87	0.054	0.978
FCR (kg/kg)	3.15[a]	2.91[b]	3.00[b]	2.79[c]	0.040	< 0.001
0–13 weeks						
ADG (kg)	0.85[c]	0.88[b]	0.87[bc]	0.91[a]	0.007	< 0.001
ADFI (kg)	2.22	2.24	2.22	2.23	0.021	0.896
FCR (kg/kg)	2.61[a]	2.55[a]	2.55[a]	2.47[b]	0.024	0.001

[1)]CON, basal diet; T1, CON + 0.2% *Bacillus subtilis*; T2, CON + 0.2% *Saccharomyces cerevisiae*; T3, CON + 0.2% *Bacillus subtilis* + 0.2% *Saccharomyces cerevisiae*.

[a–c]Means in the same row with different letters indicate different significantly ($p < 0.05$).

BW, body weight; ADG, average daily gain; ADFI, average daily feed intake; FCR, feed conversion ratio.

Table 3. Effects of single and complex probiotics supplementation on nutrient digestibility in growing-finishing pigs (Exp 1)

Items	CON[1)]	T1	T2	T3	SE	*p*-value
6 weeks						
DM	84.01	84.46	84.77	84.88	0.287	0.193
GE	74.95	75.38	75.77	75.96	0.347	0.225
CP	70.72	71.29	71.15	72.79	0.592	0.130
13 weeks						
DM	83.32[b]	83.73[ab]	83.74[ab]	84.03[a]	0.144	0.032
GE	70.55	71.51	71.70	72.64	0.545	0.113
CP	70.56	70.65	70.82	71.63	0.805	0.780

[1)]CON, basal diet; T1, CON + 0.2% *Bacillus subtilis*; T2, CON + 0.2% *Saccharomyces cerevisiae*; T3, CON + 0.2% *Bacillus subtilis* + 0.2% *Saccharomyces cerevisiae*. [a,b]Means in the same row with different letters indicate different significantly ($p < 0.05$).

DM, dry matter; GE, gross energy; CP, crude protein.

no significant difference ($p > 0.05$) in DM, GE, and CP at 6 weeks among treatments. Also, there was no significant difference ($p < 0.05$) in GE, and CP at 13 weeks among treatments. However, T3 showed significantly higher ($p < 0.05$) DM than CON at 13 weeks.

Blood profiles

The effects of supplemental probiotics on blood profile are presented in Table 4. There was no

Table 4. Effects of single and complex probiotics supplementation on blood characteristics in growing-finishing pigs (Exp 1)

Items	CON[1)]	T1	T2	T3	SE	*p*-value
6 weeks						
Total protein (g/dL)	5.78	6.00	6.03	6.03	0.187	0.742
BUN (mg/dL)	8.25[a]	6.75[b]	7.25[b]	6.50[b]	0.260	0.002
Creatinine (mg/dL)	1.18	1.19	1.40	1.39	0.080	0.121
WBC (10^3/μL)	23.03	23.47	23.18	23.21	0.794	0.983
RBC (10^6/μL)	7.80	7.69	7.99	7.86	0.168	0.657
13 weeks						
Total Protein (g/dL)	6.28	6.70	6.58	6.70	0.168	0.284
BUN (mg/dL)	14.00	12.25	13.00	13.25	1.365	0.839
Creatinine (mg/dL)	1.20	1.21	1.43	1.41	0.084	0.141
WBC (10^3/μL)	17.00	19.02	18.04	17.43	1.180	0.658
RBC (10^6/μL)	6.75	6.94	7.14	6.77	0.117	0.121

[1)]CON, basal diet; T1, CON + 0.2% *Bacillus subtilis*; T2, CON + 0.2% *Saccharomyces cerevisiae*; T3, CON + 0.2% *Bacillus subtilis* + 0.2% *Saccharomyces cerevisiae*. [a,b]Means in the same row with different letters indicate different significantly ($p < 0.05$).
BUN, blood urea nitrogen; WBC, white blood cell, RBC, red blood cell.

significant difference ($p > 0.05$) in total protein, creatinine, WBC, and RBC at 6 weeks among treatments. Probiotics did not affect the total protein, BUN, creatinine, WBC, and RBC at 13 weeks. However, supplementation of probiotic groups was significantly lower ($p < 0.05$) BUN than non-supplementation of the probiotic group at 6 weeks.

Compost maturity

The effects of supplemental probiotics on the maturity score of pigs (Exp 1) are presented in Fig. 1. The chi-square test showed no difference ($p > 0.05$) in the maturity score among treatments. However, supplementation of probiotic groups was significantly higher ($p < 0.001$) average maturity

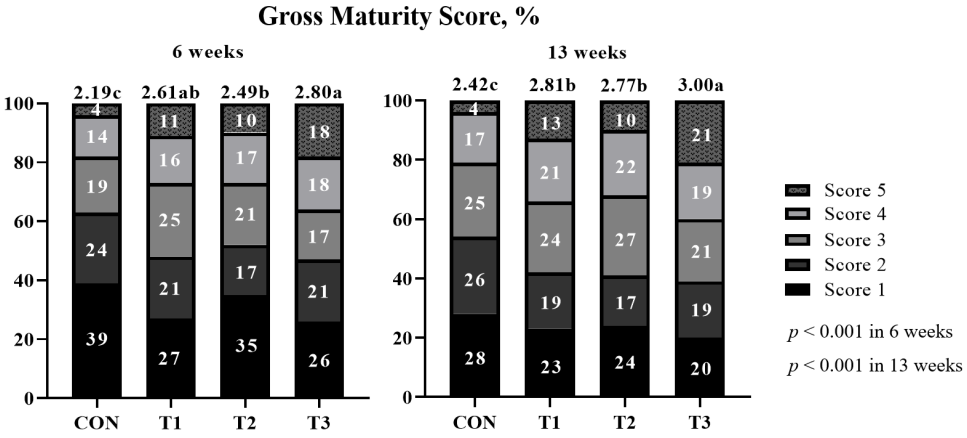

Fig. 1. Effects of supplemental probiotics on maturity score from pigs (Exp 1). 6 weeks, collected manure 2 pigs per pen at 6 weeks and composting with sawdust; 13 weeks, collected manure 2 pigs per pen at 13 weeks and composting with sawdust; CON, basal diet; T1, CON + 0.2% *Bacillus subtilis*; T2. CON + 0.2% *Saccharomyces cerevisiae*; T3, CON + 0.2% *Bacillus subtilis* + 0.2% *Saccharomyces cerevisiae*. n = 6 pen/treatment. $\chi^2 = 13.972$, $p = 0.303$ in 6 weeks. $\chi^2 = 13.387$, $p = 0.342$ in 13 weeks. Numbers inside the bar indicates percentage of score out of total (100%) as shown in legend. [a–c]Means scores with different upward letters in the graph bar different significantly by the one-way ANOVA ($p < 0.05$).

https://doi.org/10.5187/jast.2023.e88

score than non-supplementation of the probiotic group at 6 weeks. Moreover, T3 showed a significantly higher ($p < 0.001$) average maturity score than other groups at 13 weeks.

Odorous gas emissions

The effects of supplemental probiotics on gas emissions are presented in Table 5. There was no significant difference ($p > 0.05$) in H_2S, NH_3, and CH_3COOH at 6 weeks among treatments. Also, there was no significant difference ($p > 0.05$) in NH_3 and CH_3COOH at 13 weeks among treatments. However, T3 showed significantly lower ($p < 0.05$) H_2S than CON at 13 weeks.

Fecal microflora

The effects of supplemental probiotics on fecal microflora are presented in Table 6. There was no significant difference ($p > 0.05$) in *Lactobacillus* at 6 weeks among treatments. Also, there was no significant difference ($p > 0.05$) in *E. coli* at 13 weeks among treatments. However, T1 and T3 showed significantly lower ($p < 0.001$) *E. coli* than CON and T2 at 6 weeks. Moreover, T3 showed significantly higher ($p < 0.05$) *Lactobacillus* than CON and T2 at 13 weeks.

Experiment 2

Moisture content of compost

The effects of supplemental probiotics on MC of compost are presented in Fig. 2. Supplementation of probiotic groups was only numerically decreasing ($p > 0.05$) MC compared to the non-supplementation group for 6 to 12 weeks.

Table 5. Effects of single and complex probiotics supplementation on odorous gas emissions in growing-finishing pigs (Exp 1)

Items (ppm)	CON[1)]	T1	T2	T3	SE	*p*-value
6 weeks						
H_2S	5.47	5.32	5.42	5.23	0.180	0.788
NH_3	8.10	7.99	8.00	7.95	0.113	0.815
CH_3COOH	3.11	3.09	3.10	3.06	0.049	0.888
13 weeks						
H_2S	6.42[a]	6.23[ab]	6.31[ab]	6.11[b]	0.066	0.036
NH_3	9.35	9.24	9.27	9.21	0.083	0.685
CH_3COOH	3.14	3.06	3.10	3.04	0.041	0.384

[1)]CON, basal diet; T1, CON + 0.2% *Bacillus subtilis*; T2, CON + 0.2% *Saccharomyces cerevisiae*; T3, CON + 0.2% *Bacillus subtilis* + 0.2% *Saccharomyces cerevisiae*. [a,b]Means in the same row with different letters indicate different significantly ($p < 0.05$).

H_2S, hydrogen sulfide; NH_3, ammonia; CH_3COOH, acetic acid.

Table 6. Effects of single and complex probiotics supplementation on fecal bacteria counts in growing-finishing pigs (Exp 1)

Items (Log CFU/g)	CON[1)]	T1	T2	T3	SE	*p*-value
6 weeks						
Lactobacillus	8.89	9.01	8.98	9.07	0.158	0.864
E. coli	6.43[a]	6.21[b]	6.33[a]	6.15[b]	0.038	< 0.001
13 weeks						
Lactobacillus	9.02[c]	9.09[ab]	9.08[bc]	9.14[a]	0.020	0.003
E. coli	6.45	6.33	6.36	6.30	0.798	0.976

[1)]CON, basal diet; T1, CON + 0.2% *Bacillus subtilis*; T2, CON + 0.2% *Saccharomyces cerevisiae*; T3, CON + 0.2% *Bacillus subtilis* + 0.2% *Saccharomyces cerevisiae*. [a–c]Means in the same row with different letters indicate different significantly ($p < 0.05$).

E. coli, *Escherichia coli*.

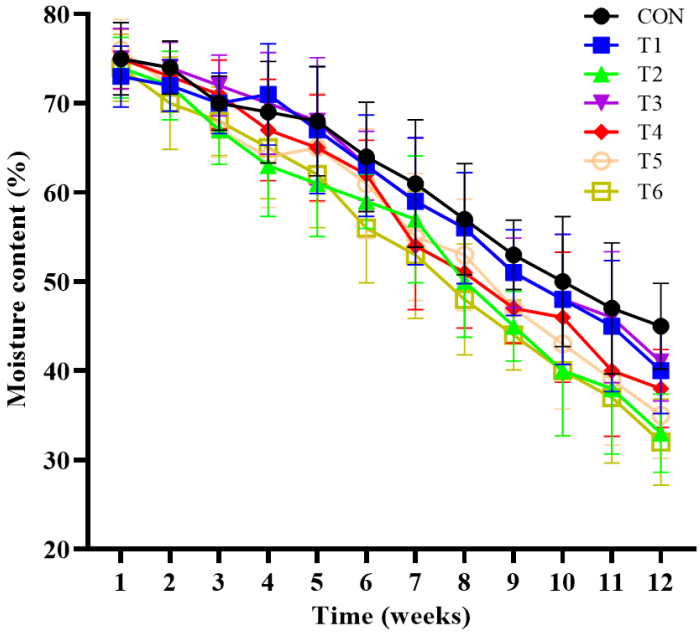

Fig. 2. Effects of probiotics supplementation on moisture content change in growing-finishing pigs manure compost during composting (Exp 2). CON, normal compost without probiotics; T1, spray *Bacillus subtilis* 10 g per 3.306 m^2; T2, spray *Bacillus subtilis* 40 g per 3.306 m^2; T3, spray *Saccharomyces cerevisiae* 10 g per 3.306 m^2; T4, spray *Saccharomyces cerevisiae* 40 g per 3.306 m^2; T5, spray (*Bacillus subtilis* 5 g + *Saccharomyces cerevisiae* 5 g) per 3.306 m^2; T6, spray (*Bacillus subtilis* 20 g + *Saccharomyces cerevisiae* 20 g) per 3.306 m^2; T6, spray (*Bacillus subtilis* 20 g + *Saccharomyces cerevisiae* 20 g) per 3.306 m^2. Bars denote standard errors.

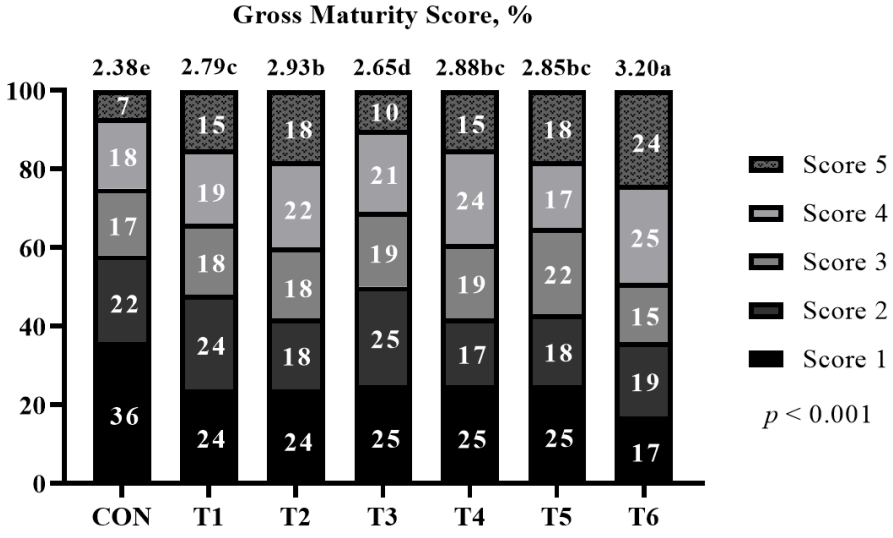

Fig. 3. Effects of supplemental probiotics on maturity score (Exp 2). CON, normal compost without probiotics; T1, spray *Bacillus subtilis* 10 g per 3.306 m^2; T2, spray *Bacillus subtilis* 40 g per 3.306 m^2; T3, spray *Saccharomyces cerevisiae* 10 g per 3.306 m^2; T4, spray *Saccharomyces cerevisiae* 40 g per 3.306 m^2; T5, spray (*Bacillus subtilis* 5 g + *Saccharomyces cerevisiae* 5 g) per 3.306 m^2; T6, spray (*Bacillus subtilis* 20 g + *Saccharomyces cerevisiae* 20 g) per 3.306 m^2; T6, spray (*Bacillus subtilis* 20 g + *Saccharomyces cerevisiae* 20 g) per 3.306 m^2. n = 6 pen/treatment. $\chi^2 = 19.558$, $p = 0.722$. Numbers inside the bar indicates percentage of score out of total (100%) as shown in legend. [a–e]Means scores with different upward letters in the graph bar different significantly by the ANOVA ($p < 0.05$).

https://doi.org/10.5187/jast.2023.e88

Table 7. Effects of probiotics supplementation on gas emissions in growing-finishing pigs manure compost (Exp 2)

Items, ppm	CON[1)]	T1	T2	T3	T4	T5	T6	SE	*p*-value
1 week									
H_2S	6.00	3.00	2.47	3.18	2.49	2.52	1.85	0.980	0.124
NH_3	16.64[a]	8.83[b]	6.24[bc]	8.98[b]	5.29[c]	5.34[c]	2.43[d]	0.922	< 0.001
CH_3SH	2.86	1.45	1.24	1.53	1.30	1.31	0.99	0.504	0.232
4 weeks									
H_2S	1.09	0.41	0.34	0.42	0.36	0.36	0.19	0.205	0.111
NH_3	5.05[a]	2.17[b]	1.67[b]	2.34[b]	1.92[b]	1.99[b]	0.80[c]	0.212	< 0.001
CH_3SH	0.95	0.81	0.69	0.82	0.71	0.72	0.52	0.097	0.127
8 weeks									
H_2S	0.30	0.15	0.12	0.16	0.13	0.12	0.09	0.059	0.289
NH_3	0.70	0.31	0.25	0.32	0.26	0.26	0.16	0.126	0.129
CH_3SH	0.11	0.07	0.05	0.08	0.07	0.07	0.03	0.198	0.243
12 weeks									
H_2S	0.01	0.01	0.00	0.01	0.00	0.00	0.00	0.003	0.123
NH_3	0.02	0.01	0.00	0.13	0.01	0.01	0.00	0.004	0.156
CH_3SH	0.01	0.01	0.00	0.01	0.01	0.01	0.00	0.005	0.605

[1)]CON, normal compost without probiotics; T1, spray *Bacillus subtilis* 10 g per 3.306 m^2; T2, spray *Bacillus subtilis* 40 g per 3.306 m^2; T3, spray *Saccharomyces cerevisiae* 10 g per 3.306 m^2; T4, spray *Saccharomyces cerevisiae* 40 g per 3.306 m^2; T5, spray (*Bacillus subtilis* 5 g + *Saccharomyces cerevisiae* 5 g) per 3.306 m^2; T6, spray (*Bacillus subtilis* 20g + *Saccharomyces cerevisiae* 20 g) per 3.306 m^2.

[a–d]Means in the same row with different letters indicate different significantly ($p < 0.05$).

H_2S, hydrogen sulfide; NH_3, ammonia; CH_3SH, methyl mercaptan.

Compost maturity

The effects of supplemental probiotics on MC of compost are presented in Fig. 3. Supplementation of probiotic groups was only numerically decreasing ($p > 0.05$) MC compared to the non-supplementation group for 6 to 12 weeks.

Odorous gas emissions of compost

The effects of supplemental probiotics on gas emissions of compost are presented in Table 7. There was no significant difference ($p > 0.05$) in H_2S and CH_3SH emissions of compost during the overall measurement period among treatments. Although there was no significant difference in NH_3 emission at 8 and 12 weeks among treatments, T6 showed significantly lower ($p < 0.05$) NH_3 emissions than other groups at 1 week and 4 weeks, respectively.

DISCUSSION

Experiment 1

In addition to promoting the growth of beneficial bacteria, probiotics may also produce microbicidal substances that have effects against harmful microbes and gastrointestinal pathogens [16,17]. Furthermore, probiotics can improve growth performance by improving digestion, absorption, and uptake of nutrients in pigs [18]. Especially, *Bacillus* spp. can produce various digestive enzymes to degrade complex carbohydrates in feed and improve feed utilization [19]. Previous studies have indicated that supplementation of *B. subtilis* and *B. licheniformis* can increase ADFI and ADG [20] and decrease FCR [21] in pigs. The addition of *Lactobacillus acidophilus*, *S. cerevisiae*, and *B. subtilis* can also increase ADG [22] in pigs. Likewise, the results of this study revealed that the inclusion

of complex probiotics such as *B. subtilis* and *S. cerevisiae* in the diets of growing-finishing pigs increased ADG and ADFI, while decreasing FCR. These study results agree with previous studies showing that complex probiotics have enhanced benefits in the gastrointestinal tract by integrating effects of different strains compared with a single probiotic [23].

The improved growth performance after adding probiotics might be related to enhanced nutrient digestibility by improving the gastrointestinal tract [24]. The mechanism of probiotics involves production of antimicrobials that can affect the composition and function of microbial communities, thus promoting overall gut health [25]. Previous studies have indicated that supplementation of complex probiotics (*B. subtilis*, *Clostridium butyricum*, *B. liceniformis,* and *B. coagulans*) can improve DM and nitrogen digestibility in growing-finishing pigs that addition of complex probiotics (*B. subtilis* and *S. cerevisiae*) can improve DM and GE digestibility in growing pigs [26,27]. Similarly, in this study, dietary addition of probiotics to pigs improved DM digestibility in the grower phase. However, another study has suggested that Supplements containing complex probiotics (*B. subtilis*, *B. licheniformis*, and *S. cerevisiae*) has no effect on nutrient digestibility in growing pigs [28]. Such inconsistent results on nutrient digestibility might be due to different probiotic species and dose levels.

In the current study, there was no significant difference in blood profile including WBC, RBC, creatinine, or total protein after supplementing probiotics to diets. However, there was a significant decrease in BUN concentration in groups supplemented with probiotics. BUN concentration might be used as a method for quantifying nitrogen utilization in livestock [29]. In addition, Otsuka et al. [30] have reported that increased BUN concentration is associated with an increase in feed intake. However, another study has revealed that high concentrations of BUN-impaired kidneys are harmful to pigs [31]. Probiotics can increase the efficiency of nitrogen utilization, improve nitrogen utilization, and increase BUN concentrations in pigs [32,33]. On the other hand, other studies have demonstrated that supplementation of probiotics has no effect on blood profiles of growing pigs [24,34]. These results were probably due to feed intake time or amount and gender differences.

Immature manure can generate odorous gas and cause civil complaints in nearby livestock facilities [35]. Its solutions include reducing nitrogen excretion in urine and feces and supplying feed additives to improve gastrointestinal microbial manipulation [18]. Scheuermann [36] has reported that supplementation of *Lactobacillus* in growing pigs can increase nitrogen retention and reduce nitrogen content in manure. In addition, Ramons et al. [37] have revealed that a reduction of nitrogen content can accelerate maturity period. Similarly, the present study showed that manure composting of dietary supplementation probiotics pigs accelerated the maturity period. These results might reduce BUN to enhance pig intestinal N retention. However, there have been few studies that have measured the maturity of manure excreted after feeding probiotics to pigs, so more research is needed.

High levels of noxious gases such as NH_3, volatile sulfur, and volatile organic compounds can negatively affect animal health and performance. They, not only affect the health of workers but also cause environmental pollution [18]. Volatile sulfur-degrading properties of *Bacillus* spp. and increased absorption of nutrients in the gut by *S. cerevisiae* can reduce the substrate for microbial fermentation and decrease emissions of these gases [38,39]. Prior studies have reported that addition of *Bacillus*-based can reduce H_2S emissions in growing pigs [24] and sows [40]. At the end of the experiment, H_2S emissions were reduced in groups supplemented with probiotics. Supplementation of complex probiotics significantly decreased H_2S from 4 weeks. This indicates that *Bacillus*-based complex probiotics might have potential to reduce gas emissions in pigs and improve air quality of swine farms efficiently with positive effects on pigs.

E. coli and *Lactobacillus* are representative intestinal pathogens and beneficial bacteria, respectively.

https://doi.org/10.5187/jast.2023.e88

Moreover, these bacteria are associated with gastrointestinal conditions, health status, and immune system [41]. Prior studies have shown that *S. cerevisiae* can decrease the level of potential pathogens in the intestinal lumen and generate antibacterial substances and that *Bacillus* can generate some effective enzymes (such as α-amylase, α-galactosidase, β-glucanase, β-mannanase, cellulase) to improve the intestinal condition [42,43]. In this study, adding complex probiotics increased *Lactobacillus* but decreased *E. coli*. Similarly, previous studies have reported that supplementation of probiotics can increase the counts of gastrointestinal *lactobacillus* but decrease the counts of *E. coli* [26,44]. The present result was consistent with Balasubramanian et al. [26] showing that continuous feeding of probiotics could maintain beneficial intestine microbiota by generating organic acids and hydrogen peroxide, thereby preventing pathogenic bacteria activation into the intestine and excreting antagonistic activity.

Experiment 2

In compost, there are beneficial microorganisms that take charge of regular composting process and potentially harmful microorganisms for humans and the environment. These deactivations of harmful microorganisms and beneficial microbiome development are important goals of composting [45]. In addition, previous studies have reported that providing sufficient quantity of probiotics as beneficial microorganisms could enhance microbial enzyme activity and offset effects of pathogenic microorganisms [46,47].

In addition to microorganisms, factors that affect composting include porosity, aeration, moisture, and temperature [48]. Especially, low values of MC, an important environmental parameter during composting, can cause premature dehydration known to arrest biological processes, resulting in biologically unstable compost, while high values of MC will halt composting activity due to creation of anaerobic conditions caused by water logging [49]. Besides, moisture is related to heat capacity in compost [50]. It can influence metabolic activities of probiotics [51]. Lee et al. [52] have reported that microorganisms produce heat during enzymatic catabolism of substrates and synthesis of cell material. Therefore, we hypothesize that reducing MC during composting due to microorganisms could generate heat as they decompose organic material. However, in this study, single and complex probiotics (*B. subtilis*, *S. cerevisiae*) supplementation only numerically decreased MC compared to the non-supplemented group. Since no study has reported the relationship between probiotics and MC of compost, further research is needed.

Normal composting involving aerobic decomposition proceeds with the following phases: (i) fermentation, (ii) acid formation, (iii) thermophilic activity, and (iv) temperature decline [53]. Shortening the thermophilic activity phase during the degradation phase can delay the maturity period [54]. Prior studies have suggested that adding *B. subtilis* can prolong the thermophilic phase by increasing high-temperature-resistant bacteria [55]. Xu and Li. [56] have shown that inoculating *B. subtilis*, *B. licheniformis*, *Phanerochaete chrysosporium*, *Trichoderma koningii*, and *S. cerevisiae* into compost can promote compost maturity. Results of this study agree with those of previous studies showing that supplementation of *B. subtilis* and *S. cerevisiae* can reduce the maturation time of composting [57]. For this reason, complex probiotics such as *B. subtilis* can be used to accelerate the maturation.

Accelerating compost mature period can improve several problems such as greenhouse gas emissions, including loss of nitrogen via NH_3 volatilization, and leaking of inorganic/organic pollutants from compost substrates [58]. In general, the genus *Bacillus* grows by assimilating ammonium nitrogen during composting, which causes NH_3 emissions reduction [59]. A prior study has suggested that supplementation of *Bacillus*-based probiotics can reduce emissions of gases such as NH_3, H_2S, CH_3COOH, CO_2, and CH_3SH [60]. In addition, supplementation of *S.*

cerevisiae can reduce NH_3 by 10.2% and amine gas by 45.5% in swine manure [61]. In this study, supplementation of probiotics decreased NH_3 emissions compared with non-supplementation. However, other odorous gas emissions (CH_3SH and H_2S) were not affected by supplementation of probiotics. These inconsistent results are attributed to temperature and humidity.

CONCLUSION

This study indicates that supplementation of probiotics at the complex probiotics are more improved on growth performance, nutrient digestibility, blood profile, compost maturity, gas emissions, and fecal microflora in pigs and on MC, compost maturity, and gas emissions in compost than single and non-supplementation. Therefore, these results revealed that complex probiotics (*S. cerevisiae* and *B. subtilis*) had positive effects in pigs and compost, respectively. However, supplementation of complex probiotics in compost rarely investigates so more studies are needed.

REFERENCES

1. Jang YN, Hwang O, Jung MW, Ahn BK, Kim H, Jo G, et al. Comprehensive analysis of microbial dynamics linked with the reduction of odorous compounds in a full-scale swine manure pit recharge system with recirculation of aerobically treated liquid fertilizer. Sci Total Environ. 2021;777:146122. https://doi.org/10.1016/j.scitotenv.2021.146122
2. Park S, Cho S, Hwang O. Effects of Italian ryegrass (IRG) supplementation on animal performance, gut microbial compositions and odor emission from manure in growing pigs. Agronomy. 2020;10:647. https://doi.org/10.3390/agronomy10050647
3. Kim E, Kim BU, Kim HC, Kim S. Sensitivity of fine particulate matter concentrations in South Korea to regional ammonia emissions in Northeast Asia. Environ Pollut. 2021;273:116428. https://doi.org/10.1016/j.envpol.2021.116428
4. Loyon L. Overview of manure treatment in France. Waste Manag. 2017;61:516-20. https://doi.org/10.1016/j.wasman.2016.11.040
5. Awasthi MK, Wang Q, Ren X, Zhao J, Huang H, Awasthi SK, et al. Role of biochar amendment in mitigation of nitrogen loss and greenhouse gas emission during sewage sludge composting. Bioresour Technol. 2016;219:270-80. https://doi.org/10.1016/j.biortech.2016.07.128
6. Lu Y, Gu W, Xu P, Xie K, Li X, Sun L, et al. Effects of sulphur and Thiobacillus thioparus 1904 on nitrogen cycle genes during chicken manure aerobic composting. Waste Manag. 2018;80:10-6. https://doi.org/10.1016/j.wasman.2018.08.050
7. Vasquez R, Oh JK, Song JH, Kang DK. Gut microbiome-produced metabolites in pigs: a review on their biological functions and the influence of probiotics. J Anim Sci Technol. 2022;64:671-95. https://doi.org/10.5187/jast.2022.e58
8. Li J, Kim IH. Effects of Saccharomyces cerevisiae cell wall extract and poplar propolis ethanol extract supplementation on growth performance, digestibility, blood profile, fecal microbiota and fecal noxious gas emissions in growing pigs. Anim Sci J. 2014;85:698-705. https://doi.org/10.1111/asj.12195
9. Wang Y, Cho JH, Chen YJ, Yoo JS, Huang Y, Kim HJ, et al. The effect of probiotic BioPlus 2B® on growth performance, dry matter and nitrogen digestibility and slurry noxious gas emission in growing pigs. Livest Sci. 2009;120:35-42. https://doi.org/10.1016/j.livsci.2008.04.018
10. Gong YL, Liang JB, Jahromi MF, Wu YB, Wright AG, Liao XD. Mode of action of

https://doi.org/10.5187/jast.2023.e88

Saccharomyces cerevisiae in enteric methane mitigation in pigs. Animal. 2018;12:239-45. https://doi.org/10.1017/S1751731117001732
11. NRC [National Research Council]. Nutrient requirement of swine. 11th rev. ed. Washington, DC: The National Academies Press; 2012.
12. Williams CH, David DJ, Iismaa O. The determination of chromic oxide in faeces samples by atomic absorption spectrophotometry. J Agric Sci. 1962;59:381-5. https://doi.org/10.1017/S002185960001546X
13. AOAC [Association of Official Analytical Chemists] International. Official methods of analysis of AOAC International. 18th ed. Washington, DC: AOAC International; 2007.
14. Song JM, Phong NH, Kim JY, Kang DS, Yu JY, Kang HW. Physicochemical changes and plant growth effect on composting of spent mushroom substrates. J Mushroom. 2020;18:268-73. https://doi.org/10.14480/JM.2020.18.3.268
15. Singh J, Kalamdhad AS. Assessment of compost quality in agitated pile composting of water hyacinth collected from different sources. Int J Recycl Org Waste Agric. 2015;4:175-83. https://doi.org/10.1007/s40093-015-0097-z
16. Lee SH, Ingale SL, Kim JS, Kim KH, Lokhande A, Kim EK, et al. Effects of dietary supplementation with Bacillus subtilis LS 1–2 fermentation biomass on growth performance, nutrient digestibility, cecal microbiota and intestinal morphology of weanling pig. Anim Feed Sci Technol. 2014;188:102-10. https://doi.org/10.1016/j.anifeedsci.2013.12.001
17. Mun D, Kyoung H, Kong M, Ryu S, Jang KB, Baek J, et al. Effects of Bacillus-based probiotics on growth performance, nutrient digestibility, and intestinal health of weaned pigs. J Anim Sci Technol. 2021;63:1314-27. https://doi.org/10.5187/jast.2021.e109
18. Kim YJ, Cho SB, Song MH, Lee SI, Hong SM, Yun W, et al. Effects of different Bacillus licheniformis and Bacillus subtilis ratios on nutrient digestibility, fecal microflora, and gas emissions of growing pigs. J Anim Sci Technol. 2022;64:291-301. https://doi.org/10.5187/jast.2022.e12
19. Hu J, Kim IH. Effect of Bacillus subtilis C-3102 spores as a probiotic feed supplement on growth performance, nutrient digestibility, diarrhea score, intestinal microbiota, and excreta odor contents in weanling piglets. Animals. 2022;12:316. https://doi.org/10.3390/ani12030316
20. Ahmed ST, Hoon J, Mun HS, Yang CJ. Evaluation of Lactobacillus and Bacillus-based probiotics as alternatives to antibiotics in enteric microbial challenged weaned piglets. Afr J Microbiol Res. 2014;8:96-104. https://doi.org/10.5897/AJMR2013.6355
21. Jørgensen JN, Laguna JS, Millán C, Casabuena O, Gracia MI. Effects of a Bacillus-based probiotic and dietary energy content on the performance and nutrient digestibility of wean to finish pigs. Anim Feed Sci Technol. 2016;221:54-61. https://doi.org/10.1016/j.anifeedsci.2016.08.008
22. Chen YJ, Son KS, Min BJ, Cho JH, Kwon OS, Kim IH. Effects of dietary probiotic on growth performance, nutrients digestibility, blood characteristics and fecal noxious gas content in growing pigs. Asian-Australas J Anim Sci. 2005;18:1464-8. https://doi.org/10.5713/ajas.2005.1464
23. Adamberg S, Sumeri I, Uusna R, Ambalam P, Kondepudi KK, Adamberg K, et al. Survival and synergistic growth of mixed cultures of bifidobacteria and lactobacilli combined with prebiotic oligosaccharides in a gastrointestinal tract simulator. Microb Ecol Health Dis. 2014;25:23062. https://doi.org/10.3402/mehd.v25.23062
24. Kang J, Lee JJ, Cho JH, Choe J, Kyoung H, Kim SH, et al. Effects of dietary inactivated probiotics on growth performance and immune responses of weaned pigs. J Anim Sci Technol. 2021;63:520-30. https://doi.org/10.5187/jast.2021.e44

25. Luise D, Bertocchi M, Motta V, Salvarani C, Bosi P, Luppi A, et al. Bacillus sp. probiotic supplementation diminish the Escherichia coli F4ac infection in susceptible weaned pigs by influencing the intestinal immune response, intestinal microbiota and blood metabolomics. J Anim Sci Biotechnol. 2019;10:74. https://doi.org/10.1186/s40104-019-0380-3
26. Balasubramanian B, Lee SI, Kim IH. Inclusion of dietary multi-species probiotic on growth performance, nutrient digestibility, meat quality traits, faecal microbiota and diarrhoea score in growing–finishing pigs. Ital J Anim Sci. 2018;17:100-6. https://doi.org/10.1080/1828051X.2017.1340097
27. Liu W, Devi S, Park J, Kim I. Effects of complex probiotic supplementation in growing pig diets with and without palm kernel expellers on growth performance, nutrient digestibility, blood parameters, fecal microbial shedding and noxious gas emission. Anim Sci J. 2018;89:552-60. https://doi.org/10.1111/asj.12965
28. Wang H, Ha BD, Kim IH. Effects of probiotics complex supplementation in low nutrient density diet on growth performance, nutrient digestibility, faecal microbial, and faecal noxious gas emission in growing pigs. Ital J Anim Sci. 2021;20:163-70. https://doi.org/10.1080/1828051X.2020.1801358
29. Sampath V, Han K, Sureshkumar S, Kim IH. Impact of yeast hydrolysate (Saccharomyces cerevisiae) supplementation on the growth performance, nutrient digestibility, fecal microflora, noxious gas emission, blood profile, and meat quality of finishing pigs. Can J Anim Sci. 2022;102:98-107. https://doi.org/10.1139/cjas-2021-0056
30. Otsuka M, Ishida A, Nakayama Y, Saito M, Yamazaki M, Murakami H, et al. Dietary supplementation with cellooligosaccharide improves growth performance in weanling pigs. Anim Sci J. 2004;75:225-9. https://doi.org/10.1111/j.1740-0929.2004.00180.x
31. Liang Z, Ren Z, Gao S, Chen Y, Yang Y, Yang D, et al. Individual and combined effects of deoxynivalenol and zearalenone on mouse kidney. Environ Toxicol Pharmacol. 2015;40:686-91. https://doi.org/10.1016/j.etap.2015.08.029
32. Tan BF, Lim T, Boontiam W. Effect of dietary supplementation with essential oils and a Bacillus probiotic on growth performance, diarrhoea and blood metabolites in weaned pigs. Anim Prod Sci. 2021;61:64-71. https://doi.org/10.1071/AN18752
33. Jang YD, Oh HK, Piao LG, Choi HB, Yun JH, Kim YY. Evaluation of probiotics as an alternative to antibiotic on growth performance, nutrient digestibility, occurrence of diarrhea and immune response in weaning pigs. J Anim Sci Technol. 2009;51:25-32. https://doi.org/10.5187/JAST.2009.51.1.025
34. Lefter NA, Hăbeanu M, Gheorghe A, Dumitru M, Gal C, Vlaicu PA. Effects of microencapsulated probiotics on performance, organ development, diarrhoea incidences, blood parameters, intestinal histomorphology and microflora in weaning piglets. Agriculture. 2023;13:39. https://doi.org/10.3390/agriculture13010039
35. Prasad CS, Anandan S, Gowda NKS, Schlecht E, Buerkert A. Managing nutrient flows in Indian urban and peri-urban livestock systems. Nutr Cycl Agroecosyst. 2019;115:159-72. https://doi.org/10.1007/s10705-018-9964-0
36. Scheuermann SE. Effect of the probiotic Paciflor® (CIP 5832) on energy and protein metabolism in growing pigs. Anim Feed Sci Technol. 1993;41:181-9. https://doi.org/10.1016/0377-8401(93)90011-8
37. Ramos RF, Santana NA, de Andrade N, Romagna IS, Tirloni B, de Oliveira Silveira A, et al. Vermicomposting of cow manure: effect of time on earthworm biomass and chemical, physical, and biological properties of vermicompost. Bioresour Technol. 2022;345:126572. https://doi.org/10.1016/j.biortech.2021.126572

https://doi.org/10.5187/jast.2023.e88

38. Lan R, Kim IH. Effects of Bacillus licheniformis and Bacillus subtilis complex on growth performance and faecal noxious gas emissions in growing-finishing pigs. J Sci Food Agric. 2019;99:1554-60. https://doi.org/10.1002/jsfa.9333
39. Sampath V, Duk Ha B, Kibria S, Kim IH. Effect of low-nutrient-density diet with probiotic mixture (Bacillus subtilis ms1, B. licheniformis SF5-1, and Saccharomyces cerevisiae) supplementation on performance of weaner pigs. J Anim Physiol Anim Nutr. 2022;106:61-8. https://doi.org/10.1111/jpn.13544
40. Jeong J, Kim J, Lee S, Kim I. Evaluation of Bacillus subtilis and Lactobacillus acidophilus probiotic supplementation on reproductive performance and noxious gas emission in sows. Ann Anim Sci. 2015;15:699-710. https://doi.org/10.1515/aoas-2015-0018
41. Lee JH, Lee B, Rousseau X, Gomes GA, Oh HJ, Kim YJ, et al. Stimbiotic supplementation modulated intestinal inflammatory response and improved broilers performance in an experimentally-induced necrotic enteritis infection model. J Anim Sci Biotechnol. 2022;13:100. https://doi.org/10.1186/s40104-022-00753-9
42. Latorre JD, Hernandez-Velasco X, Wolfenden RE, Vicente JL, Wolfenden AD, Menconi A, et al. Evaluation and selection of Bacillus species based on enzyme production, antimicrobial activity, and biofilm synthesis as direct-fed microbial candidates for poultry. Front Vet Sci. 2016;3:95. https://doi.org/10.3389/fvets.2016.00095
43. Czerucka D, Rampal P. Experimental effects of Saccharomyces boulardii on diarrheal pathogens. Microbes Infect. 2002;4:733-9. https://doi.org/10.1016/S1286-4579(02)01592-7
44. Huang S, Rong X, Liu M, Liang Z, Geng Y, Wang X, et al. Intestinal mucosal immunity-mediated modulation of the gut microbiome by oral delivery of Enterococcus faecium against Salmonella enteritidis pathogenesis in a laying hen model. Front Immunol. 2022;13:853954. https://doi.org/10.3389/fimmu.2022.853954
45. Tanaji CS, Shahaji SP, Suhas JA. Stabilization of dairy industry sludge with leaf litter using as composting and its effect on Spinacia oleracea plant growth. Mater Today Proc. 2023;73:455-63. https://doi.org/10.1016/j.matpr.2022.09.600
46. Dubreuil JD. Enterotoxigenic Escherichia coli and probiotics in swine: what the bleep do we know? Biosci Microbiota Food Health. 2017;36:75-90. https://doi.org/10.12938/bmfh.16-030
47. Greff B, Szigeti J, Nagy Á, Lakatos E, Varga L. Influence of microbial inoculants on co-composting of lignocellulosic crop residues with farm animal manure: a review. J Environ Manage. 2022;302:114088. https://doi.org/10.1016/j.jenvman.2021.114088
48. Bernal MP, Alburquerque JA, Moral R. Composting of animal manures and chemical criteria for compost maturity assessment. A review. Bioresour Technol. 2009;100:5444-53. https://doi.org/10.1016/j.biortech.2008.11.027
49. Liang C, Das KC, McClendon RW. The influence of temperature and moisture contents regimes on the aerobic microbial activity of a biosolids composting blend. Bioresour Technol. 2003;86:131-7. https://doi.org/10.1016/S0960-8524(02)00153-0
50. Ahn HK, Sauer TJ, Richard TL, Glanville TD. Determination of thermal properties of composting bulking materials. Bioresour Technol. 2009;100:3974-81. https://doi.org/10.1016/j.biortech.2008.11.056
51. Khater ESG. Some physical and chemical properties of compost. Int J Waste Resour. 2015;5:72-9. https://doi.org/10.4172/2252-5211.1000172
52. Lee D, Goh TW, Kang MG, Choi HJ, Yeo SY, Yang J, et al. Perspectives and advances in probiotics and the gut microbiome in companion animals. J Anim Sci Technol. 2022;64:197-217. https://doi.org/10.5187/jast.2022.e8
53. Ahmad S, Khalid R, Abbas S, Hayat R, Ahmed I. Potential of compost for sustainable crop

production and soil health. In: de Mandal SD, Passari AK, editors. Recent advancement in microbial biotechnology: agricultural and industrial approach. London: Academic Press; 2021. p. 123-70.
54. Wang K, Li W, Li Y, Gong X, Wu C, Ren N. The modelling of combined strategies to achieve thermophilic composting of sludge in cold region. Int Biodeterior Biodegradation. 2013;85:608-16. https://doi.org/10.1016/j.ibiod.2013.03.005
55. Li C, Li H, Yao T, Su M, Li J, Liu Z, et al. Effects of microbial inoculation on enzyme activity, available nitrogen content, and bacterial succession during pig manure composting. Bioresour Technol. 2020;306:123167. https://doi.org/10.1016/j.biortech.2020.123167
56. Xu P, Li J. Effects of microbial inoculant on physical and chemical properties in pig manure composting. Compost Sci Util. 2017;25:S37-42. https://doi.org/10.1080/1065657X.2017.1295886
57. Jiang J, Liu X, Huang Y, Huang H. Inoculation with nitrogen turnover bacterial agent appropriately increasing nitrogen and promoting maturity in pig manure composting. Waste Manag. 2015;39:78-85. https://doi.org/10.1016/j.wasman.2015.02.025
58. Xiao R, Awasthi MK, Li R, Park J, Pensky SM, Wang Q, et al. Recent developments in biochar utilization as an additive in organic solid waste composting: a review. Bioresour Technol. 2017;246:203-13. https://doi.org/10.1016/j.biortech.2017.07.090
59. Kuroda K, Tanaka A, Furuhashi K, Nakasaki K. Application of Bacillus sp. TAT105 to reduce ammonia emissions during pilot-scale composting of swine manure. Biosci Biotechnol Biochem. 2017;81:2400-6. https://doi.org/10.1080/09168451.2017.1389607
60. Sureshkumar S, Park JH, Kim IH. A preliminary evaluation on mixed probiotics as an antimicrobial spraying agent in growing pig barn. J Anim Sci Technol. 2022;64:1035-45. https://doi.org/10.5187/jast.2022.e69
61. Kim JA, Bayo J, Cha J, Choi YJ, Jung MY, Kim DH, et al. Investigating the probiotic characteristics of four microbial strains with potential application in feed industry. PLOS ONE. 2019;14:e0218922. https://doi.org/10.1371/journal.pone.0218922

https://doi.org/10.5187/jast.2023.e88