Effects of Herbiotic FS supplementation on growth performance, nutrient digestibility, blood profiles, and faecal scores in weanling pigs

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ABSTRACT
This study was conducted to investigate the effects of Herbiotic FS supplementation on growth performance, nutrient digestibility, blood characteristics, and faecal scores in weanling pigs. A total of 150 weanling pigs [(Landrace×Yorkshire)×Duroc] with an average initial body weight of 8.02 ± 0.92 kg were randomly assigned to 5 dietary treatments (5 replicate pens per treatment with 6 pigs per pen). The dietary treatments were as follows: (1) negative control: basal diet, (2) positive control: basal diet + 150 mg/kg apramycin, (3) Herbiotic1: basal diet + 250 mg/kg Herbiotic FS, (4) Herbiotic2 (basal diet + 500 mg/kg Herbiotic FS), and (5) Herbiotic3: (basal diet + 250 mg/kg Herbiotic FS + 75 mg/kg apramycin). During days 1–7, pigs fed positive control, Herbiotic2 and Herbiotic3 diets had improved average daily gain and gain:feed ratio compared with those fed negative control or Herbiotic1 diets (P < 0.05). On day 21, pigs fed positive control and Herbiotic3 diets had increased nitrogen digestibility compared with those fed negative control or Herbiotic1 diets (P < 0.05). In conclusion, Herbiotic FS supplementation decreased the pathogenic count in the gut owing to its antimicrobial activity and this in turn caused the nutrients more available to the animal contributing to the growth.

1. Introduction
During the last decades, phytotherapeutic feed additives have been widely used as an alternative to antibiotics because of their plant-derived property and growth-promoting effects (Hong et al. 2004; Wang et al. 2007; Wang et al. 2008; Windisch et al. 2008; Jang et al. 2010; Ao et al. 2011). Valchev et al. (2009) demonstrated that herbal extracts (150 mg/kg) in pig diet could increase growth performance, feed efficiency, and immune-related blood characteristics. It is suggested that the beneficial effects of herbs or herbal extracts may arise from increased feed intake and secretion of digestive enzymes, immune stimulation, anti-bacterial, anti-viral, and antioxidant properties (Wenk 2003; Kim et al. 2010). Krotkiewski and Janiak (2008) and Yang et al. (2009) also demonstrated a synergistic interaction between the components of herbal mixture in vivo and in vitro.

Herbiotic FS is a mixture of thyme, buckwheat, turmeric, black pepper, and ginger, and is developed for a better and safer alternative to antibiotics and chemical growth promoters. It has lower minimum inhibitory concentration as compared to zinc bacitracin and other synthetic antibiotics and has better growth-promoting effects (Dinodiya et al. 2015). Herbiotic FS augmented the population of beneficial microbiota in the gut through its prebiotic oligosaccharides, prevented the irritation of the intestine and increased the number and height of the intestinal villi in broilers (Dinodiya 2012). However, the information about Herbiotic FS supplementation in weanling pigs is still scarce. This study was conducted to investigate the effects of Herbiotic FS supplementation on growth performance, nutrient digestibility, blood characteristics, and diarrhoea score in weanling pigs.

2. Materials and methods
The experiment was conducted at the Experimental Unit of the Dankook University (Cheonan, Republic of Korea). The protocol for the current experiment was approved by the Animal Care and Use Committee of Dankook University.

2.1. Experimental design and animals
A total of 150 pigs [(Landrace × Yorkshire) × Duroc] with an average initial body weight of 8.02 ± 0.92 kg were assigned to 5 treatments (5 replicate pens per treatment and 6 pigs per pen) in a randomized complete block. The experiment lasted for 6 weeks. The dietary treatments were as follows: (1) negative control: basal diet (NC), (2) positive control: basal diet + 150 mg/kg apramycin (PC), (3) basal diet + 250 mg/kg Herbiotic FS (H1), (4) basal diet + 500 mg/kg Herbiotic FS (H2), and (5) basal diet + 250 mg/kg Herbiotic FS + 75 mg/kg apramycin (H3). The Herbiotic FS used in this study included buckwheat (15.00%), thyme (7.50%), turmeric (3.75%), black pepper (1.25%), and ginger (1.25%). A 3-period feeding programme was employed in the current experiment, which consisted of phase 1 (days 1–7), phase 2 (days 8–21), and phase 3 (days 22–42). All diets (Table 1) were formulated to meet or exceed the nutrient requirements of pigs.
The DM and N in feed and faecal samples were determined by oven-drying at 70°C for 72 h, after which feed and faecal samples were stored immediately at −20°C until analysis. Faecal samples were collected via jugular venipuncture into both a nonheparinized and a K3EDTA vacuum tube (Becton Dickinson Vacutainer Systems, Franklin Lakes, NJ) to enable evaluation of the serum and whole blood, respectively. The white blood cell (WBC), red blood cell (RBC) and lymphocyte counts were analysed using an automatic blood analyser (ADVIA 120, Bayer, USA). The serum samples were then centrifuged (2000 × g) for 30 minutes at 4°C, after which the immunoglobulin G (IgG) was determined using a nephelometer analyser (Behring, Marburg, Germany).

Subjective diarrhoea score was recorded daily from days 1 to 7 by the same person and was based on the following: 1 = well-formed faeces, 2 = sloppy faeces, and 3 = diarrhoea. Score was recorded on a pen basis following observations of individual pig and signs of stool consistency in the pen. The score is reported as average daily diarrhoea of individual pig score.

### 2.3. Statistical analysis

In this experiment, all data were analysed using a randomized complete block design following general linear model procedures of SAS (1996), with each pen being used as the experimental unit. Differences between treatments were detected by Tukey’s multiple range test. The data were expressed as means and pooled standard error of the mean. Significance was defined as *P* < 0.05.

### 3. Results

#### 3.1. Growth performance

During phase 1, pigs fed PC, H2 and H3 diets had improved (*P* < 0.05) ADG and G:F compared with those fed NC and H1 diets, but ADFI was not affected (*P* > 0.10) by treatments (Table 2). During phase 2, ADFI in pigs fed PC and H3 was increased (*P* < 0.05) compared with those fed NC diet, whereas ADG and G:F were unaffected (*P* > 0.10). During phase 3, pigs fed PC and H3 diets showed higher (*P* < 0.05) ADG than that of pigs fed NC diet, whereas ADG and G:F were not affected (*P* > 0.10). Overall, pigs fed PC, H2, and H3 diets had improved (*P* < 0.05) ADG when compared with those fed NC diet, although ADG and G:F were not affected (*P* > 0.10).

#### 3.2. Nutrient digestibility

Pigs fed PC and H3 diets had increased (*P* < 0.05) ATTD of N compared with those fed NC and H1 diets (Table 3) at day 21.

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**Table 1.** Compositions of basal weanling pig diets for experiment 1 (as-fed basis).

| Items | Phase 1 (days 1–7) | Phase 2 (days 8–21) | Phase 3 (days 22–42) |
|-------|------------------|-------------------|------------------|
| Ingredient, g/kg | | | |
| Extruded corn | 111.5 | 349.2 | 451.0 |
| Extruded oat | 100.0 | — | — |
| Biscuit meal | — | 50.0 | 90.0 |
| Soybean meal | 80.0 | 200.0 | 296.5 |
| Fermented soybean meal | 78.0 | 82.0 | — |
| Fish meal | 50.0 | 40.0 | 25.0 |
| Soy oil | 41.5 | 48.0 | 30.0 |
| Lactose | 100.0 | 60.0 | — |
| Whey | 170.0 | 107.0 | 68.5 |
| Milk product* | 13.00 | 20.0 | 20.0 |
| Monocalcium phosphate | 12.5 | 10.0 | 6.0 |
| Sugar | 40.0 | 20.0 | — |
| Plasma powder | 65.0 | — | — |
| L-Lys·HCl, 78% | 1.2 | 2.5 | 1.6 |
| DL-Met, 50% | 2.6 | 1.5 | 1.4 |
| L-Thr, 89% | 7.7 | 0.8 | — |
| Choline chloride, 25% | 2.0 | 1.0 | 1.0 |
| Vitamin premixa | 1.0 | 1.0 | 1.0 |
| Trace mineral premixa | 2.0 | 2.0 | 2.0 |
| Limestone | 2.0 | 2.0 | 3.0 |
| Salt | 3.0 | 3.0 | 3.0 |
| Calculated composition, g/kg | | | |
| Metabolizable energy, MJ/kg | 14.8 | 14.8 | 14.6 |
| Crude protein | 220.0 | 210.0 | 205.0 |
| Lys | 15.7 | 14.1 | 13.3 |
| Met | 6.0 | 4.9 | 4.7 |
| Calcium | 8.0 | 7.8 | 7.5 |
| Total phosphorus | 7.6 | 7.6 | 6.4 |
| Lys | 226.3 | 215.4 | 207.9 |
| Calcium | 8.1 | 7.9 | 7.4 |
| Phosphorus | 7.8 | 7.4 | 6.7 |

*Provided per kg of complete diet: vitamin A, 11,025 IU; vitamin D₃, 1,103 IU; vitamin E, 44 IU; vitamin K, 4.4 mg; d-pantothenic, 29 mg; choline, 166 mg; and vitamin B₁₂, 3 μg.*

*Provided per kg of complete diet: Fe (as FeSO₄·7H₂O), 80 mg; Cu (as CuSO₄·5H₂O), 4 mg; d-pantothenic, 29 mg; choline, 166 mg; and vitamin B₁₂, 3 μg.*
Table 2. Effect of Herbiotic FS on growth performance in weanling pigs.

| Dietary treatments | NC     | PC     | H1     | H2     | H3     | SEM | P-value |
|--------------------|--------|--------|--------|--------|--------|-----|---------|
| Phase 1, days 1–7  |        |        |        |        |        |     |         |
| ADG, g             | 180b   | 210a   | 184b   | 212a   | 219b   | 5.8 | 0.010   |
| ADFI, g            | 254    | 265    | 257    | 264    | 260    | 7.5 | 0.521   |
| G:F                | 0.709b | 0.793a | 0.716b | 0.803b | 0.842a | 0.04| 0.021   |
| Phase 2, days 8–21 |        |        |        |        |        |     |         |
| ADG, g             | 487    | 521    | 497    | 516    | 509    | 15.7| 0.010   |
| ADFI, g            | 707b   | 775a   | 768ab  | 764ab  | 772a   | 12.1| 0.025   |
| G:F                | 0.689  | 0.672  | 0.647  | 0.675  | 0.659  | 0.03| 0.113   |
| Phase 3, days 22–42|        |        |        |        |        |     |         |
| ADG, g             | 605b   | 632a   | 624ab  | 629ab  | 638a   | 10.5| 0.021   |
| ADFI, g            | 992    | 1,005  | 995    | 999    | 1,000  | 21.3| 1.020   |
| G:F                | 0.610  | 0.629  | 0.627  | 0.630  | 0.638  | 0.06| 0.995   |
| Overall, days 1–42 |        |        |        |        |        |     |         |
| ADG, g             | 495b   | 525a   | 508ab  | 526a   | 525a   | 7.2 | 0.023   |
| ADFI, g            | 774    | 805    | 796    | 798    | 801    | 10.2| 0.556   |
| G:F                | 0.640  | 0.652  | 0.638  | 0.659  | 0.655  | 0.05| 0.441   |

Notes: The dietary treatments were as follows: (1) NC, basal diet; (2) PC, basal diet + 150 mg/kg apramycin; (3) H1, basal diet + 250 mg/kg Herbiotic FS; (4) H2, basal diet + 500 mg/kg Herbiotic FS; (5) H3, basal diet + 250 mg/kg Herbiotic FS + 75 mg/kg apramycin. Abbreviation: ADFI: average daily feed intake; ADG: average daily gain; G:F: gain:feed ratio; SEM: standard error of the mean.

a,bMeans in the same row with different superscripts differ (P < 0.05).

However, other determined ATTD were not affected by dietary treatments (P > 0.10).

3.3. Blood profiles

On day 7, RBC concentration in PC, H2 and H3 treatments was higher (P < 0.05) than that in NC treatment (Table 4). Pigs fed H1, H2, and H3 diets had increased (P < 0.05) lymphocyte (day 21) and IgG (day 21) concentrations compared with those fed NC and PC diets. No other effects were observed among treatments (P > 0.10).

3.4. Diarrhoea score

No significant difference (P > 0.10) was observed on diarrhoea score among dietary treatments during the experiment (Table 5).

Table 3. Effect of Herbiotic FS on apparent total tract digestibility in weanling pigs.

| Dietary treatments | NC     | PC     | H1     | H2     | H3     | SEM | P-value |
|--------------------|--------|--------|--------|--------|--------|-----|---------|
| Day 7              |        |        |        |        |        |     |         |
| Dry matter         | 79.25  | 79.52  | 80.16  | 80.46  | 81.02  | 1.11| 0.887   |
| Nitrogen           | 79.47  | 79.83  | 80.15  | 79.52  | 80.53  | 1.26| 1.112   |
| Gross energy       | 81.25  | 80.15  | 80.14  | 80.94  | 80.57  | 0.88| 0.994   |
| Day 21             |        |        |        |        |        |     |         |
| Dry matter         | 80.25  | 80.17  | 80.56  | 80.53  | 81.46  | 0.95| 0.884   |
| Nitrogen           | 79.24b | 82.19b | 79.24b | 80.60ab| 82.92a | 0.61| 0.001   |
| Gross energy       | 79.56  | 81.99  | 79.85  | 81.94  | 80.65  | 1.10| 0.992   |
| Day 42             |        |        |        |        |        |     |         |
| Dry matter         | 78.41  | 80.11  | 79.40  | 80.26  | 80.37  | 1.28| 0.555   |
| Nitrogen           | 79.01  | 80.26  | 80.19  | 80.04  | 81.66  | 1.43| 0.233   |
| Gross energy       | 79.51  | 81.94  | 81.46  | 81.20  | 80.29  | 1.80| 0.611   |

Notes: The dietary treatments were as follows: (1) NC, basal diet; (2) PC, basal diet + 150 mg/kg apramycin; (3) H1, basal diet + 250 mg/kg Herbiotic FS; (4) H2, basal diet + 500 mg/kg Herbiotic FS; (5) H3, basal diet + 250 mg/kg Herbiotic FS + 75 mg/kg apramycin. Abbreviation: SEM: standard error of the mean.

a,bMeans in the same row with different superscripts differ (P < 0.05).
apramycin or a combination of 250 mg/kg Herbiotic FS and 75 mg/kg apramycin increased the ATTD of N, whereas no effects on ATTD of DM and GE were observed with any of the dietary treatments. Herbs may increase activity of digestive enzymes of gastric mucosa and balance the intestinal microbiota, thereby improving nutrient digestibility (Jamroz et al. 2003; Huang et al. 2012). The improved ATTD of N may help to, at least partially, explain the improved growth performance.

The gastrointestinal tract is the largest immunological competent organ in the body, and studies have indicated that the maturation of gastrointestinal tract and the development of the immune system depend on the composition of the indigenous microbiota (Insoft et al. 1996; de Vrese and Marteau 2007). Inclusion of herbs or herbal extracts could influence the gut microbiota, thereby improving nutrient digestibility (Jamroz et al. 2003; Huang et al. 2012). The increased ATTD of N may help to, at least partially, explain the improved growth performance.

In the present study, pigs fed with Herbiotic FS diets showed increased RBC (day 7), IgG (day 21), and lymphocyte (day 42) concentrations compared to pigs fed with NC and PC diets, suggesting that Herbiotic FS had beneficial effects on immune function. Herbs may stimulate immune function in livestock by influencing the growth of pathogenic microorganisms in gastrointestinal ecosystems (Wenk 2003; Ali et al. 2008; Windisch et al. 2008). The improved immunity function may consequently contribute to the improved growth performance.

Post-weaning diarrhoea is one of the serious problems faced in swine production, especially within the first week after weaning. Herbs may reduce diarrhoea occurrence via improving intestinal microbial balance. Huang et al. (2012) reported that inclusion of medical herbs reduced diarrhoea score during the first 10 days of the experiment period. However, in the present study, diarrhoea score was not affected by dietary treatments. Similarly, Zhang et al. (2012) demonstrated that phytoncide supplementation (flavonoid, phenolic compounds, alkaloid, tannin, terpene, and saponin) had no effect on diarrhoea score. The lack of effect on diarrhoea score in the present can be explained by the generally good health status of the pigs and good hygienic conditions of the research facilities.

In conclusion, Herbiotic FS supplementation decreased the pathogenic count in the gut owing to its antimicrobial activity and this in turn causes the nutrients more available to the animal contributing to the growth.

Disclosure statement
No potential conflict of interest was reported by the authors.

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Table 4. Effect of Herbiotic FS on blood profiles in weanling pigs.

| Items          | NC    | PC    | H1    | H2    | H3    | SEM  | P-value |
|---------------|-------|-------|-------|-------|-------|------|---------|
| White blood cell, × 10^3/μL |       |       |       |       |       |      |         |
| Day 7         | 19.6  | 21.4  | 19.2  | 21.9  | 20.3  | 3.25 | 0.884  |
| Day 21        | 19.4  | 20.2  | 22.9  | 21.1  | 21.3  | 3.04 | 0.511  |
| Day 42        | 19.6  | 22.7  | 21.8  | 22.5  | 21.1  | 2.68 | 0.774  |
| Red blood cell, × 10^6/μL |       |       |       |       |       |      |         |
| Day 7         | 5.2^b | 6.1^a | 5.7^ab| 6.3^a | 6.2^a | 0.53 | 0.002  |
| Day 21        | 6.8   | 7.5   | 7.5   | 7.2   | 7.1   | 0.47 | 0.888  |
| Day 42        | 7.3   | 7.2   | 7.4   | 7.3   | 7.4   | 0.23 | 0.771  |
| Lymphocyte, % |       |       |       |       |       |      |         |
| Day 7         | 44.8  | 46.5  | 50.4  | 49.2  | 48.7  | 5.05 | 0.665  |
| Day 21        | 47.9  | 47.0  | 54.9  | 50.7  | 49.5  | 3.95 | 0.112  |
| Day 42        | 49.8^b| 47.4^a| 56.9^a| 54.3^a| 55.9^a| 3.08 | 0.020  |
| Ig G, mg/dL   |       |       |       |       |       |      |         |
| Day 7         | 410   | 426   | 445   | 462   | 458   | 83.2 | 0.102  |
| Day 21        | 567^b | 651^b | 815^a | 785^a | 724^a | 79.0 | 0.020  |
| Day 42        | 653   | 592   | 575   | 624   | 642   | 52.9 | 0.996  |

Notes: The dietary treatments were as follows: (1) NC, basal diet; (2) PC, basal diet + 150 mg/kg apramycin; (3) H1, basal diet + 250 mg/kg Herbiotic FS; (4) H2, basal diet + 500 mg/kg Herbiotic FS; (5) H3, basal diet + 250 mg/kg Herbiotic FS + 75 mg/kg apramycin. Abbreviation: IgG: immunoglobulin G; SEM: standard error of the mean. abMeans in the same row with different superscripts differ (P < 0.05).

Table 5. Effect of Herbiotic FS on diarrhoea score in weanling pigs.

| Dietary treatments | NC    | PC    | H1    | H2    | H3    |
|-------------------|-------|-------|-------|-------|-------|
| Diarrhoea score   | 2.0   | 1.5   | 1.8   | 1.6   | 1.5   |

Notes: The dietary treatments were as follows: (1) NC, basal diet; (2) PC, basal diet + 150 mg/kg apramycin; (3) H1, basal diet + 250 mg/kg Herbiotic FS; (4) H2, basal diet + 500 mg/kg Herbiotic FS; (5) H3, basal diet + 250 mg/kg Herbiotic FS + 75 mg/kg apramycin. Abbreviation: IgG: immunoglobulin G; SEM: standard error of the mean.

Subjective diarrhoea score was recorded daily from days 1 to 7 by the same person and were based on the following: 1 = well-formed faeces, 2 = sloppy faeces, 3 = diarrhoea.

Diarrhoea score was not affected by dietary treatments during the first 10 days of the experiment period. However, in the present study, diarrhoea score was not affected by dietary treatments. Similarly, Zhang et al. (2012) demonstrated that phytoncide supplementation (flavonoid, phenolic compounds, alkaloid, tannin, terpene, and saponin) had no effect on diarrhoea score.

The lack of effect on diarrhoea score in the present can be explained by the generally good health status of the pigs and good hygienic conditions of the research facilities.

In conclusion, Herbiotic FS supplementation decreased the pathogenic count in the gut owing to its antimicrobial activity and this in turn causes the nutrients more available to the animal contributing to the growth.

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