Effects of dietary supplementation of quillaja saponin or fructooligosaccharide and a mixture of both on the growth performance, nutrient utilisation, faecal microbial and faecal noxious gas emissions in growing pigs

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ABSTRACT

The objective of this experiment was to evaluate the effect of an alone or in combination quillaja saponin (QS) and fructooligosaccharide (FOS) on the growth performance, nutrient utilisation, faecal microbial and faecal noxious gas emissions in growing pigs. A total of 100 pigs (Landrace × Yorkshire × Duroc) were selected and randomly allocated to 4 treatments with 5 replicates per group and 5 pigs per replicate according to the initial body weight and sex. The experimental treatments were as follows: (1) basal diet (Ctrl); (2) Ctrl + 0.2% QS; (3) Ctrl + 0.10% FOS; (4) Ctrl + 0.2% QS + 0.10% FOS (QSFOS). The experiment period lasted for 56 days. Pigs fed a diet supplemented with QS, FOS and QSFOS resulted in an increase in body weight (BW) on days 28 and 56, average daily gain and average daily feed intake during days 28–56 and 50–56, as well as improved apparent total tract digestibility (ATTD) of dry matter and nitrogen (N) during day 56 compared with Ctrl. The faecal Lactobacillus counts were increased and a reduction in faecal Escherichia coli (E. coli) counts and ammonia (NH3) and carbon dioxide (CO2) emission from the faeces was observed from pigs fed QS, FOS and QSFOS supplemented diets compared with those fed a Ctrl diet. In conclusion, dietary QS, FOS and QSFOS supplementation have beneficial effects on growth performance, nutrient utilisation, faecal microbial and noxious gas emissions in growing pigs.

HIGHLIGHTS

- Dietary supplementation of QS, FOS and QSFOS increased body weight, average daily gain and average daily feed intake of growing pigs.
- Dietary supplementation of QS, FOS and QSFOS could improve ATTD of dry matter and nitrogen of growing pigs.
- Dietary supplementation of QS, FOS and QSFOS increased Lactobacillus counts and reduced E. coli counts of growing pigs.
- Dietary supplementation of QS, FOS and QSFOS could reduce faecal NH3 and CO2 emissions of growing pigs.

Introduction

Phytogenic feed additives (PFAs) are plant-based products that are used to improve the performance of swine and poultry (Wati et al. 2015; Bafundo et al. 2020). Additionally, PFAs have become an additional tool for lowering NH3 emissions from swine production. Triterpenoid saponin is a surface-active triterpene glycoside, which is a naturally occurring compound in different fruits and plants. Triterpenoid saponin exerts biological and pharmacological activities such as anti-microbial (Kaczorek et al. 2016), anti-inflammatory (Sarkhel 2016), anti-fungal (Chludil et al. 2002), anti-oxidant (Chaudhary et al. 2018), anti-viral (Chludil et al. 2002). In livestock husbandry, triterpenoid saponins have beneficial effects on the production, intestinal health, immunity and meat quality of monogastric animals (Chaudhary et al. 2018). Quillaja saponin (QS) which is acquired from the soap bark (Quillaja Saponaria Molina) tree, contains a triterpenoid (non-steroidal) sapogenin structural component (Francis et al. 2002). Quillaja saponin exhibits various effects.
when used as a feed additive. Additionally, supplementation of Quillaja saponaria in diets is effective in improving growth performance, faecal microbiota and nutrient digestibility and decrease gas emissions in pigs (Bartoš et al. 2016; Dang and Kim 2020).

A prebiotic that has elicited a great interest to improve intestinal health and productivity in livestock is fructooligosaccharide(s) (FOS). FOS consists of several β(1-2)- or β(1-6)-linked fructose units, which may be linked to glucose residues (Gibson 1995; Estrada et al. 2001; Kolida et al. 2002). FOS are known for their ability to stimulate the growth of bifidobacteria (Davani-Davari et al. 2019) and to inhibit that of potentially pathogenic bacteria such as enterobacteria (Scott et al. 2014), clostridia (Kullen et al. 1998) and salmonella (Oyarzabal and Conner 1996). FOS has been shown to be resistant to intestinal glycolytic enzymes and to pass unaltered to the large intestine where they are fermented by the microflora (Prata et al. 2010). Fermentation of FOS leads to the production of short-chain fatty acids, which are a substrate for energy metabolism in the colonic mucosa, stimulating epithelial cell growth (Roediger 1982). FOS was recently reported to have antitumor and antiviral effects in vitro (Esawy et al. 2011). According to Chaudhary et al. (2018) and Lei et al. (2017) dietary FOS supplementation improve growth performance, nutrient digestibility, faecal microbiota, intestinal health and reduce faecal gas emissions in pigs.

We hypothesised that the inclusion of QS and FOS in combination may improve growth performance, nutrient utilisation and reduce ammonia emissions in pigs. To our knowledge, no data is available about the combined supplementation of QS and FOS of growing pigs. Therefore, the objective of this study was to evaluate the effects of combined or single supplementation of QS and FOS to the diets on growth performance, nutrient utilisation, faecal microbial and faecal noxious gas emissions in growing pigs.

Materials and methods

Animal ethics

All animal procedures used in this experiment were reviewed and approved by the Institutional Animal Care and Use Committee of Dankook University (Cheonan, Republic of Korea). (DK-1-1934).

Experimental design, animals and housing

A total of 100 pigs (Landrace × Yorkshire × Duroc); weighted 23.83 ± 1.95 kg, were selected and randomly allocated to 4 treatments with 5 replicates per group and 5 pigs per replicates according to the initial body weight and sex. The experimental treatments were as follows: (1) basal diet (control diet; Ctrl); (2) Ctrl + 0.2% QS (QS) (DelaconBiotechnik GmbH, Steyrregg, Austria); (3) Ctrl + 0.10% FOS (FOS) (FOS-MAX; Dreamfeed Inc., Seoul, South Korea); (4) Ctrl + 0.2% QS + 0.10% FOS (QSFOS). The diets were formulated to meet or exceed the National Research Council recommendation (NRC 2012) for growing pigs (Table 1). During the 56-day experimental period, all pigs had free access to feed and water.

Sampling and measurements

Pigs were weighted on days 0, 28 and 56 of the trial as well as the feed consumption to calculate average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR = ADFI/ADG) were recorded.

On days 21–28 and 50–56, pigs were fed diets containing 0.25% chromic oxide (Cr₂O₃) added to the diet as an indigestible marker. At the end of days 28 and 56, pigs’ rectums were gently massaged by trained

Table 1. Composition of finishing pig diets (as fed-basis).

| Ingredients                  | % Content |
|------------------------------|-----------|
| Corn                         | 64.19     |
| Soybean meal                 | 17.43     |
| Sesame meal                  | 2.00      |
| Distillers dried grains with soluble (Corn, USA) | 4.00 |
| Palm kernel meal             | 2.00      |
| Condensed molasses soluble   | 1.50      |
| Lipid                        | 4.20      |
| Molasses                     | 1.50      |
| Limestone                    | 1.06      |
| Dicalcium phosphate          | 0.31      |
| Salt                         | 0.25      |
| Methionine, 99%              | 0.14      |
| Lysine, 50%                  | 0.71      |
| Threonine, 98.5%             | 0.14      |
| Tryptophan, 20%              | 0.25      |
| Vitamin mix<sup>a</sup>      | 0.10      |
| Mineral mix<sup>a</sup>      | 0.01      |
| Phytase                      | 0.01      |
| Total                        | 100.00    |

Calculated value, %

| Moisture                      | 13.29     |
| Crude protein                 | 15.87     |
| Crude fat                     | 7.17      |
| Crude fibre                   | 2.55      |
| Crude ash                     | 4.55      |
| Digestible non starch polysaccharide | 121 |
| Digestible energy (kcal kg⁻¹) | 2471      |
| Calcium                       | 0.70      |
| Total Phosphorus              | 0.38      |
| SID Lysine<sup>b</sup>        | 1.07      |

<sup>a</sup>Provided per kilogram of complete diet: vitamin A, 11,025 IU; vitamin D₃, 1750 IU; vitamin E, 44 IU; vitamin K, 4.4 mg; riboflavin, 8.3 mg; niacin, 50 mg; thiamine, 4 mg; d-pantothenic acid, 29 mg; choline, 166 mg and vitamin B₁₂, 33 μg. Fe (as FeSO₄ × 7H₂O), 80 mg; Cu (as CuSO₄ × 5H₂O), 12 mg; Zn (as ZnSO₄), 85 mg; Mn (as MnO₂), 8 mg; I (as KI), 0.28 mg and Se (as Na₂SeO₃ × 5H₂O), 0.15 mg.

<sup>b</sup>Standardised ileal digestible lysine.
personnel, and fresh faecal samples were collected from at least two pigs per pen (one gilt and one barrow) and then stored at $-20^\circ C$ for determination of the apparent total tract digestibility of nutrients. Moreover, approximately 1 kg of representative feed samples was harvested weekly during the experiment. The faecal samples were thawed at 4°C, then dried at 65°C for 72 h, and the samples of feed and faeces were ground to pass through a 1-mm sieve. Dry matter (DM) and nitrogen (N) were analysed in accordance with the Association of Official Analytical Chemists (AOAC 2005). Chromium concentrations were determined via UV absorption spectrophotometry (UV-1201, Shimadzu, Kyoto, Japan), and the apparent total tract digestibility (ATTD) of DM and N was calculated using indirect methods, as described by (Wang et al. 2021) for calculating the ATTD with the following equation.

$$\text{Apparent total tract digestibility (ATTD, %)} = 1 - \frac{(\text{Cr}_{\text{feed}} \times \text{Nutrient}_{\text{feces}})}{(\text{Cr}_{\text{feces}} \times \text{Nutrient}_{\text{feed}})}$$

On days 0, 28 and 56, faecal samples were collected via massaging the rectum from two pigs randomly selected from each pen (1 barrow and 1 gilt) and pooled and placed on ice for transportation to the laboratory, where analysis was immediately carried out. At the time of investigation, the content of each faecal was thawed and squeezed into sterile bottles and serially diluted in 0.85% sterile saline solution. An aliquot (0.1 mL) of each diluted sample was cultivated on a specific media [MacConkey Agar for coliforms (E. coli), and MRS agar for Lactic acid bacteria (lactobacilli)] and incubated at 37°C for 24 h. After incubation, the bacterial colonies were counted according to Wang et al. (2021). The counted bacteria were expressed as log CFU g$^{-1}$ of faecal sample.

On days 0, 28 and 56, faecal samples were taken from at least two pigs in each pen (1 barrow and 1 gilt) for analysing faecal gas emission and stored in 2.6 L plastic boxes in duplicate. Each box had a small hole in the middle of one sidewall, which was sealed with adhesive plaster. Noxious gas emissions were determined according to the method described by Wang et al. (2021). Briefly, a total of 300 g of faeces were stored in 2.6 L sealed plastic boxes in duplicates. These samples were permitted to ferment at 32°C for 30 h. After the fermentation period, an instrument (Gas Detector, GV-100S; Gastec Corp., Kanagawa, Japan) was used for gas detection. The plastic boxes were punctured and headspace air was sampled approximately 2.0 cm above the samples at a rate of 100 mL/min. Levels of ammonia (NH$_3$), hydrogen sulphide (H$_2$S), Carbon dioxide (CO$_2$), acetic acid and total mercaptans were measured using Gastec Detector Tube No. 3La, No. 4LK and No.70 L (Gastec Corp.), respectively.

### Statistical analysis

All data were subjected to statistical analysis in a randomised complete block based on sex and BW design using the GLM procedure of SAS (SAS Inst. Inc., Cary, NC, USA). The pen was considered as the experimental unit. Differences among treatments were separated by Tukey’s range test. Variability in the data is expressed as the standard error means. Probability values less than 0.05 were considered significant.

### Results

#### Growth performance

As shown in Table 2, the supplementation of QS, FOS and QS-FOS increased ($p < 0.05$) BW of growing pigs on days 28 and 56 compared with the Ctrl diet. Also, no differences were detected in ADG, ADFI and G:F between dietary treatments during the days 0–28. During days 28–56 and 0–56, dietary treatments showed no effect on FCR but pigs fed QS, FOS and QS-FOS diets had greater ($p < 0.05$) ADG and ADFI than pigs fed Ctrl diet.

#### Apparent total tract digestibility

On day 56, pigs fed QS, FOS and QS-FOS diets had an increase ($p < 0.05$) ATTD of dry matter and nitrogen than pigs fed Ctrl diet (Table 3).

#### Faecal microbial shedding

As shown in Table 4, on days 28 and 56, pigs fed diets supplemented with QS, FOS and QS-FOS had increased lactic acid bacteria counts and reduce coliform bacteria counts than those pigs fed Ctrl diet ($p < 0.05$).

#### Noxious gas emission

The effect of supplementation of FOS and QS (single or combined) on faecal noxious gas emission is presented in Table 5. The concentration of Methyl mercaptans, H$_2$S, and acetic acid emission did not differ between dietary treatments. However, pigs fed QS, FOS and QS-FOS diets had lower faecal NH$_3$ and CO$_2$ emission on days 28 and 56 ($p < 0.05$) compared with pigs fed Ctrl diet.
In the current study, the inclusion of the QS significantly improved pigs’ performance and nutrient digestibility. The positive performance response could be attributed to the synergistic effect between different active ingredients in this blend which are characterised by possessing anti-microbial, anti-inflammatory, anti-fungal, and anti-oxidant activities besides the stimulant property of the digestive enzymes with subsequent beneficial effects on the gut microbial ecosystem, nutrient digestibility and performance parameters. Significant improvements in the pigs’ performance and nutrient digestibility were observed due to feeding on diets supplemented by QS at levels of 0.2% diet during the growing phase. Previously, Dang and Kim (2020) and Bartoš et al. (2016) also reported that pigs fed QS had improved the growth performance and ATTD of DM and N. Similarly, Vaclavkova and Beckova (2008) reported that QS at a level of 125 mg/kg had increased the ADG and ADFI of weaning pigs. On the other hand, Ilsley et al. (2003) stated that dietary supplemented with 250 mg/kg QS had improved DM and N of sows. However, Kang et al. (2010) and Turner et al. (2000) did not observe a

### Table 2. Effects of dietary supplementation of quillaja saponin or fructooligosaccharide and a mixture of both on the growth performance in growing pigs.

| Items            | Ctrl  | QS    | FOS   | QSFO  | SEM  | p-Value |
|------------------|-------|-------|-------|-------|------|---------|
| Body weight, kg  |       |       |       |       |      |         |
| Day 0            | 23.84 | 23.82 | 23.82 | 23.82 | 0.010| 1.000   |
| Day 28           | 43.77 | 44.00 | 44.00 | 44.20 | 0.300| <.0001  |
| Day 56           | 66.20 | 67.09 | 66.80 | 67.47 | 0.510| 0.002   |
| Days 0–28        |       |       |       |       |      |         |
| ADG, g           | 712   | 725   | 723   | 729   | 11.000| 0.755   |
| ADFI, g          | 1634  | 1652  | 1653  | 1666  | 19.000| 0.698   |
| FCR              | 2.295 | 2.281 | 2.287 | 2.286 | 0.012| 0.695   |
| Days 28–56       |       |       |       |       |      |         |
| ADG, g           | 802a  | 856b  | 840ab | 882a  | 14.000| 0.013   |
| ADFI, g          | 2058a | 2187b | 2185b | 2246b | 24.000| 0.012   |
| FCR              | 2.566 | 2.551 | 2.560 | 2.533 | 0.021| 0.332   |
| Days 0–56        |       |       |       |       |      |         |
| ADG, g           | 752b  | 791ab | 782ab | 805a  | 9.000 | 0.018   |
| ADFI, g          | 1846a | 1920ab| 1919ab| 1956a | 14.000| 0.013   |
| FCR              | 2.438 | 2.424 | 2.431 | 2.417 | 0.013| 0.378   |

Abbreviations: ADG: average daily gain; ADFI: average daily feed intake; FCR: feed conversion ratio; Ctrl: basal diet; QS: quillaja saponin; FOS: fructooligosaccharide; SEM: standard error of the mean.

### Table 3. Effects of dietary supplementation of quillaja saponin or fructooligosaccharide and a mixture of both on the nutrient digestibility in growing pigs.

| Items, %          | Ctrl  | QS    | FOS   | QSFO  | SEM  | p-Value |
|-------------------|-------|-------|-------|-------|------|---------|
| Day 28            |       |       |       |       |      |         |
| Dry matter        | 77.14 | 78.10 | 78.89 | 78.49 | 0.80 | 0.425   |
| Nitrogen          | 75.57 | 76.65 | 77.86 | 76.64 | 0.78 | 0.336   |
| Day 56            |       |       |       |       |      |         |
| Dry matter        | 72.51b| 76.57ab| 76.36ab| 77.60a| 1.13 | 0.030   |
| Nitrogen          | 72.27b| 74.78ab| 76.03a | 76.57a| 1.21 | 0.036   |

Abbreviations: Ctrl: basal diet; QS: quillaja saponin; FOS: fructooligosaccharide; SEM: standard error of the mean.

### Table 4. Effects of dietary supplementation of quillaja saponin or fructooligosaccharide and a mixture of both on the on faecal microbial in growing pigs.

| Items, log10cfu/g | Ctrl  | QS    | FOS   | QSFO  | SEM  | p-Value |
|-------------------|-------|-------|-------|-------|------|---------|
| Day 0             |       |       |       |       |      |         |
| Lactobacillus     | 6.90  | 6.95  | 7.01  | 6.97  | 0.04 | 0.946   |
| E. coli           | 5.84  | 5.94  | 5.95  | 5.94  | 0.05 | 0.132   |
| Day 28            |       |       |       |       |      |         |
| Lactobacillus     | 7.02ab| 7.08ab| 7.14a | 7.15a | 0.03 | 0.048   |
| E. coli           | 5.99a | 5.80b | 5.77a | 5.76a | 0.06 | 0.024   |
| Day 56            |       |       |       |       |      |         |
| Lactobacillus     | 7.13b | 7.21ab| 7.26ab| 7.28a | 0.04 | 0.033   |
| E. coli           | 6.20a | 6.00ab| 6.05ab| 5.98a | 0.09 | 0.043   |

Abbreviations: Ctrl: basal diet; QS: quillaja saponin; FOS: fructooligosaccharide; SEM: Standard error of the mean.

### Discussion

In the current study, the inclusion of the QS significantly improved pigs’ performance and nutrient digestibility. The positive performance response could be attributed to the synergistic effect between different active ingredients in this blend which are characterised by possessing anti-microbial, anti-inflammatory, anti-fungal, and anti-oxidant activities besides the stimulant property of the digestive enzymes with subsequent beneficial effects on the gut microbial ecosystem, nutrient digestibility and performance parameters. Significant improvements in the pigs’
significant difference in the growth performance and nutrient digestibility of dietary supplementation of a 450 and 500 mg/kg QS. Similarly, Ilsley et al. (2005) reported that dietary inclusion of 750 mg/kg QS had no influence on growth performance in pigs.

FOS has been shown to be an excellent prebiotic fibre source, selectively enhancing the growth of beneficial gut microbiota in broilers while suppressing the growth of pathogenic bacteria. This results in health benefits, such as preventing constipation and reducing the absorption of heavy metal ions (Tungland 2003; Bogusławska-Tryk et al. 2012). In an experiment with fructooligosaccharides, an increase in villus height and crypt depth ratio in the distal small intestine was observed in pigs. The increased villus height to crypt depth ratio is favourable for nutrient absorption, and the increased absorptive capacity of the intestinal tract results in improved feed efficiency and faster growth (Shim 2005). The positive performance response could be attributed to the synergistic effect between different active ingredients in this blend which are characterised by possessing antioxidant, anti-inflammatory and antibacterial activities besides the stimulant property of the digestive enzymes with a subsequent positive impact on the gut microbial ecosystem, nutrient utilisation and performance parameters (Grela et al. 2021). In the present study, supplementation of 0.10% FOS significantly increased ADG and ADFI and ATTD of DM and N, which was consistent with Lei et al. (2018), who reported that the dietary inclusion of FOS increased ADG and ADFI and ATTD of DM and N in pigs. The beneficial effects of FOS on nutrient digestibility may be attributed to the effects of FOS on gastrointestinal development and the concomitant increase in digestive enzyme secretion (Hu and Wang 2001). Additionally, several studies have reported that fructooligosaccharides have beneficial effects on young pigs, improving their growth performance and intestinal health (Houdijk 1998; Oli et al. 1998; Shim et al. 2005). Similar results were also reported by Zhao et al. (2012) and Zhang and Kim (2014) in which pigs fed FOS diet had greater growth performance and ATTD of DM, and N in pigs compared with those fed the Ctrl dietary treatment. Reasons for these different results were likely due to different compositions of diets and different doses and sources of FOS suppletations.

In the current study, pigs fed a combination of QS and FOS improved the ADG and ADFI and ATTD of DM and N when compared with the Ctrl diet. However, there were differences in growth performance and nutrient utilisation between QS and QSFOS dietary treatments, indicating that supplementation of QS and FOS in combination exhibits synergism in growth performance and nutrient digestibility in growing pigs fed the Ctrl diet. Lei et al. (2017) investigated the supplemented with a combination of protease and FOS had improved growth performance and nutrient digestibility in growing pigs fed the Ctrl diet. Lei et al. (2017) investigated the supplemented with a combination of protease and FOS had improved growth performance and nutrient digestibility in growing pigs fed the Ctrl diet.

### Table 5. Effects of dietary supplementation of quillaja saponin or fructooligosaccharide and a mixture of both on the gas emission in growing pigs.

| Items, ppm | Ctrl | QS | FOS | QSFOS | SEM^2 | p-Value |
|-----------|------|----|-----|-------|-------|---------|
| Day 0     |      |    |     |       |       |         |
| Methyl mercaptans | 4.85 | 4.55 | 4.18 | 4.25 | 1.28 | 0.995 |
| NH₃       | 2.08 | 2.08 | 2.20 | 2.25 | 0.59 | 0.999 |
| H₂S       | 5.10 | 5.30 | 5.50 | 5.60 | 0.40 | 0.933 |
| CO₂       | 1325 | 1375| 1350 | 1375 | 245  | 1.000 |
| Acetic acid | 4.80 | 4.40 | 5.00 | 4.80 | 1.00 | 0.703 |
| Day 28    |      |    |     |       |       |         |
| Methyl mercaptans | 4.95 | 5.05 | 4.93 | 5.05 | 0.58 | 0.191 |
| NH₃       | 2.88 | 2.63 | 2.35 | 2.10 | 0.32 | 0.010 |
| H₂S       | 6.20 | 5.70 | 5.30 | 5.80 | 0.60 | 0.227 |
| CO₂       | 1525 | 1410| 1450 | 1400 | 243  | 0.034 |
| Acetic acid | 5.80 | 5.90 | 5.20 | 3.80 | 0.70 | 0.333 |
| Day 56    |      |    |     |       |       |         |
| Methyl mercaptans | 6.13 | 5.08 | 5.08 | 5.20 | 1.07 | 0.198 |
| NH₃       | 3.12 | 1.47 | 2.00 | 1.37 | 0.49 | 0.054 |
| H₂S       | 7.00 | 7.40 | 6.50 | 6.20 | 0.80 | 0.806 |
| CO₂       | 1675 | 1575| 1625 | 1550 | 248  | 0.048 |
| Acetic acid | 6.00 | 5.40 | 5.20 | 5.00 | 0.80 | 0.890 |

**Abbreviations:** Ctrl: basal diet; QS: quillaja saponin; FOS: fructooligosaccharide; NH₃: ammonia; H₂S: hydrogen sulphide; CO₂: carbon dioxide; SEM: standard error of the mean.

^a,bMeans in the same row with different superscript differ significantly (p < 0.05).
and pigs' performance. *Escherichia coli* as an example of a harmful bacteria can increase gut thickness, accelerate gut tissue turnover rate, and/or compete with the host for the feed nutrients and hence decrease feed efficiency. In contrast, *Lactic acid bacteria* as an example of the beneficial bacteria can inhibit pathogenic bacteria via competitive exclusion, bacteriocin/acid production, and stimulation of the immune system and therefore improve pigs' growth and health (Del Hierro et al. 2018; Lei et al. 2018).

In the current study, pigs fed QS diets showed QS improved faecal lactic acid bacteria counts and decreased faecal *E. coli* counts compared to those fed on the control diet. Similar findings were reported by other researchers (Dang and Kim 2021; Ilsley et al. 2005). Dietary inclusion of QS could improve *Lactobacillus* counts in pigs. On the other hand, Vaclavkova and Beckova (2008) reported that dietary inclusion of QS had increased faecal *lactic acid bacteria* counts and decreased faecal *E. coli* counts in pigs.

**Dietary supplementation of the FOS increases faecal *Lactobacillus* counts and lowers faecal *E. coli* counts.** Similarly, Zhao et al. (2013) reported significantly increased faecal *Lactobacillus* counts and lowered faecal *Escherichia coli* counts of pigs fed dietary 2% FOS supplementation. Additionally, Xu et al. (2003) found a significant increase in *Bifidobacteria* and *Lactobacillus* pigs fed dietary 6 g/kg FOS supplementation. Furthermore, Li and Kim (2013) reported a significant improve in faecal *Lactobacillus* counts due to dietary inclusion of FOS at levels 0.5, 1.0 and 2.0 mg/kg. In the present study, QSFOS dietary treatment had enhanced faecal *Lactobacillus* counts whereas decreased faecal *E. coli* counts compared with Ctrl, QS and FOS dietary treatments, indicating that a combination of FOS and QS was more effective in increasing faecal *Lactobacillus* counts and lowering faecal *E. coli* counts than QS and FOS alone. On the other end, Mountzouris et al. (2006) reported that dietary inclusion of 10 g/kg did not significantly influence the faecal *bacterial* count. Similarly, Mikkelsen and Jensen (2004) reported that supplementation of FOS did not affect the faecal bacterial count in pigs. This differential response could be due to the product composition, the level of ingestion and the hygiene status of the farm.

A serious environmental problem in the pig industry is the production of harmful gases. Ammonia, total mercaptans and hydrogen sulphide are the main air pollutants in swine production. The results of the current study showed that dietary supplementation with QS and FOS decreased faecal NH₃ and CO₂ emissions in growing pigs. Previous studies have reported that dietary supplementation with QS has positive effects on NH₃ in pigs (Philippe et al. 2007; Veit et al. 2011; Dang and Kim 2020). Additionally, Bartoš et al. (2016) reported a significant decrease faecal NH₃ emission of growing pigs fed dietary 150 mg/kg QS supplementation. Some studies have reported that prebiotics can lower faecal gas emissions in pigs (Cho and Kim 2015; Deng et al. 2015). In the present study, QSFOS dietary treatment had reduced ammonia emissions compared with a Ctrl diet, indicating that a combination of FOS and QS was more effective in reducing ammonia emissions than QS and FOS alone. FOS, is a kind of prebiotic, because of the beta-linkages between fructose monomers, it cannot be decomposed by digestive enzymes; however, it can selectively stimulate the growth and activity of beneficial microorganisms (Saulnier et al. 2007). Xu et al. (2003) and Dang and Kim (2020) indicated that supplementation of FOS and QS increased the *Lactobacillus* counts and decreased faecal *coliiform bacteria* counts and nutrient digestibility by reducing faecal noxious gas emission. Additionally, it is suggested that in a lower gastric pH gut environment, fermentation bacteria of carbohydrates may incorporate more undigested protein and their metabolites, peptides, amino acids, microbial protein and thereby reducing the potential for ammonia emission (Lei et al. 2017). Therefore, the possible reasons for reduced ammonia emissions may be possibly due to the increased nutrient digestibility and increase in faecal *Lactobacillus* counts, reduction in faecal *coliiform bacteria* counts and lowered gastric pH.

The CO₂ emissions measured in the present study averaged 3.6 kg per animal per day and were higher as reported in the study of Bartoš et al. (2016). As anaerobic fermentation or aerobic degradation of organic matter in the manure is one source of CO₂ (Philippe and Nicks 2015), it might be speculated that the content of fermentable/degradable organic matter in the manure was higher in the present study compared to the study of Bartoš et al. (2016). There was a significantly lower CO₂ emission in QS and FOS or QSFOs compared with a Ctrl. Elevated concentration of CO₂ reduces the frequency and increases the depth of respiration in animals, while the long-lasting and excessive concentration of CO₂ leads to metabolic disorders or even acidosis (Bartoš et al. 2016). Between the tested QS and FOS and QSFOs, there were small differences in terms of NH₃ reduction. The NH₃ reducing effect of QS, FOS and QSFOs is discussed to be either due to NH₃ binding characteristics of saponins and prebiotics or because of inhibitory effects of
saponins and prebiotics on microorganism urease enzymes (Bartos et al. 2016; Lei et al. 2017). Both possibly modes of action are in agreement with the observation that the QS and prebiotics containing feed additives tested in this study exclusively affect NH₃ and no other gas emissions. The slight reduction feed additives tested in this study exclusively affect

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Writing
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Conclusion

Supplementation of QS, FOS alone and the combination of QS and FOS diet improved growth performance, nutrient digestibility and Lactobacillus counts. Compared with supplementation of QS and FOS alone, the combination of QS and FOS on reducing E. coli counts and NH₃ and CO₂ emissions.

Ethical approval

All animal procedures used in this experiment were reviewed and approved by the Institutional Animal Care and Use Committee of Dankook University (Cheonan, Republic of Korea). (DK-1-1934).

Disclosure statement

No potential conflict of interest was reported by the author(s).

Data availability statement

The data presented in this study are available on request from the corresponding author.

Author contributions

Madesh Muniyappan: Investigation, Data curation, Formal analysis, writing – original draft preparation, Conceptualisation, Methodology, Software, Writing – review & editing. In Ho Kim: Conceptualisation, Methodology, Writing – review & editing, Supervision.

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