



## Analysis of the effect of dietary protected organic acid blend on lactating sows and their piglets

Subramaniam Mohana Devi<sup>1</sup>, Kwang Yong Lee<sup>1</sup>, In Ho Kim<sup>1</sup>

<sup>1</sup> Dankook University, Department of Animal Resource & Science, Cheonan, Chungnam, South Korea.

**ABSTRACT** - The objective of the present study was to evaluate the effects of blends of dietary protected organic acid supplementation on growth performance, nutrient digestibility, blood profiles, faecal microflora, and gas emission on sows and piglets with emphasis on their modes of action to improve pig performance. A total of 12 sows with an average initial body weight (BW) of 252.40±11.7 kg were used in this trial. Growth performance, blood profiles, and nutrient digestibility of sows and piglets fed protected organic acid were evaluated. The dietary treatments included a basal diet (CON); CON + 0.1% protected organic acid; and CON + 0.2% protected organic acid. The BW and back fat of sows was checked four days prior to farrowing and at the weaning day to calculate BW loss and back fat loss during that period. Inclusion of 0.2% protected organic acid provided a greater digestibility than CON diets throughout the experimental period in lactating sows. Dietary supplementation with 0.2% protected organic acid led to a higher white blood cell and lymphocyte concentration than CON treatment in sucking piglets. Immunoglobulin G concentration observed was greater in protected organic acid groups in lactating sow and sucking piglets. Increased faecal *Lactobacillus* counts with decreased *E. coli* concentrations were observed with the diets of protected organic acid fed to lactating sows. The *E. coli* counts were decreased in weaning piglets. The faecal H<sub>2</sub>S contents were decreased in 0.2% protected organic acid diets during farrowing on day 1. Dietary supplementation with protected organic acid blends beneficially affects the nutrient digestibility, ileal noxious gas (NH<sub>3</sub> and H<sub>2</sub>S) emission, as well as intestinal microbial balance in lactating sows.

Key Words: blood profiles, digestibility, growth performance, noxious gas, pigs

### Introduction

Supplementation of pig diets with organic acids has become an important nutritional strategy aimed at improvement, performance, and health status of animals fed diets devoid of antibacterial growth promoters. Positive effects of acids are associated mainly with increased gastric acidity, antibacterial activity, reduced coliform populations, and improved digestibility (Jensen et al., 2003).

Studies prior to 2006 indicate that health strategies widely used antibiotic growth promoters to reduce enteric infections and the occurrence of pathogens able to adhere to intestinal mucosa (Budino et al., 2005). Among a variety of candidates for the replacement of antibiotics, organic acids have been broadly applied worldwide with reasonable success (Mroz, 2005). The successful use of organic acids in the diets of pigs requires understanding of their mechanisms of action. Organic acids may influence the physiology of the

intestinal mucosa by their action on the villi, maintaining their integrity, promoting an increase in the number of cells and preventing its flattening, as well as serving as a substrate in the intermediary metabolism of the citric acid-cycle (Partanen and Mroz, 1999). Organic acids can also reduce the diets' buffering capacity, inhibit the proliferation and colonisation of undesirable microorganisms, act on the physiology of the gastrointestinal mucosa, and promote the availability of nutrients in the diet, improving their digestion, absorption, and retention (Costa et al., 2011). In some countries, organic acids are used to decontaminate microorganisms in feed ingredients (Koyuncu et al., 2013). According to Overland et al. (2008), dietary inclusions of organic acids have positive effects in improving growth rate and feed efficiency. A combination of organic acids would enhance the effectiveness of acidification and maintains optimum pH (Ravindran and Kornegay, 1993). Our previous study suggested that inclusion of bacteriophages could be used as an antibiotics alternative for growing pigs (Yan et al., 2012). However, the effectiveness varies with the types and combinations of acid, the health status of the animal, and feed characteristics (Mroz et al., 2006). The protected organic acid used in the current study is a blend of organic acids and medium-chain fatty acids (MCFAs) with matrix coating.

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Corresponding author: inhokim@dankook.ac.kr

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Protected organic acids and MCFA are released in the small intestine by lipase and those undissociated organic acids released inhibit the growth of hazardous bacteria (*Salmonella* and *E. coli*, etc.) and increase the population of desired bacteria (lactic acid bacteria). The protective lipid matrix used for microencapsulation of the organic acid blend allowed slow-release of the active ingredients, preventing the immediate disappearance of such compounds upon exiting the stomach. The longer permanence along the gastrointestinal tract of active compounds allowed them to act synergistically on the intestinal microflora and to reduce coliform counts.

In the present study, the protected organic acid consists of MCFA for animal nutrition and metabolism, especially made by technology of Joint Matrix coating. Therefore, we hypothesised that the blends of different organic acids with MCFA in matrix coating could play an influential role in improving growth performance, microbial population, nutrient digestibility, blood profiles and gas emission of lactating sows and their piglets. The objective of the present study was to evaluate the effects of blends of dietary protected organic acid supplementation on growth performance, nutrient digestibility, blood profiles, faecal microflora, and gas emission in sows and piglets with emphasis on their modes of action to improve pig performance.

## Material and Methods

The study was carried out following the procedures approved by the Animal Care and Use Committee of Dankook University, in Cheonan, Chungnam, South Korea, for experiments with animals.

The matrix coated organic acid used in the current experiment was provided by a commercial company (Morningbio Co., Ltd., Cheonan, Korea). This protected organic acid consists of MCFA and composite organic acids. The active ingredients were 17% fumaric acid, 13% citric acid, 10% malic acid, 1.2% MCFA (capric and caprylic acids), and a carrier.

A total of 12 crossed (Landrace × Yorkshire) sows and 120 piglets from Dankook University experimental farm were used in this current study to investigate the effect of protected organic acid on the production performance of sows and piglets. Pigs were randomly allocated into one of three treatments (four replications) in a randomized complete block design. The pigs were cross-fostered after farrowing to ten piglets per sow. Dietary treatments included: a basal diet (CON); CON + 0.1% protected organic acid; and CON + 0.2% protected organic acid. Sows were

fed a same commercial gestation diet from 28 to 95 days of lactation. After 95 days, the gestating sows were fed the experimental diet. The farrowing parity of experimental sows consisted of four sows for each treatment. The diets (Table 1) were formulated to meet or exceed the nutrient requirements recommended by NRC (2012). The farrowing crate (2.1 × 0.6 m) contained an air-conditioned area for newborn pigs on each side, and the temperature in the farrowing house was maintained at a minimum of 20 °C with supplemental heat provided by heat lamps. Piglets were treated according to routine management practices that included teeth clipping, tail docking, and ear tagging for labelling, and received injections of 1 mL of iron dextran and males were castrated five days after birth. After farrowing, sows received 1 kg of standard lactation diet and increased feed by 1 kg each day *ad libitum* to avoid overconsumption. All diets were provided in meal form, and sows were provided with free access to drinking water throughout the experimental period.

Sows were weighed at the start of the experiment, before farrowing, after farrowing, and at the weaning period. The initial treatment was the experimental diet. The back fat thickness and body weight of the sows were measured within a few hours after the start of the experiment, at farrowing, and at weaning (21 days). The daily feed intake of the sows

Table 1 - Composition of experimental diets (as-fed basis)

Item	Lactation diet
Ingredient, g/kg	
Ground maize	510.0
Soybean meal (480 g/kg CP)	267.3
Wheat bran	10.0
Rice bran	50.0
Rapeseed meal (430 g/kg CP)	35.0
Tallow	60.5
Molasses	35.0
Dicalcium phosphate	16.4
Limestone	7.6
NaCl	5.0
L-lysine-HCl (780 g/kg)	1.2
Vitamin premix <sup>1</sup>	1.0
Trace mineral premix <sup>2</sup>	1.0
Analysed nutrient content, g/kg	
Dry matter	888.7
Metabolisable energy, MJ/kg	14.47
Crude protein	183.4
Crude fat	91.6
Lysine	10.8
Calcium	10.6
Total phosphorus	7.3
Iron, mg/kg	25.0

<sup>1</sup> Provided per kilogram of complete diet: vitamin A - 12,100 IU; vitamin D3 - 2,000 IU; vitamin E - 48 IU; vitamin K3 - 1.5 mg; riboflavin - 6 mg; niacin - 40 mg; D-pantothenic acid - 17 mg; biotin - 0.2 mg; folic acid - 2 mg; choline - 166 mg; vitamin B6 - 2 mg; vitamin B12 - 28 µg.

<sup>2</sup> Provided per kilogram of complete diet: Cu (as CuSO<sub>4</sub>·5H<sub>2</sub>O) - 15 mg; Zn (as ZnSO<sub>4</sub>) - 50 mg; Mn (as MnO<sub>2</sub>) - 54 mg; I (as KI) - 0.99 mg; Se (as Na<sub>2</sub>SeO<sub>3</sub>·5H<sub>2</sub>O) - 0.25 mg.

was determined as the difference between feed allowance and the refusals collected after feeding. After farrowing, piglets were ear-notched and weighed individually. Individual piglet body weight (BW) was assessed on days 0, 7, 14, and 21 (weaning), whereas the number of piglets of every sow was recorded on farrowing day and weaning day to evaluate the survival rate of piglets. The back fat thickness (6 cm of the midline at the 10th rib) measurements were taken using a real-time ultrasound instrument (Piglot 105, SFK Technology, Herlec, Denmark).

Chromium oxide was added to all the diets at 2 g/kg as an indigestible marker for determination of apparent total tract digestibility of dry matter (DM), nitrogen (N), and gross energy (Ball and Aherne, 1987). On days 26, 27, and 28, faecal samples were collected from all pigs in each pen in the afternoon via rectal massage and pooled within pen. Faecal samples (four samples per treatment) and feed samples were stored in a freezer at  $-20^{\circ}\text{C}$  until further analysis. For chemical analysis, faecal and feed samples were freeze-dried and ground to pass through a 1-mm screen. Dietary DM, calcium, phosphorus, crude protein, lysine, and methionine were analysed according to the procedures described by AOAC (2000). Chromium was analysed by UV absorption spectrophotometry (Shimadzu, UV-1201, Kyoto, Japan) (Williams et al., 1962), and N was measured using a Kjeltac 2300 analyser (Foss Tecator AB, Hoeganaes, Sweden). Gross energy was determined using a Parr 6100 Oxygen Bomb Calorimeter (Parr Instrument Co., Moline, IL, USA). The apparent total tract digestibility of DM, N and gross energy were calculated using indirect methods as described by Zhao et al. (2012).

The incidence of diarrhoea in piglets was observed and recorded from five days after farrowing to weaning every day. To assess the severity of diarrhoea, faeces from each pig were scored by determining the moisture content according to the method described by Hart and Dobb (1988). In short, scores were 0 for normal, firm faeces; 1 for possible slight diarrhoea; 2 indicated definitely unformed, moderately fluid faeces; or 3 in case of very watery and frothy diarrhoea. A cumulative diarrhoea score per diet and day was further assessed (Montagne et al., 2004).

Blood from sows was collected via vena cava puncture before feeding at farrowing and weaning (21 days) whereas six piglets per treatment were selected randomly at weaning. Blood from the same sows and piglets were collected on day 21. Blood samples were collected into 5-mL vacuum tubes with  $\text{K}_3\text{EDTA}$  (Becton Dickinson Vacutainer Systems, Franklin Lakes, NJ, USA) and placed on ice until analysis. The concentration of white blood cells (WBC) and lymphocyte in the whole blood samples were

determined using an automatic blood analyser (ADVIA 120, Bayer, Tarry town, NY, US). Whole blood samples were subsequently centrifuged for 15 min at  $3000 \times g$  at  $4^{\circ}\text{C}$  and the harvested serum was used to determine blood profiles. Serum immunoglobulin G (IgG) was particularly analysed using nephelometry (Dade Behring, Marburg, Germany).

On farrowing and weaning, faecal samples (for individual sows) were collected by rectal massage from all sows from each pen, pooled, and transported to the lab for immediate analysis. One gram of the composite faecal sample from each pen was diluted with 9 mL of 1% peptone broth (Becton, Dickinson and Co., Franklin Lakes, USA) and homogenised. Viable counts of bacteria in the faecal samples were then conducted by plating serial 10-fold dilutions (in 1% peptone solution) onto MacConkey agar plates (Difco Laboratories, Detroit, MI, USA) and Lactobacilli spp. medium III agar plates (Medium 638, DSMZ, Braunschweig, Germany) to isolate the *Escherichia coli* and *Lactobacillus*, respectively. The Lactobacilli medium III agar plates were then incubated for 48 h at  $39^{\circ}\text{C}$  under anaerobic conditions and the MacConkey agar plates were incubated for 24 h at  $37^{\circ}\text{C}$  and counted.

At the end of farrowing and weaning, faecal  $\text{NH}_3$ , R.SH,  $\text{H}_2\text{S}$ , and acetic acid concentrations were determined using the methods described by Yan and Kim (2013). A total of 300 g fresh faecal samples were collected from at least two pigs in each pen (14 pigs per treatment), then transferred to a sealed box and fermented for 48 h at  $32^{\circ}\text{C}$  in an incubator. At day 5, levels of  $\text{NH}_3$ , R.SH,  $\text{H}_2\text{S}$ , and acetic acid were measured within the range of 5.0-100.0 (No. 3La, detector tube; Gastec Corp.), 2.0-20.0 (4LK, detector tube; Gastec Corp.), 0.5-120.0 (No.70 L and 70, detector tube; Gastec Corp.), and 2.0-50.0 (No.81 L, detector tube; Gastec Corp.) ppm. Prior to the measurements, slurry samples were shaken manually for approximately 30 s to disrupt any crust formation on the surface and homogenised. Adhesive plasters were punctured, and 100 mL of headspace air were sampled, approximately 2.0 cm above the slurry surface. Two samples from each pen were measured and then the average was calculated.

Data were subjected to general linear model (GLM) procedure of SAS (Statistical Analysis System, version 7.0) in a randomised complete block design. Effects of treatments (Control; CON + 0.1% protected organic acid; and CON + 0.2% protected organic acid) were analysed by ANOVA for assessing the design, and linear and quadratic effects of each treatment. Results are presented as least square mean and the variability in data was expressed as standard error of the mean (SEM). Before conducting statistical

analysis of the microbial counts, the value was transformed logarithmically. Linear and quadratic polynomial contrasts were performed to determine the effects of inclusion level of 0.1% and 0.2% protected organic acid blends in the diet. Probability values higher than or equal to 0.05 were considered significant.

## Results

There was no significant difference ( $P>0.05$ ) in before farrowing, after farrowing, and weaning body weight of sows (Table 2), and nor did the sow body weight loss differ in the dietary treatments ( $P>0.05$ ) during the lactating period. No difference ( $P>0.05$ ) was observed in ADFI and back fat thickness loss during the experiments. Piglet survival was not affected by dietary treatments, birth weight (1-week weight, 2-week weight, weaning weight), or ADG (Table 3,  $P>0.05$ ). The number of diarrhoeal piglets and faecal score showed no difference ( $P>0.05$ ) in suckling piglets during the experimental period (Table 4). During the experimental period, digestibility of DM, N, and energy was found to increase linearly by the dietary supplementation of 0.2% protected organic acid diets (Table 5;  $P<0.01$ ).

No difference ( $P>0.05$ ) was observed in WBC, lymphocyte, and IgG in farrowing sows. The WBC and

lymphocyte of 0.2% protected organic acid were increased (linear effect,  $P = 0.003$ ) after sow weaning. Moreover, the IgG concentration of protected organic acid increased (linear effect,  $P<0.001$ ) after sow weaning. Besides, IgG concentration was higher (linear effect,  $P<0.001$ ) in the groups fed protected organic acid diets (Table 6). Supplementation of 0.2% protected organic acid diet to suckling piglets (Table 6) provided greater (linear effect,  $P = 0.01$ ) WBC counts than CON diet.

Table 2 - Effect of dietary protected organic acid blend supplementation on performance in lactating sows<sup>1</sup>

Item	CON	POA1	POA2	SE	Linear <sup>2</sup>	Quadratic <sup>2</sup>
Parity	4	4	4			
Litter						
Number of pigs	11.8	13.0	11.0	1.4	0.8	0.5
Weaned pigs	9.3	11.0	9.8	1.2	0.9	0.5
Body weight, kg						
Before farrowing <sup>3</sup>	267.0	274.2	275.6	10.0	0.6	0.8
After farrowing	245.9	253.5	252.8	10.1	0.6	0.7
Weaning	236.9	245.4	245.5	10.2	0.6	0.7
Body weight loss <sup>4</sup>	9.0	8.0	7.3	0.9	0.3	0.9
ADFI, kg						
1 week	4.14	4.31	4.33	0.19	0.5	0.8
2 week	6.23	5.98	6.35	0.66	0.9	0.7
3 week	7.25	7.51	7.19	0.37	0.9	0.5
Overall	5.85	5.93	5.90	0.32	0.9	0.9
Backfat thickness, mm						
Before farrowing <sup>3</sup>	19.3	19.3	20.9	1.0	0.3	0.5
After farrowing	17.0	18.1	18.3	1.0	0.4	0.7
Weaning	14.8	17.1	17.5	1.4	0.2	0.5
Backfat thickness loss <sup>5</sup>	2.3	1.0	1.6	0.7	0.5	0.2
Oestrus interval, d	4.0	3.5	3.8	0.4	0.6	0.4

ADFI - average daily feed intake; SE - standard error.

<sup>1</sup> CON - basal diet; POA1 = CON + 0.1% POA; POA2 = CON + 0.2% POA.

<sup>2</sup> CON vs. POA1 vs. POA2.

<sup>3</sup> Farrowing before 4 days.

<sup>4</sup> Body weight loss from farrowing to weaning.

<sup>5</sup> Backfat thickness loss from farrowing to weaning.

Table 3 - Effect of dietary protected organic acid blend supplementation on performance in suckling piglets<sup>1</sup>

Item	CON	POA1	POA2	SE	Linear <sup>2</sup>	Quadratic <sup>2</sup>
Piglet survival, %	94.4	96.9	97.2	3.1	0.5	0.8
Body weight, kg						
Birth weight	1.52	1.56	1.69	0.14	0.4	0.8
1-week weight	2.96	2.94	3.04	0.17	0.8	0.8
2-week weight	4.74	4.83	4.89	0.23	0.7	1.0
Weaning weight	6.70	6.86	6.96	0.39	0.6	1.0
Average daily gain, g						
1 week	202	193	190	8	0.4	0.8
2 week	255	270	265	16	0.6	0.6
3 week	280	289	295	30	0.7	1.0
Overall	246	251	250	15	0.8	0.9

SE - standard error.

<sup>1</sup> CON - basal diet; POA1 = CON + 0.1% POA; POA2 = CON + 0.2% POA.

<sup>2</sup> CON vs. POA1 vs. POA2.

Table 4 - Effect of protected organic acid blend supplementation on faecal score in suckling piglets<sup>1</sup>

Item	CON	POA1	POA2	SE	Linear <sup>2</sup>	Quadratic <sup>2</sup>
Farrowing						
Number of piglets	1.5	1.0	0.8	0.3	0.3	0.8
Faecal score <sup>3</sup>	3.4	3.3	3.3	0.2	0.6	0.8
Weaning						
Number of piglets	0.5	0.3	0.0	0.3	0.1	1.0
Faecal score <sup>3</sup>	3.3	3.1	3.0	0.1	0.1	1.0

SE - standard error.

<sup>1</sup> CON - basal diet; POA1 = CON + 0.1% POA; POA2 = CON + 0.2% POA.

<sup>2</sup> CON vs. POA1 vs. POA2.

<sup>3</sup> Faecal score: 1 - hard, dry pellets in a small, hard mass; 2 - hard, formed stool that remains firm and soft; 3 - soft, formed, and moist stool that retains its shape; 4 - soft, unformed stool that assumes the shape of the container; 5 - watery, liquid stool that can be poured.

Table 5 - Effect of dietary protected organic acid blend supplementation on nutrient digestibility in lactating sows<sup>1</sup>

Item, %	CON	POA1	POA2	SE	Linear <sup>2</sup>	Quadratic <sup>2</sup>
Weaning						
Dry matter	65.20b	67.08ab	68.30a	0.70	0.005	0.69
Nitrogen	70.17b	72.05ab	73.56a	0.94	0.02	0.87
Energy	65.39b	66.49ab	69.16a	0.92	0.008	0.50

SE - standard error.

<sup>1</sup> CON - basal diet; POA1 = CON + 0.1% POA; POA2 = CON + 0.2% POA.

<sup>2</sup> CON vs. POA1 vs. POA2.

a,b - means in the same row with different letters differ ( $P<0.05$ ).

Sows fed protected organic acid diets had lower (linear effect,  $P < 0.05$ ) *E. coli* contents in the farrowing and weaning periods compared with sows fed the CON diet (Table 7). In addition, protected organic acid treatments lead to a higher (linear effect,  $P < 0.04$ ) *Lactobacillus* concentration in the farrowing and weaning periods (Table 7). Results for faecal gas emission of sows (Table 8) indicated the protected organic acid treatment provided a lower (linear effect,  $P = 0.01$ )  $H_2S$  than non-protected organic acid treatment at the end of farrowing. Moreover, supplementation of protected organic acid resulted in lower (linear effect,  $P < 0.04$ )  $NH_3$  levels compared with that of CON treatment on days 1 and 5.

## Discussion

Livestock production can make good use of resources, which contributes with high-quality nutrients to the human diet (Mohana Devi et al., 2014a). Organic acids are widely distributed in plants and animals produced by microbial

Table 6 - Effect of dietary protected organic acid blend supplementation on blood profiles in lactating sows and suckling piglets<sup>1</sup>

Item, log <sub>10</sub> cfu/g	CON	POA1	POA2	SE	Linear <sup>2</sup>	Quadratic <sup>2</sup>
<b>Farrowing</b>						
WBC, 10 <sup>3</sup> /μL	10.52	11.01	11.32	0.89	0.54	0.94
Lymphocyte, %	19.05	20.90	19.68	2.75	0.88	0.66
IgG, mg/dL	833	873	866	31	0.47	0.55
<b>Weaning</b>						
WBC, 10 <sup>3</sup> /μL	10.07b	12.07ab	14.60a	0.75	0.003	0.90
Lymphocyte, %	43.1b	44.0ab	46.0a	0.7	0.01	0.54
IgG, mg/dL	815b	1,383a	1,300a	43	<0.0001	0.0002
<b>Suckling</b>						
WBC, 10 <sup>3</sup> /μL	9.75b	11.06ab	11.88a	0.56	0.01	0.72
Lymphocyte, %	48.1	54.4	51.2	2.2	0.34	0.09
IgG, mg/dL	273b	382a	375a	15	<0.0001	0.003

WBC - white blood cells; IgG - immunoglobulin G; SE - standard error.

<sup>1</sup> CON - basal diet; POA1 = CON + 0.1% POA; POA2 = CON + 0.2% POA.

<sup>2</sup> CON vs. POA1 vs. POA2.

a, b - means in the same row with different letters differ ( $P < 0.05$ ).

Table 7 - Effect of dietary protected organic acid blend supplementation on faecal microflora in lactating sows<sup>1</sup>

Item, log <sub>10</sub> cfu/g	CON	POA1	POA2	SE	Linear <sup>2</sup>	Quadratic <sup>2</sup>
<b><i>E. coli</i></b>						
Farrowing	5.67a	4.93b	5.18b	0.10	0.008	0.001
Weaning	5.99a	5.23b	5.33b	0.10	0.07	0.60
<b><i>Lactobacillus</i></b>						
Farrowing	6.13b	7.49a	7.34a	0.07	0.02	0.92
Weaning	7.32b	7.75a	7.81a	0.06	0.001	0.14

SE - standard error.

<sup>1</sup> CON - basal diet; POA1 = CON + 0.1% POA; POA2 = CON + 0.2% POA.

<sup>2</sup> CON vs. POA1 vs. POA2.

a, b - means in the same row with different letters differ ( $P < 0.05$ ).

fermentation of carbohydrates and other fermentable material, predominantly in the large intestine of pigs. Acidifiers have received much attention in pig production due to their beneficial effects on growth performance of pigs (Papatsiros et al., 2011). In the case of early weaning, a sudden dietary change leads to digestive disturbances (Wu et al., 2012). The addition of organic acids to pig diets has been examined for decades, clearly demonstrating to improve growth performance and apparent total tract digestibility (Suryanarayana et al., 2012). Walsh et al. (2007)

Table 8 - Effect of dietary protected organic acid blend supplementation on faecal noxious gas emission in lactating sows<sup>1</sup>

Item, %	CON	POA1	POA2	SE	Linear <sup>2</sup>	Quadratic <sup>2</sup>
<b>Farrowing</b>						
<b>Ammonia</b>						
1 d	3.8	3.8	4.8	0.6	0.3	0.5
3 d	4.3	3.8	3.3	0.9	0.4	1.0
5 d	2.8	3.8	3.0	0.5	0.7	0.2
7 d	0.5	1.0	1.5	0.6	0.2	1.0
<b>Total mercaptans</b>						
1 d	0.8	0.6	0.5	0.2	0.5	1.0
3 d	0.8	1.3	0.8	0.3	1.0	0.2
5 d	0.5	0.6	0.6	0.3	0.7	0.6
7 d	0.4	0.3	0.4	0.2	1.0	0.7
<b>Hydrogen sulphide</b>						
1 d	6.3a	4.8b	3.3c	0.6	0.01	1.0
3 d	9.5	7.8	8.3	1.4	0.5	0.5
5 d	4.8	4.8	6.5	1.5	0.4	0.6
7 d	5.3	4.0	4.0	1.0	0.4	0.6
<b>Acetic acid</b>						
1 d	3.8	2.0	3.8	0.9	1.0	0.1
3 d	5.0	4.3	5.0	1.0	1.0	0.6
5 d	2.8	3.3	3.3	0.7	0.7	0.8
7 d	1.3	1.0	1.5	0.3	0.8	0.7
<b>Weaning</b>						
<b>Ammonia</b>						
1 d	9.3a	5.8b	6.0b	0.8	0.05	0.2
3 d	9.5	7.5	8.5	1.1	0.5	0.3
5 d	5.8a	3.5b	3.3b	0.4	0.6	0.04
7 d	3.0	2.0	3.0	0.7	1.0	0.2
<b>Total mercaptans</b>						
1 d	0.4	0.6	0.4	0.2	1.0	0.4
3 d	1.6	1.0	0.8	0.5	0.2	0.8
5 d	1.1	0.9	0.8	0.6	0.6	0.9
7 d	0.8	0.5	0.8	0.4	1.0	0.6
<b>Hydrogen sulphide</b>						
1 d	18.8	15.8	16.8	5.4	0.8	0.7
3 d	17.0	14.0	12.5	2.7	0.3	0.8
5 d	6.5	7.0	7.8	1.6	0.7	1.0
7 d	1.0	1.0	3.0	0.8	0.1	0.4
<b>Acetic acid</b>						
1 d	3.5	2.5	3.0	1.1	0.8	0.6
3 d	2.8	1.8	2.8	0.7	1.0	0.4
5 d	1.0	1.0	0.8	0.4	0.7	0.8
7 d	1.0	0.5	0.8	0.3	0.6	0.4

SE - standard error.

<sup>1</sup> CON - basal diet; POA1 = CON + 0.1% POA; POA2 = CON + 0.2% POA.

<sup>2</sup> CON vs. POA1 vs. POA2.

a,b,c - means in the same row with different letters differ ( $P < 0.05$ ).

demonstrated that addition of 0.4% organic acid blend (fumaric, lactic, propionic, citric, and benzoic acids) to the diets of nursery pigs resulted in better growth rate, feed intake, and feed efficiency compared with the control diet. In the present study, there was no significant difference in the pre- and post-farrowing body weight and weaning body weight of sows. There was no difference in body weight loss among the dietary treatments during the lactating period of sows and nor was there any difference in the ADFI and backfat thickness loss during the experiments. Piglet survival, birth weight, and ADG were not affected by the dietary treatments. These results are in agreement with a study conducted by Canibe et al. (2005), who reported that ADG and ADFI were not affected, but the growth:feed ratio tends to be greater for growing pigs fed the diet containing 1.8% formic acid. Feed additives such as enzymes, essential oils, and benzoic acid and their combination can improve growth performance in broiler chickens (Giannenas et al., 2014). Mohana Devi et al. (2014b) reported that supplementation of protein sources in growing pig diets improved the growth rate and feed intake. Dietary supplementation with medium-chain fatty acids together with probiotic in weaning pigs can improve growth performance, increase nutrient digestibility, and enhance biochemical profiles (Mohana Devi and Kim, 2014).

Mroz et al. (2000) suggested that organic acids had a beneficial effect on the apparent ileal and total tract digestibilities as well as calcium digestibility in growing pigs. This may be because of higher diet acidity due to the addition of organic acid to the diet. In agreement with this study, Partanen et al. (2007) demonstrated that dietary organic acid improved the apparent ileal digestibility in 34-kg pigs. In addition, the inclusion of soy bean in weaned piglet diets increased the microbial biodiversity in distal intestinal segments (Roca et al., 2014). Partanen et al. (2001) documented that formic acid tends to decrease bacterial nitrogen in different parts of the small intestine in pigs and improve apparent ileal digestibility of protein sources, certain essential amino-acids, lipids, calcium, and phosphorus.

In the present study, digestibility of DM, N, and energy were increased (linear effect,  $P < 0.01$ ) by the dietary supplementation of 0.2% protected organic acid diets. The growth efficiency of market pigs was significantly increased by the addition of formic acid and this effect was further enhanced by the combination of formic acid with potassium sorbate (Partanen et al., 2002). Research implies an improved apparent ileal digestibility of protein and amino acids (Mroz et al., 1997) and an improved absorption of minerals (Jongbloed and Jongbloed, 1996).

Likewise, other previous reports with different types of organic acid indicated that the inclusion of organic acids such as 2% benzoic acid (Kluge et al., 2010) in the diet of lactating sows improved the digestibility of nutrients, and 0.5% phenyl lactic acid (Wang et al., 2009) improved nutrient digestibility in weaning pig and trends of nitrogen digestibility in growing pigs. Franco et al. (2005) also reported that combination of lactic acid with formic or fumaric acids numerically increased DM digestibility in weaning pigs. The increase in nutrient digestibility might have resulted from the increased microbial activity in the gastrointestinal tract (Yin et al., 2001). However, Kil et al. (2006) indicated that there was no positive response from nutrient digestibility with the inclusion of lactic, formic, or fumaric acids in weaning pigs.

As acidifiers, diets with organic acid improved growth performance (Partanen and Mroz, 1999), nutrient digestibility, gut health (Partanen et al., 2007; Wang et al., 2009), increased blood lymphocyte and WBC concentration, and decreased digesta pH value in the gastrointestinal tract (Ravindran and Kornegay, 1993) in pigs mainly due to its antibacterial and antifungal activities (Lavermicocca et al., 2003). In the present investigation, the WBC and lymphocyte of 0.2% protected organic acid were increased after sow weaning. Moreover, the IgG concentration of protected organic acid increased in sows after weaning. The supplementation of 0.2% protected organic acid in the diet of sucking piglets provided greater WBC counts and IgG concentration than the CON diet. Lymphocytes were more densely populated in the lamina propria and submucosa of caecal tonsil and ileum in diets supplemented with citric acid (0.5%) (Chowdhury et al., 2009). In addition, a reduction in subclinical infections due to antimicrobial effects may contribute to improved nutrient digestibility and a reduction in the demand for nutrients by the gut-associated immune tissue.

It has been reported that addition of organic acids to pig diets alters the total microbial load, which can be particularly effective against *E. coli* and other acid-intolerant organisms (Dibner and Buttin, 2002). The presence of *Lactobacillus* in the gastrointestinal tract is believed to be beneficial for the pig. In the current study, the number of *lactobacilli* increased because of the inclusion of protected organic acid. Sows fed protected organic acid diets had decreased *E. coli* contents during farrowing and weaning period with a higher *Lactobacillus* concentration. Protected organic acid led to lower *E. coli* counts in the ileum and higher *Lactobacillus* counts in the colon, indicating that protected organic acid is more effective in retarding absorption of dietary acids and allowing more effective delivery of acids

to the distal ileum, caecum, and colon of piglets (Bosi et al., 1999). Thus, including MCFA with organic acid blends would enhance its anti-microbial effects. Therefore, we hypothesised that the blends of different organic acids with MCFA in matrix coating could play an influential role in improving growth performance, meat quality, and microbial population of finishing pigs. However, *E. coli* population was significantly reduced in the faeces obtained from pigs fed diets supplemented with protected organic acid, which is in agreement with Li et al. (2008). In addition, Ahmed et al. (2014) stated that blends of organic acid supplementation reduce *E. coli* count and increase *Lactobacilli* population in weaned pigs.

Biagi et al. (2006) described a favourable effect of gluconic acid on intestinal microflora, morphology of intestinal villi, and general condition of piglets. Several studies documenting an improvement in growth efficiency have been reported in the literature (Missotten et al., 2009). The probable mode of actions of organic acids includes reducing the pH value of digesta in the gastrointestinal tract (Ravindran and Kornegay, 1993), regulating the balance of microbial populations in the gut, stimulating the secretion of digestive enzyme (Thaela et al., 1998), and promoting the growth and recovery of the intestinal morphology (Galfi and Bokori, 1990). According to Willamil et al. (2011), microencapsulated acids significantly modify caecal fermentation. Galfi and Bokori (1990) reported that sodium-butyrate supplementation in piglets reduced coliform bacteria and increased the number of *Lactobacillus* spp. in the ileum. In poultry, this supplementation also reduced coliform bacteria such as *E. coli* and *Salmonella* spp. (Van Immerseel et al., 2006). Our findings corroborate the above results by showing a significant increase in amounts of *Lactobacillus* with diets supplemented with coated protected organic acid. Besides, supplementation of 0.2% coated protected organic acid reduced the *E. coli* population in the excreta.

During recent years, different dietary strategies have been successfully assessed to reduce the gas emissions in piglet production. Cortus (2006) observed that urinary pH has a major impact on the volatilisation potential of  $\text{NH}_3$ . As pH is reduced, the  $\text{NH}_4^+$  concentration increases and  $\text{NH}_3$  concentration decreases within a solution. The pH of urine can be lowered by adding acidifying components to the diet, e.g., benzoic acid. These effects have been proven especially for pigs (Aarnink and Verstegen, 2007). Mean gas emissions of different dietary treatments are presented. The experimental diets with protected organic acid evaluated decreased  $\text{NH}_3$ , Mercaptan, and hydrogen sulphide emission in the trial with respect to the control.

The most effective treatment was seen in the treatments with 0.1% and 0.2% protected organic acid compared with control diet. Effect of dietary protected organic acid on gas emission in weaning pigs shows some little difference but there was no significant difference observed among the treatments in weaning pigs. The faecal gas emission of sows was presented and the protected organic acid treatment had a lower  $\text{H}_2\text{S}$  than control treatment at the beginning of farrowing. Moreover, supplementation of protected organic acid treatment shows decreased (linear effect,  $P < 0.04$ )  $\text{NH}_3$  compared with that of CON treatment on days 1 and 5 in the weaning period. Faecal noxious gas emission in lactating sows did not display any significant difference between each treatment. It is possible that, with a higher proportion of protected organic acid blend,  $\text{NH}_3$ , mercaptan, and hydrogen sulphide emission reduction would have been significant. According to Eriksen et al. (2010), 2% benzoic acid supplementation in the diet of pigs reduced ammonia emissions by 60-70%. The noxious bacterial metabolites and ammonia were reduced by acidifiers with the reduction in pathogen concentration (Dibner and Buttin, 2002). In our recent study, harmful gas emissions (RSH and  $\text{H}_2\text{S}$ ) were influenced by the supplementation of dietary fibre and the addition of 5 g/kg benzoic acid in the diet of growing pigs (Mohana Devi et al., 2015).

## Conclusions

Dietary protected organic acids can actually become the most common and efficacious to improve health status and performance of pigs. Acidifying the diet with protected organic acid blend reduces the pathogenic bacterial load. It also improves nutrient digestibility, reduces fecal emission of ammonia and hydrogen sulphide, and increases beneficial bacterial counts. Best results are observed with inclusion of 0.02% organic acid blend. Thus, the use of organic acids in pig production could be a part of a general nutritional strategy focusing on better gastrointestinal health and productivity.

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