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Sows fed with synergistic blend of short- and medium chain organic acid has a carryover effect on postweaning growth rate

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Abstract

This study investigated the effect of a synergistic blend of free and buffered organic acid (FMP) on the performance of piglets born to sows supplemented with a blend of short- and medium-chain organic acids (SGG) during the late gestation and lactation period. A total of 150 multiparous sows (n = 50/treatment, Landrace × Yorkshire) were blocked (2.4 parity) and assigned to 1 of 3 dietary treatments: CON - corn-soybean meal-based basal diet, SGG-Low -CON+ 1.5 kg/ton SGG, and SGG-High - CON + 3kg/ton SGG. During weaning, 600 piglets (6.72 ± 0.5kg) which weaned from sows supplemented with 3 levels of SGG were allocated to 2 weaner diets (Control and FMP - 3kg/ton) following 3 × 2 factorial arrangement. Supplemental effects on performance were measured at d0-d21 and d 21-42, and the entire period. Pigs fed with FMP and born to sows supplemented with SGG-High gained more weight and ate more (p < 0.05) compared with those in the CON group in both phases, and with SGG-Low in the second phase. Over the entire post-weaning period, piglets born to sows supplemented with SGG-Low and SGG-High had a higher average daily gain (ADG) and body weight (BW) (p < 0.05). Regardless of sow treatment, pigs fed with an FMP diet had higher ADG (p < 0.001), BW (p = 0.045), and a lower feed conversion ratio (p = 0.033). Also, feeding FMP diets reduced the fecal Escherichia coli and Clostridium perfringens counts at d42. The current study indicates that sows fed SGG supplement had a positive carry-over effect on the post-weaning growth rate, and FMP supplement enhances the growth performance and reduced the number of C. perfringens and E. coli. Thus, the application of 3 kg/ton of SGG in sows' diet and subsequent feeding of piglets with FMP would be an effective strategy to improve growth rate and reduce pathogenic bacteria in post-weaned piglets.

Keywords: Growth performance, Fecal microbiome, Organic acid, Weaning pig

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Competing interests

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

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Availability of data and material

Upon reasonable request, the datasets of this study can be available from the corresponding author.

Authors' contributions

Conceptualization: Sampath V, Park JH, Pineda L, Kim IH. Data curation: Sampath V, Han Y. Formal analysis: Sampath V. Writing - review & editing: Sampath V, Park JH, Pineda L, Han Y, Cho S, Kim IH.

Ethics approval and consent to participate

The research protocol license (No: DK-2-2005) was approved by Dankook University (Korea) ethical committee, including the Animal Care and Use, prior to the trail.

INTRODUCTION

A modernistic challenge in livestock industry is to exploit the use of certain additives that could boost the livestock production [1]. Over the past centuries, antibiotic growth promoters (AGP) are widely accepted as a potential alternative [2]. However, overuse on specific antibiotics in animals' feedstuff generate bacterial resistance and cause severe health issues to the consumers. As a consequence, the European Union (EU) and other countries including, Korea have prohibited the use of certain antibiotics in animal feedstuff since 2011 [3]. Farmers, researchers, and feed industries have concerns on both animal and consumers health and thus prompted them to find suitable alternatives strategies to replace AGP. Among different strategies, the products based on organic acids have been addressed several times with a promising result in controlling gut disease and enhancing the growth performance of pigs and poultry [4,5]. For instance, Upadhaya et al. [6] reported that inclusion of protected organic acid in the diet of growing pig enhanced the growth performance by improving the microbial inhabitants. Similarly, Devi et al. [7] noted that protected organic acid blend supplement improved the performance of sows and their litters. Besides, medium-chain fatty acids (MCFA) have been shown as a good alternative for antibiotics in piglets, due to its antibacterial activity. In 2013, Zentek et al. [8] addressed that combination of organic acid and MCFA enhanced the intestinal microbial-ecology of piglets.

Acidifiers, especially formic acid, and its salt have been permitted to use in animal feeding. Even EU has approved to use 12 g/kg formic acid (as a flavoring agent) in the diet of pigs [9]. The addition of formic acid showed a positive result in weaning, growing and finishing pigs [10-12]. Previously, Øverland et al. [13] stated that formic acid had a stronger antibacterial effect on coliform bacteria in the small intestine compared to benzoic acid. Bolduan et al. [14] and Maribo et al. [15] demonstrated that increasing level of formic acid from 0.35% to 1.4% (combined with lactic acid) in the diet of pigs reduced the gastric pH. Similarly, Mroz et al. [16] noted the impact of formic acid supplementation in gestation and lactation sows' and reported that the inclusion of 1% formic acid has no effect on the body mass of sows from pregnancy to lactation, whereas it significantly increased the feed intake (FI) during lactation, and slightly improved litter size and piglet birth weight. Though previous studies showed the beneficial effects of dietary organic acids and formic acid in pigs. To the best of our knowledge, this is the first report to find, sows fed diet supplemented with blend of short- and medium chain organic acids (SGG) during late gestation and lactation period has a carry-over effect on the post-weaning growth rate. We hypothesized that inclusion of 1.5 kg/ton and/or 3 kg/ton SSG supplement to sow diet during gestation and lactation period and subsequent feeding with a synergistic blend of free and buffered organic acid (FMP) could enhance the growth performance of piglets. Thus, the purpose this study is to examine whether sows fed with SGG has a carryover effect on post-weaning growth rate.

MATERIALS AND METHODS

Animal ethics

The research protocol (License No: DK-2-2005) was approved by Dankook University (Korea) ethical committee, including the Animal Care and Use, prior to the trail.

Source of additives

SGG is a free-flowing powder based on a blend of a synergistic blend of short chain organic acids (formic acid, acetic acid, lactic acid, propionic acid, citric acid, and sorbic acid) combined with MCFA (C8-caprylic acid, C10- caproic acid, and C12- lauric acid). FMP is a synergistic blend

of free and buffered organic acid (ammonium formate). SGG and FMP supplements used in this study was commercially procured from Trouw Nutrition (Amersfoort, Netherlands).

Trial design, diet formulation and feeding schedule

A total of one-hundred and fifty multiparous sows (n = 50/treatment, Landrace × Yorkshire) (average parity, 2.4) were assigned to 1 of 3 dietary treatments: CON - corn-soybean meal-based basal diet, SGG-Low – CON+ 1.5 kg/ton SGG, and SGG-High – CON + 3 kg/ton SGG during gestation and lactation. The basal diets (gestation and lactation) were formulated to be isocaloric and isonitrogenous according to the NRC [17] nutrient recommendations of sows (Tables 1 and 2). At 107th day sows were moved to farrowing crates and fed with gestation diet. After parturition sows were fed with 2.5 kg/day lactation diet, the quantity of diet offerings was gradually increased to ad libitum access during day 5 of lactation. The progenies were weighed immediately after farrowing,

Items	Gestation
Ingredient, %	
Corn	42.52
Wheat	22.00
Wheat bran	10.00
Soybean hull	6.20
Parm kernell meal	2.00
Soybean meal	3.00
Dehulled soybean meal	5.94
Soybean oil	1.52
Molasses	3.00
Limestone	1.18
MCP	0.74
Salt	0.50
Methionine (98%)	0.02
Threonine (98%)	0.09
Lysine (25%)	0.59
Choline chloride (50%)	0.15
Vitamin/Mineral mixture ¹⁾	0.55
Total	100.00
Calculated value	
DE (kcal/kg)	3,260
ME (kcal/kg)	3,020
C. Protein (%)	13.00
C. Fat (%)	3.80
C. Ash (%)	4.90
C. Fiber (%)	5.80
Lysine (%)	0.68
Calcium (%)	0.75
Phosphorus (%)	0.50

Table 1. Diet composition of gestation sow (as fed basis)

¹⁾Provided per kg of complete diet: 16,800IU vitamin A; 2,400IU vitamin D₃; 108mg vitamin E; 7.2mg vitamin K; 18mg Riboflavin; 80.4mg Niacin; 2.64mg Thiamine; 45.6mg D-Pantothenic; 0.06mg. Cobalamine; 12mg Cu (as CuSO₄); 60mg Zn (as ZnSO₄); 24mg Mn (as MnSO₄); 0.6mg I (as Ca(IO₃)₂); 0.36mg Se (as Na₂SeO₃).

Ingredient (%)	CON	SSG-Low	SSG- High
Maize (ground)	50.99	50.99	50.99
Soybean meal (48%)	26.73	26.73	26.73
Wheat bran	1.00	1.00	1.00
Rice bran	5.00	5.00	5.00
Rapeseed meal (43%)	3.50	3.50	3.50
Tallow	6.05	6.05	6.05
Molasses	3.50	3.50	3.50
Dicalcium phosphate	1.64	1.64	1.64
Limestone	0.76	0.76	0.76
NaCl (sodium chloride)	0.50	0.50	0.50
L-Lysine-HCI (78%)	0.12	0.12	0.12
Vitamin premixa ¹⁾	0.10	0.10	0.10
Trace mineral premixb ²⁾	0.10	0.10	0.10
Phytase (5000 G)	0.01	0.01	0.01
SGG		0.15	0.30
Nutrient content			
Dry matter		88.87	
Metabolizable energy (kcal/kg)		3,456	
Crude protein (%)		18.34	
Crude fat (%)		9.16	
Lysine (%)		1.08	
Calcium (%)		1.06	
Total phosphorus (%)		0.73	

Table 2. Diet composition of lactating sow (as fed basis)

¹⁾Provided per kilogram of complete diet: vitamin A, 12,100 IU; vitamin D₃, 2000 IU; vitamin E, 48 IU; vitamin K₃, 1.5 mg; riboflavin, 6 mg; niacin, 40 mg; D-pantothenic, 17 mg; biotin, 0.2 mg; folic acid, 2 mg; choline, 166 mg; vitamin B₆, 2 mg; vitamin B₁₂, 28 μg.

²⁾Provided per kilogram of complete diet: Cu (as CuSO₄·5H₂O), 15 mg; Zn (as ZnSO₄), 50 mg; Mn (as MnO₂), 54 mg; I (as KI), 0.99 mg; Se (asNa₂SeO₃·5H₂O), 0.25 mg.

the litter size at birth per sow, and mortality ratio were also recorded. At the time of weaning (day 21 of age), sows were removed from the farrowing crate, then pigs within treatments were divided into two groups and randomly distributed in 20 replicates (n = 5 piglets/pen).

At weaning, six hundred piglets (6.72 \pm 0.5 kg) which weaned from sows supplemented with three levels of SGG (Control, SGG-Low, and SGG-High) were allocated to two weaner diets (CON and FMP – 3 kg/ton) following 3 × 2 factorial arrangement for 42 days. A corn-soybean-meal based basal diets were formulated to meet or exceed the nutrient recommendations of NRC [17] (Table 3). A master-batch of the basal diet (weaning) was prepared in mash form then FMP additive was top dressed into pre-marked feed bags, mixed well using DK-801 feed mixer (Daedong Tech, Anyang, Korea), and offered to pigs from days 0–21 (Phase 1) and days 22–42 (Phase 2). All pigs were housed in an environmentally controlled room with slatted plastic floor and had free access to feed and water throughout the experimental period. Ambient temperature was approximately 30 °C and it was reduced by 1 °C in each week of the experiment. To impose a small challenge to the animals and create stress, less cleaning was done at the facility during the experimental period.

Ingredients (%)	Days (0–21) Phase 1	Days (22–42) Phase 2		
Extruded corn	47.51	55.54		
Soybean meal (dehulled)	18.00	24.00		
Fermented soybean meal	8.00	5.00		
LT fish meal	2.70	-		
Soy oil	3.20	3.25		
Dicalcium phosphate	1.34	1.63		
Limestone	0.74	0.82		
Sugar	2.00	2.00		
Whey protein	8.00	3.00		
Lactose	6.70	3.00		
L-Lysine – HCl	0.46	0.48		
DL-Met	0.17	0.19		
Threonine	0.29	0.20		
Choline Chl 50%	0.10	0.10		
Salt	0.10	0.10		
Vitamin premix ¹⁾	0.20	0.20		
Mineral premix ²⁾	0.20	0.20		
Phytase (5000G)	0.01	0.01		
Zinc oxide	0.28	0.28		
FMP	0.30	0.30		
Nutrient content				
Protein (%)	19.00	18.50		
Fat (%)	4.80	4.20		
Calcium (%)	0.75	0.75		
Phosphorus (%)	0.65	0.65		
Digestible energy (kcal/kg)	3,900	3,800		
Lys (%)	1.50	1.40		
Met (%)	0.45	0.42		
Lactose (%)	12.00	5.00		

Table 3. Diet composition of weaning pig (as fed basis)

¹⁾Provided per kg of diet: vitamin A, 11,025 IU; vitamin D₃, 1103 IU; vitamin E, 44 IU; vitamin K, 4.4 mg; riboflavin, 8.3 mg; niacin, 50 mg; thiamine, 4 mg; D-pantothenic, 29 mg; choline, 166 mg; vitamin B₁₂, 33 µg.

 $^{2)}\mbox{Provided}$ per kg of diet: Se, 0.3 mg; I, 1 mg; Mn, 30 mg; Cu, 20 mg; Fe, 15.

Measurements

Growth performance

The body weight (BW) of individual pigs was recorded at initial and at the end of d 21 and 42 to determine the average daily gain (ADG). The amount of FI and remaining was recorded at the end of d 21 and 42 to determine the ADG and feed conversion ratio (FCR).

Fecal microbiome

Fresh and clean fecal samples (50g) were randomly collected from 2 pigs pen (both sex) (40 pigs / treatment) at d 21 and 42 by rectal palpation to determine the effect of additives on the microbiota. After collection, the samples were homogenized, diluted with sterile saline and immediately taken to laboratory to check for the presence of *Lactobacillus*, *Clostridium perfringens* and *E. coli* counts. One gram of fecal sample was diluted (1:10) with peptone water and mixed with vortex mixer. Then 0.1% peptone solution was serially diluted (10^1 to 10^9), and given to MRS agar (Difco,

USA), MacConkey agar (Difco, USA), and Clostridium perfringens agar (MB cell, Korea) to determine the *Lactobacillus, E. coli*, and *C. perfringens*, respectively. MRS agar plates were incubated anaerobically at 37° C to 38° C for 24 hours, whereas MacConkey agar and Clostridium perfringens agar plates were incubated anaerobically at 37° C for 48 hours. Later the plates were taken out, the colonies were enumerated and log transferred for statistical analysis.

Statistical analysis

The current data were analyzed as a completely randomized block design, with a 3 × 2 factorial design using the GLM procedure of SAS (Cary, NC, USA). The statistical model considered the main effects of sow treatment and weaner diet and their interactions. The batch was included as a random effect. Phases of growth were demonstrated as repeated measures on the pen as a subject. Variability in the data is expressed as the standard error means. Probability values p < 0.05 and p < 0.10 was considered as significant and trends, respectively.

RESULT

Effect of sow treatment (blend of short- and medium chain organic acids)

The dietary effect of sow diet on growth performance and fecal microbial shedding of weaning pigs is presented in Tables 4 and 5. In the first growth phase (days 0 to 21), no difference was observed in the BW of pigs. However, pigs born to sows supplemented with SGG- Low and high had a higher ADG (p < 0.001) and average daily feed intake (ADFI) (p = 0.044). Whereas, during the second growth phase (days 22 to 42), except BW (p < 0.036) and ADG (p < 0.053), the sow treatments did not affect any of the measured growth parameters. Over the entire growth phases, pigs born to

ltomo	Sow diet ¹⁾	C	CON		SGG Low		SGG High		<i>p</i> -value ³⁾		
Items	Weaner diet ²⁾	CON	FMP	CON	FMP	CON	FMP	SEM	SGG effect	FMP effect	Interaction
Body weight	t (kg)										
Initial (dag	y 0)	6.73	6.73	6.65	6.65	6.79	6.00	0.02	0.463	0.977	0.999
d 21 (Pha	ase 1)	14.24	14.27	14.28	14.32	14.47	14.52	0.05	0.124	0.713	0.995
d 42 (Pha	ase 2)	24.29	24.45	24.37	24.71	24.64	24.97	0.12	0.036	0.045	0.813
d 21											
ADG (g)		358	359	363	365	366	368	2	< 0.001	0.281	0.977
ADFI (g)		492	493	497	498	499	501	3	0.044	0.583	0.956
FCR		1.377	1.374	1.367	1.363	1.365	1.362	0.007	0.206	0.564	0.998
d 42											
ADG (g)		457	459	463	472	462	475	4	0.053	< 0.001	0.502
ADFI (g)		659	663	660	666	663	669	4	0.434	0.076	0.929
FCR		1.444	1.434	1.441	1.413	1.435	1.41	0.012	0.379	0.033	0.715
Overall (0–4	2)										
ADG (g)		408	412	412	420	415	423	3	0.006	0.005	0.682
ADFI (g)		613	615	616	620	618	623	3	0.066	0.088	0.919
FCR		1.502	1.494	1.495	1.476	1.490	1.474	0.008	0.161	0.052	0.802

¹⁾SGG Low, 1.5 kg/ton SGG; SGG High, 3 kg/ton SSG.

²⁾FMP, 3 kg/ton FMP.

 $^{3)}p < 0.05$ and p < 0.1 was considered as significant and trends, respectively.

CON, basal diet; SGG, blend of short- and medium chain organic acids; FMP, synergistic blend of free and buffered organic acid; ADG, average daily gain; FCR, feed conversion ratio.

Items	Sow diet ¹⁾	CON		SGG Low		SGG High		0514	<i>p</i> -value ³⁾		
	Weaner diet ²⁾	CON	FMP	CON	FMP	CON	FMP	- SEM	SGG effect	FMP effect	Interaction
Log ₁₀ CFU/g											
d 21 (Phase	1)										
Lactobacil	lus	9.09	9.11	9.11	9.13	9.12	9.14	0.02	0.665	0.330	0.979
Escherichi	ia coli	7.20	7.16	7.17	7.14	7.16	7.13	0.02	0.399	0.079	0.992
Clostridiun	n perfringens	8.10	8.00	8.06	7.99	8.04	7.97	0.04	0.599	0.031	0.974
d 42 (Phase 2	2)										
Lactobacil	lus	9.05	9.07	9.07	9.10	9.10	9.12	0.03	0.417	0.259	0.989
Escherichi	ia coli	7.16	7.09	7.15	7.08	7.13	7.06	0.03	0.681	0.008	0.991
Clostridiun	n perfringens	8.16	7.91	8.13	7.88	8.11	7.86	0.04	0.603	< 0.001	0.984

¹⁾SGG Low, 1.5 kg/ton SGG; SGG High, 3 kg/ton SSG.

²⁾FMP, 3 kg/ton FMP.

 $^{3)}p < 0.05$ and p < 0.1 was considered as significant and trends, respectively.

Probability values p < 0.05 and p < 0.1 was considered as significant and trends, respectively.

CON, basal diet; SGG, blend of short- and medium chain organic acids; FMP, synergistic blend of free and buffered organic acid

sow supplemented with SGG- high and-low showed higher BW (p = 0.036). Similarly, pigs born to sows supplemented with SGG-Low (+6 g/d) and -High (+9 g/d) had greater ADG than pigs born to sows fed CON diets. However, there were no differences in ADFI and FCR among the sow treatments. The fecal *Lactobacillus*, *E coli* and *C. perfringens* remains similar among the sow treatments in both sampling period.

Effect of weaner diet (synergistic blend of free and buffered organic acid)

The dietary effect of weaner diet on growth performance and fecal microbial shedding of weaning pigs is presented in Tables 4 and 5. The ADG, ADFI and FCR in the first growth phase were similar among the weaner diets. However, in the second phase, FMP supplementation increased the ADG (p < 0.001) and tendency to improve the ADFI (p = 0.076), and significantly reduced the FCR of pigs (p = 0.033). Over the entire growth phases, pigs supplemented with FMP had higher BW (p = 0.045), ADG (p = 0.005), and tendency to improve ADFI (p = 0.088), and significantly reduce FCR (p=0.052) compared to pigs in CON group. Also, FMP diet reduced the number of *E coli* (p = 0.07 and 0.008) and *C. perfringens* (p = 0.031 and < 0.001) population in pigs at both sampling period.

Interaction effect between sow treatment and weaner diet

Overall, there were no significant interactions observed between sow treatment and weaner diet on any of the measured growth parameters (Tables 4 and 5).

DISCUSSION

Weaning is the most important stage in a pig's life that determines the success or failure of production. As the weaning phase begins on day 21, pigs are familiar to digest milk-based protein supplements and have difficulty in accepting plant- and meat-based proteins supplements because of their immature digestive system [18]. It has been postulated that adding in-feed organic acid to the diet of pigs could be the best solution [19]. Previously, many researchers have exemplified the mechanism of organic acid and even proved the benefits of mixed and protected organic acid supplements in swine nutrition. For instance: Suryanarayana et al. [20] reported that dietary organic

acid had improved the growth performance and apparent total tract digestibility of pigs. Additionally, Devi et al. [7] demonstrated that the inclusion of 0.2% organic acids supplementation had increased the white blood cell (WBC) and immunoglobulin (IgG) counts in suckling piglets. Moreover, Walsh et al. [21] stated that the addition of 0.4% organic acid blend supplementation to the diet of nursery pigs had improved their growth rate, FI, and feed efficiency. The above reports were consistent with the current study, in which pigs born to sows supplemented with SGG- High and Low showed significantly increased BW, ADG, and ADFI. Also, dietary FMP supplementation showed higher BW, ADG, and ADFI in weaning pigs. However, pig fed diet supplemented with FMP reduce FCR in pigs, which was inconsistent with Metzler and Mosenthin [22] and Suryanarayana et al. [20] who observed an increased FCR in growing pigs fed organic acid supplement. Even Canibe et al. [23] reported that 1.8% of formic acid supplements to the diet of growing pig showed tendency to increase the gain-to-feed (G:F) ratio. Similarly, Ngoc et al. [24] stated that growing pigs diet supplemented with SGG had a positive impact on ADG and G:F. The probable reason for the improvement in BW, ADG, and ADFI of piglets that were born to SGG-Low and -High group sows in this study are mainly due to the antimicrobial activity of synergistic organic acid blend or due to the reduction of harmful microbial inhabitants, which helps to reduce the metabolic need of microbes and increase the availability of dietary energy and nutrients to host animals.

It is becoming increasingly clear that the gut microbiota has a significant impact on the overall health and production of pigs. Besides, the gastrointestinal microbiome of animals is a heterogeneous ecosystem and is dominated by bacteria [25]. Also, it is well documented that gut microbiota could modulate the host physiological regulation, digestion, metabolism, and immune system [26,27]. The current experiment tested the hypothesis that FMP supplement could improve the growth performance of weaning pigs by reducing the pathogenic bacteria. As anticipated, pigs fed FMP diet showed lower number of E coli and C. perfringens however, pigs born to sows supplemented with SGG-Low and -High showed no difference on their gut microbiome. Previously, Dibner and Buttin. [28] reported that the inclusion of organic acids in pigs' diet had increased the Lactobacillus counts however this result was not correlated with the current study. However, in this trial, pigs fed diet supplement with FMP reduced the number of Escherichia coli was agreed with Li et al. [29] who observed the similar result in pigs fed protected organic acid supplementation. Additionally, Devi et al. [7] reported that the addition of protected organic acid supplement in sows' diet increased Lactobacillus and reduced the E. coli counts during farrowing and weaning. Also, Upadhaya et al. [6] reported that inclusion of 0.1% organic acid mixture (10% malic, 13% citric, and 17% fumaric acids) in the diet of growing and finishing pigs had decreased E. coli counts. C. perfringens is a grampositive, anaerobic, and spore-forming bacillus that may produce major toxin microbiota in animals and humans [30]. Such toxic C. perfringens residents were significantly reduced in pigs fed FMP supplementation during both phases (d 21 and d42). In 2020, Nguyen et al. [31] demonstrated that organic acids could penetrate through the cell wall of bacteria and prevent bacterial growth in the intestine. Besides, many mechanisms could be responsible for the reduction of pathogenic bacteria, yet the reduction of C. perfringens count in this study is probably due to the impact of free and buffered organic acid which helps to prevent the entering of harmful bacteria in the gut.

CONCLUSION

The result of the present study demonstrates that SGG supplement in sow diet has a carry-over effect on piglet's growth performance. Regardless of sow treatment, pigs fed FMP diet had improved the BW and ADG, and lower the feed efficacy. Also, feeding FMP diets reduced the fecal *E. coli* and *C. perfringens* counts at d42. Thus, we recommend the application of 3kg/ton of SGG supplement in sows'

diet and subsequent feeding of piglets with FMP would be an effective strategy to improve growth rate and reduce pathogenic bacteria in post-weaned piglets.

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