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# Effects of salicylic acid on growth performance, fecal score, blood profile, and nutrient digestibility in weaned pigs

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## Abstract

An experiment was conducted to examine the effect of salicylic acid on growth performance in weaned pigs. A total of 80 weaning pigs ((Landrace × Yorkshire) × Duroc) with average initial body weight (BW) of  $6.40 \pm 1.42$  kg were randomly assigned by BW for a 6-week feeding trial. Pigs were randomly allotted to two treatments, CON group (basal diet) and SA group (basal diet + 0.05% salicylic acid). There were eight replications in each treatment, with five pigs per pen. The results showed that SA group at days 14, 28, and 42 ( $P < 0.05$ ) significantly improved the BW gain of the weaning pig compared with CON group. In addition, during the overall experiment period (1–42 days), a tendency in higher average daily gain and average daily feed intake ( $0.05 < P < 0.1$ ) were observed in pigs fed SA supplemented diet compared with those fed CON diet. Moreover, at the end of trial, the blood parameters of Fe concentration were higher in pigs ( $P < 0.05$ ) fed diet supplemented with SA. In conclusion, supplementation of SA positively influenced growth performance and serum Fe concentration, but there were no effects on nutrient digestibility and fecal score.

**Key words:** salicylic acid, weaning pigs, blood profile, nutrient digestibility, growth performance

## Résumé

Une expérience a été effectuée afin d'examiner l'effet de l'acide salicylique sur la performance de croissance chez les porcs sevrés. Un total de 80 porcs en sevrage [(Landrace × Yorkshire) × Duroc] ayant un poids corporel initial de  $6,40 \pm 1,42$  kg ont été assignés de façon aléatoire selon le poids corporel (BW — « body weight ») pour une expérience en alimentation d'une durée de 6 semaines. Les porcs ont été assignés de façon aléatoire à l'un de deux traitements : le groupe témoin (CON — « control »; diète de base) et groupe expérimental (SA — « salicylic acid »; diète de base + 0,05 % acide salicylique). Il y avait 8 réplicats par traitement avec cinq porcs par enclos. Les résultats ont montré que les porcs en sevrage du groupe SA avaient un gain de BW significativement amélioré ( $P < 0,05$ ) aux jours 14, 28, et 42 par rapport aux porcs du groupe CON. De plus, pendant tout le long de la période expérimentale (jours 1 à 42), une tendance ( $0,05 < P < 0,1$ ) vers des gains moyens quotidiens (ADG — « average daily gain ») et consommations moyennes quotidiennes (ADFI — « average daily feed intake ») plus élevés a été observée chez les porcs ayant reçu la diète avec suppléments de SA par rapport à ceux ayant reçu la diète CON. De plus, à la fin de l'expérience, les paramètres sanguins de concentration en fer (Fe) étaient plus élevés ( $P < 0,05$ ) chez les porcs ayant reçu la diète avec suppléments de SA. En conclusion, les suppléments de SA ont eu une influence positive sur la performance de croissance et les concentrations sériques en Fe, mais aucun effet sur la digestibilité des éléments nutritifs et l'indice fécal. [Traduit par la Rédaction]

**Mots-clés :** acide salicylique, porcs en sevrage, profil sanguin, digestibilité des éléments nutritifs, performance de croissance

## Introduction

For last several decades, antibiotic growth promoter has been used in the weanling pig's diet for good health and better performance. However, with the rising concerns on the adverse effects of the prolonged usage of antibiotics as growth promoter (AGP), a ban has been imposed by several countries. This has initiated the surge to find new alternatives to AGP. Among several alternatives, salicylic acid (SA) can be a potential candidate.

SA is an organic acid (OA) and it has been administered in veterinary medicine due to its medicinal value. It has been used as an antipyretic agent and in acute rheumatic events in humans for a long time and also for the treatment of joint disorders in dogs and horses (Buntenkötter et al. 2016). It is present in various amounts in vegetables, fruits, and various plants, and it has been important in responding to various types of physical, chemical, and biological functions. SA can improve the ability of plants to resist

**Table 1.** Composition of weanling pig basal diets (as-fed basis).

Items	Phase 1 (days 1–7)	Phase 2 (days 8–21)	Phase 3 (days 22–42)
Ingredients, %			
Extruded corn	25.15	36.15	25.05
Corn	—	—	38.20
Soybean meal	—	26.74	25.30
Extruded soybean	12.80	—	—
Soy oil	3.50	4.72	3.67
Fish meal	3.00	3.60	3.00
Spray dried plasma protein	6.50	3.60	—
Isolated soy protein	6.00	—	—
Lactose	18.00	19.50	2.50
Milk powder	22.00	2.60	—
MCP	0.90	—	—
MDCP	—	0.92	0.59
Limestone	0.90	0.92	0.59
Emulsifier	0.05	0.05	0.05
Mineral mix <sup>a</sup>	0.10	0.10	0.10
Vitamin mix <sup>b</sup>	0.10	0.10	0.05
Choline (25%)	1.00	1.00	1.00
Total	100.00	100.00	100.00
Calculated value			
NE, kcal/kg	2950	2800	2700
CP, %	22.00	20.00	19.70
SID Lys, %	1.50	1.40	1.28
Calcium, %	0.8	0.8	0.8
Phosphorus, %	0.6	0.6	0.6
Lactose, %	26.00	20.00	2.00
D Fat, %	10.84	8.45	8.45
Choline, ppm	1750	1750	1650

<sup>a</sup>Provided per kg of diet: Fe, 180 mg as ferrous sulfate; Cu, 17 mg as copper sulfate; Mn, 54 mg as manganese oxide; Zn, 90 mg as zinc oxide; Se, 0.36 mg as sodium selenite; I, 0.78 mg as potassium iodide.

<sup>b</sup>Provided per kg of diet: vitamin A, 10 800 IU; vitamin E, 40 IU; vitamin D3, 4000 IU; vitamin K3, 4 mg; vitamin B1, 6 mg; vitamin B2, 21.6 mg; vitamin B6, 9.6 mg; vitamin B12, 0.084 mg; biotin, 0.48 mg; folic acid, 3 mg; niacin, 84 mg; D-calcium pantothenate, 54 mg.

environmental stress and play a role in plant drought resistance, salt resistance, high-temperature resistance, cold resistance, heavy metal stress resistance, ozone resistance, and other adversities (Randjelovic et al. 2015). In addition, SA has some anti-inflammatory properties and has been used as an anti-inflammatory drug in medical practice for over 100 years (Stanley and Hegedus 2000; Randjelovic et al. 2015). SA is also used in the treatment of some skin diseases and as an additive in skin care products. SA can be detected in normal blood, in good physical condition population, suggesting the origin from plant-based food (Duthie and Wood 2011; Randjelovic et al. 2015). To our knowledge, there are no reported studies on the effects of SA supplementation in pig diets. Whether SA supplementation in weaned pig diets has a certain growth-promoting effect and the effect of improving intestinal inflammation and diarrhea is still unclear.

This study hypothesized that dietary supplementation of SA could improve growth performance in weaned piglets. Therefore, the present study was carried out to determine the effects of SA on growth performance, fecal score, blood profile, and nutrient digestibility in weaned pigs.

## Materials and methods

The experimental protocols describing the management and care of animals were reviewed and approved by the Animal Care and Use Committee of Dankook University. Pigs were handled according to the guidelines described by the Canadian Council on Animal Care (2009). In the present study, SA was used in powder form with an effective purity of 99%.

A total of 80 weanling pigs ((Landrace × Yorkshire) × Duroc) with an average initial body weight (BW) of 6.40 ± 1.42 kg were randomly assigned by BW and sex for a 6-week feeding trial in the three phases: Phase 1 (1–7 days), Phase 2 (8–21 days), and Phase 3 (22–42 days). Pigs were randomly allotted to either basal diet (CON) or a basal diet supplemented with 0.05% salicylic acid (SA) to substitute for the 0.05% corn ingredient. There were eight replications in each treatment, with five pigs per pen. All diets were provided in mash form and formulated to meet or exceed the NRC (2012) recommendations (Table 1). All pigs were raised in an individual room in 1.8 m × 1.2 m pens with plastic flooring. The room temperature was maintained at 27–29 °C, and hu-

**Table 2.** Effect of dietary salicylic acid supplementation on growth performance and nutrient digestibility in weaning pigs.

Items	CON	SA	SEM	P value
Body weight, kg				
Initial	6.38	6.42	0.026	0.568
Day 14	8.40	8.60	0.031	<0.001
Day 28	15.26	15.65	0.091	0.006
Day 42	23.02	23.60	0.152	0.012
Days 1–14				
ADG, g	144.1	146.3	2.145	0.461
ADFI, g	316.7	319.5	5.404	0.710
G:F	0.455	0.458	0.0017	0.313
Days 14–28				
ADG, g	490.4	504.0	6.327	0.128
ADFI, g	610.5	623.1	6.617	0.175
G:F	0.804	0.809	0.008	0.657
Days 28–42				
ADG, g	553.7	567.7	8.151	0.220
ADFI, g	716.7	728.9	8.119	0.281
G:F	0.773	0.779	0.011	0.684
Overall (days 1–42)				
ADG, g	396.1	405.8	3.635	0.065
ADFI, g	547.9	557.3	3.998	0.098
G:F	0.723	0.728	0.006	0.506
Nutrient digestibility				
Week 6				
Dry matter	79.47	80.87	0.680	0.167
Nitrogen	78.60	79.91	1.059	0.397
Gross energy	78.66	79.86	0.850	0.333

Note: CON, basal diet; SA, basal diet + 0.05% salicylic acid, SEM, standard error of means.

**Table 3.** Effect of dietary salicylic acid supplementation on fecal score in weaning pigs.

Items	CON	SA	SEM	P value
Fecal score <sup>a</sup>				
Initial	3.26	3.26	0.070	0.985
Week 2	3.20	3.21	0.048	0.774
Week 3	3.18	3.17	0.056	0.932
Week 4	3.16	3.15	0.059	0.878
Week 5	3.15	3.14	0.045	0.881
Week 6	3.14	3.13	0.066	0.868

Note: CON, basal diet; SA, basal diet + 0.05% salicylic acid; SEM, standard error of means.

<sup>a</sup>Fecal scores were determined using the following fecal scoring system: 1, hard, dry pellet; 2, firm, formed stool; 3, soft, moist stool that retains shape; 4, soft, unformed stool that assumes shape of container; 5, watery liquid that can be poured

midity was maintained at 60%–65%. Every pen was equipped with a plastic self-feeder and steel water nippler. All pigs had free access to their nutrition diets during the experimental period.

### Sampling and measurement

BWs of pigs were recorded initially, and at day 14, day 28, and day 42 of the experimental period. Feed consumption was recorded on a pen basis during the experiment to calculate the average daily feed intake (ADFI), average daily gain (ADG), and gain:feed ratio (G:F). Chromium ox-

ide ( $\text{Cr}_2\text{O}_3$ ) was mixed to the animal's diet at 0.5% of the diet as an indigestible marker for 7 days before fecal collection on week 6 to calculate apparent total tract digestibility (ATTD) of dry matter (DM), nitrogen (N), and gross energy (GE). Fecal grab samples were collected randomly from at least two pigs in each pen and all fresh fecal samples were pooled by pen and mixed, and immediately stored at  $-20\text{ }^\circ\text{C}$  until analysis. Before chemical analysis, the feed and fresh fecal samples were stored for 48 h at  $-10\text{ }^\circ\text{C}$  in a refrigerator (model WSFD-1900RE, Woosung Enterprise Co., Ltd, Cheonan-si, South Korea). After that, all samples were moved to dry for 72 h at  $60\text{ }^\circ\text{C}$  in a forced-air oven

**Table 4.** Effect of dietary salicylic acid supplementation on blood profile in weaning piglets.

Items	CON	SA	SEM	P value
Initial				
Fe, µg/dL	56.88	68.63	7.595	0.292
TIBC, µg/dL	579.1	655.6	21.27	0.023
Red blood cell, 10 <sup>6</sup>	6.25	6.08	0.433	0.783
Hb, mg/dL	11.33	10.38	0.890	0.462
Hematocrit, %	40.70	37.53	2.516	0.387
Week 6				
Fe, µg/dL	88.25	124.53	5.154	<0.001
TIBC, µg/dL	557.5	534.3	11.14	0.162
Red blood cell, L	6.95	6.66	0.162	0.221
Hb, mg/dL	10.73	10.68	0.217	0.874
Hematocrit, %	44.20	44.95	0.788	0.512

Note: CON, basal diet; SA, basal diet + 0.05% salicylic acid, SEM, standard error of means; TIBC, total iron binding capacity; Hb, hemoglobin.

(model FC-610, Advantec, Toyo Seisakusho Co. Ltd, Tokyo, Japan). Finally, all feed and fecal samples were grinded to a size that could pass through a 1 mm screen. After that, all the fecal and feed samples were analyzed for N (method 990.03) and DM (method 930.15) following the procedures outlined by the association of Official Analytical Chemists International (AOAC 2010). Energy was determined by measuring the heat of combustion in the samples, using a bomb calorimeter (Parr 6100; Parr Instrument Co., Moline, IL, USA). Chromium was analyzed via UV spectrophotometer (Optizen POP, Korea) (Williams et al. 1962). For calculating the ATTD of the nutrients, we used the following formula: Digestibility =  $1 - [(N_f \times C_d)/(N_d \times C_f)] \times 100$ , where  $N_f$  is the concentration of nutrient in fecal (% DM),  $C_d$  the concentration of chromium in the diet,  $N_d$  the concentration of nutrient in the diet, and  $C_f$  the concentration of chromium in the fecal.

The fecal scores were recorded for clinical signs of diarrhea initially, and at weeks 2, 3, 4, 5, and 6 in all pigs per pen in each treatment by a single blinded observer, using the 5-grade scoring system described by O'Shea et al. (2010): 1, loose and semi-liquid feces; 2, slightly soft feces; 3, well-firmed feces; 4, soft and partially formed feces; and 5, watery and mucus-like feces. At the end of the experiment, five pigs per treatment were randomly selected and 5 mL blood samples were collected into nonheparinized tubes and vacuum tubes containing K3EDTA (Becton, Dickinson and Co., Franklin Lakes, NJ, USA). After collection, blood samples were centrifuged (3000g) for 30 min at 4 °C. Contents of iron (Fe) were analyzed with flame atomic absorption spectrophotometry (AA-6300, Shimadzu Corp., Tokyo, Japan). The whole blood samples were analyzed for red blood cells and hemoglobin counts using an automatic blood analyzer (AD-VIA 120, Bayer, Tarrytown, NY).

### Statistical analysis

All data were processed by ANOVA using the MIXED procedure of SAS 9.4. Each pen was used as the experimental unit for growth performance, fecal score, and nutrient digestibility, whereas individual pigs were the experimen-

tal unit for blood profiles. Statistical differences were determined at  $P < 0.05$ . Trends were determined at  $0.05 < P < 0.1$ .

## Results and discussion

We analyzed the content of SA in the feed by sampling and testing each batch of mixed feed. The content of Phase 1 was 0.051%. The content of Phase 2 was 0.049%. The content of Phase 3 was 0.050%.

The growth performance of weaning pigs is presented in Table 2. The weaning pig's BW was significantly increased at days 14, 28, and 42 by the dietary inclusion of SA ( $P < 0.05$ ). Similarly, Kim et al. (2016) reported that weaning pig's growth performance was improved by the addition of acetyl SA. The BW and ADG were reported to be improved by the inclusion of OAs (Papatsiros et al. 2011). In the present study, the inclusion of SA to diet of weaning pigs tended to increase ADG and ADFI ( $P < 0.05$ ), but there were no effects on G:F ratio compared with control diet ( $P > 0.05$ ). Grilli et al. (2010) reported that the final BW and ADG were improved when weaning pigs were fed an additional 0.3% OA. Moreover, Freitas et al. (2006) and Boas et al. (2016) also found that mixtures of OAs based on lactic acid have no effect on the ADG weaned pigs. Therefore, it may be associated with dose level, diet composition, and feeding strategy.

Nutrient digestibility is an important factor affecting growth performance. The composition of nutrients and trace elements in the diet and feeding management are important factors that affect the digestibility of nutrients (Liu et al. 2018; Yan et al. 2021; Zhai et al. 2022). The nutrient digestibility of DM, N, and energy were not affected by the SA supplementation in diets of weaning pigs (Table 2,  $P > 0.1$ ). Similarly, OA did not exert positive effects on nutrient digestibility in weaning pigs (Kil et al. 2011; Yang et al. 2019). In contrast, several researchers reported that addition of OAs such as 2% benzoic acid and 0.5% in the diet of weaning pigs and lactating sows improved nutrient digestibility (Wang et al. 2009; Kluge et al. 2010; Nguyen et al. 2020). In addition, the nutrient digestibility of N was improved by 0.5% benzoic acid supplementation on day 21 of weaning pigs (Zhang et al. 2016).



As well as, nutrient digestibility of DM, N, and energy were improved by the dietary supplementation of 0.2% protected OA diets (Devi et al. 2016). The differences in findings across studies may be due to the animal age and feed characteristics.

Due to psychological, nutritional, and environmental stressors, weaning is often associated with postweaning diarrhea. In the present study, during the entire period, the fecal score of weaning pigs consuming SA supplementation-containing diet was not affected (Table 3,  $P > 0.1$ ). Hence, there was no incidence of diarrhea among the CON and SA groups. However, Kim et al (2016) reported that the fecal score was decreased in weaning pigs when fed with mixed 125 ppm of OAs. The reason for the absence of diarrhea incidence can be associated with good hygienic conditions in the present study.

Iron is vital for maintaining energy levels and good health in general because it is a very important part of animal's blood. Some authors reported that serum iron level is generally increased in hypoplastic or aplastic anemia, chronic hepatopathy (dogs), acute iron toxicity, experimental pyridoxine deficiency (pigs), and following the administration of glucocorticoid steroids to dogs and horses (Harvey 1997, 2011; Haldane and Davis 2009). Adding OAs to feed can effectively improve the absorption of minerals such as iron. Fe is an important component of hemoglobin, myoglobin, cytochromes, and various oxidases. Iron is an important mineral involved in the body's metabolism (NRC 2012). The blood profile of the weaning pigs is presented in Table 4. The Fe concentration was significantly increased ( $P < 0.05$ ) with the inclusion of SA in the diet. We assume that most of the iron absorbed from digestive fed or supplements maybe increase the amount of iron in the bloodstream.

## Conclusion

The supplementation of 0.05% SA had positive effects on weaned pigs' BW, cumulative ADG and ADFI and serum Fe concentration, suggesting that SA can be used as an alternative to antibiotic growth promoter. However, fecal score and nutrient digestibility of DM, N, and energy were not affected by the inclusion of SA. More studies are needed with different doses of SA to confirm the effects of SA on the performance and immune status of the weaning pigs.

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## Data availability

Data generated or analyzed during this study are provided in full within the published article.

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### Author contributions

Conceptualization: IHK. Methodology: IHK, PT. Formal analysis: IHK, PT, SC. Investigation: SC. Writing—original draft: SC. Writing—review and editing: SC, PT, IHK.

### Competing interests

The authors declare that there are no competing interests.

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