



Dietary MCFAs and Lauric Acid as Antibiotic Alternatives: Effects on Growth, Immunity and Gut Health in Weaned Pigs

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Abstract | This study aimed to reduce antibiotics in animal feed by supplement with medium-chain fatty acids (MCFAs) and lauric acid (C12) to promote piglet health. In total 48 [Duroc × (Landrace × Yorkshire)] piglets were randomly assigned into 4 groups: basal diet (T1), a basal diet with colistin 60 ppm and halquinol 400 ppm (T2), a basal diet with colistin 60 ppm and mixtures of MCFAs 0.2% (T3) and a basal diet with colistin 60 ppm and C12 0.2% (T4). Growth performance, blood hematology, immunology, intestinal morphology, feces score and microbiological examinations were collected for analysis. The results revealed that there was no significant difference between groups in growth performance, blood hematology and feces score ($P>0.05$). Immunology was significantly different for white blood cells (WBC) at day 28 with the lowest WBC $15,937.50 \pm 684.90$ in T4 ($P<0.05$). Intestinal morphology (duodenum, jejunum and ileum) was significantly different between groups ($P<0.05$) except villous height/crypt depth ratio in duodenum ($P>0.05$). There was a significant difference in bacteria and *E. coli* ($P<0.05$) but not affected for anaerobic and *Lactobacillus spp.* ($P>0.05$). Aerobics bacteria decreased to 5.96 ± 0.32 in T4. *E. coli* reduced in T1 and T4 with 2.83 ± 0.32 and 4.02 ± 0.32 , respectively. In conclusion, this study demonstrated that MCFAs may act as promoting intestinal development and health in weaned pigs.

Keywords | Medium-chain fatty acids, Lauric acid, Growth performance, Blood hematology, Intestinal morphology, Weaning pigs

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INTRODUCTION

Weaning is regarded as one of the most important stages of pig production and related to the financial benefits of pig farms. One of the most difficult periods in a pig's life is the immediate post-weaning phase (Jayaraman and Nyachoti, 2017). Piglets are separated from the

sow, the dietary and environment changes. These factors affect weaning stress in many piglets. The stress in weaning often causes function and morphology in small intestines, digestion system and absorption abilities, damages intestinal barrier function, and leads to decrease feed intake, an increase in the rate of diarrhea, and growth retardation (Tang *et al.*, 2022). Antibiotics (AB) are mostly used to

ETHICAL APPROVAL

This study was approved by Kasetsart University Institutional Animal Care and Use Committee (AC-KU67-AGR-035).

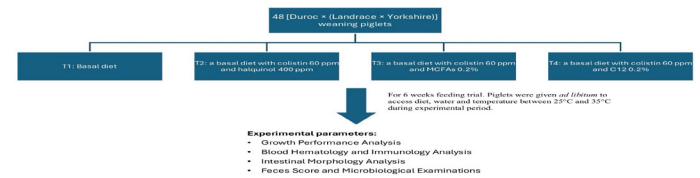


Figure 1: Animals and experimental treatments.

ANIMALS AND EXPERIMENTAL TREATMENTS

The study was conducted at the Animal Research Farm, Department of Animal Science, Faculty of Agriculture, Kasetsart University, Thailand. In total 48 [Duroc × (Landrace × Yorkshire)] piglets were randomly assigned into 4 groups: basal diet (T1), a basal diet with colistin 60 ppm and halquinol 400 ppm (T2), a basal diet with colistin 60 ppm and mixtures of MCFAs 0.2% (C6 to C12), was supplied by Nutrition Sciences NV, Drongen, Belgium (T3) and a basal diet with colistin 60 ppm and C12 0.2% (T4) (Figure 1). Diets were formulated based on the recommendations of NRC (2012) (Table 1). The piglets were assigned to 12 piglets per group. All pen sizes were approximately 0.35 square meters per piglet. Piglets were given *ad libitum* to access diet, water and temperature between 25°C and 35°C during experimental period.

GROWTH PERFORMANCE ANALYSIS

Feed intake was recorded daily and piglets were weight individual in every week to determine average daily gain (ADG), average daily feed intake (ADFI), and feed conversion ratio (FCR). The formulars were analyzed by the formula based on the research reference Yi *et al.* (2018).

BLOOD HEMATOLOGY AND IMMUNOLOGY ANALYSIS

At 28 and 42 of trial, blood samples were collected into ethylenediaminetetraacetic acid-containing tubes to prevent blood coagulation. The hematological analyses including white blood cells (WBCs), neutrophil, lymphocyte, monocyte, eosinophil and basophil were performed using a hematological analyzer (Cell Dyn 3700, Wiesbaden Hesse, Germany).

For immunology analysis, blood samples were collected into vacutainer tubes through jugular vein puncture at 28 and 42 of trial. An 8 mL blood sample was collected in the red-top serum tubes (BD Vacutainer®, UK), then immediately centrifuged at 2,000×g at 4°C for 15 minutes. Serum was separated and stored at −20°C for further analysis.

control infectious illnesses in intensive pig production systems and are a major cause of antibiotic resistance. Their prophylactic usage in feed is currently being reviewed after the EU banned the use of ABs as growth boosters. This will directly affected on piglet health after removing AB from a diet (Diana *et al.*, 2017). Short-term AB use and increasing weaning age decreased mortality and increased the weight sold per weaned pig (Faccin *et al.*, 2020). It is now commonly known that bacteria and genes resistant to antibiotics are carried from the environment and can have a negative impact on human and animal health (Hossain *et al.*, 2022). The increasing of reports related to antimicrobial resistance (AMR) in humans is recognized as one of the major health risks (Jadeja and Worrich, 2022). Since then, colistin-resistant bacteria have been discovered in humans who have not used colistin. This is strongly related to the human and animal sectors regarding the spread of AMR (Yacouba and Olowo-Okere, 2020).

Caprylic acid (C8), capric acid (C10), lauric acid (C12), and caproic acid (C6) are examples of MCFAs, which are saturated fatty acids with a carbon chain length varying from six to twelve carbons (Omachi *et al.*, 2024). Because their carbon chains are lipophilic and joined to a hydrophilic carboxyl group, MCFAs have a hydrophilic/lipophilic equilibrium with bacterial cell membranes (Hermans and De Laet, 2014). Coconut oil, milk fat, and palm kernel oil are the fat sources of MCFAs (Roopashree *et al.*, 2021). MCFAs are utilized in animal feed formulations since triglycerides are supplied at higher rates to supply free fatty acids (Lee *et al.*, 2022). MCFAs are known to have a high potential as antibacterial qualities such as against bacteria, viruses, fungus, and algae but their taste and odor are difficult to utilize in diet (Decuyper and Dierick, 2003; Zhou *et al.*, 2019). Supplementation of MCFAs in the diet affected on intestinal morphology and gut microbiota by increasing villi heights and crypt depths, and also changed the microbiota (Hanczakowska *et al.*, 2016; Świątkiewicz *et al.*, 2020). Supplements of MCFAs in diet has positive effects in pigs including increased immunity, improved gut health, promoted of gut microbial diversity and improve growth performance (Lauridsen, 2020). Antimicrobial agents from MCFAs are naturally occurring and not harmful to humans or animals. It do not cause residues or the creation of cross-resistance. They are suggested as a beneficial substitute in-feed antibiotics, used to promote growth, and prevent gastrointestinal disorders (Decuyper and Dierick, 2003).

Therefore, reducing antibiotics in animal feed by supplementing with medium-chain fatty acids (MCFAs) and lauric acid (C12) may promote piglet health. This may be a valuable alternative to in-feed antibiotics to promote animal growth and prevent gastrointestinal diseases.

Table 1: Composition of the experimental diets.

Item	Pre-Starter Feed 1	Pre-Starter Feed 2
Broken Rice	40.00	35.00
Corn	15.22	25.76
Soybean oil	1.52	2.12
SBM 48% CP	27.54	25.44
Soybean Full Fat	5.00	0.00
Rice Bran Defatted	0.00	4.38
DL-Methionine	0.36	0.17
L-Lysine HCL	1.24	0.47
L-Threonine	0.13	0.13
Calcium Carbonate	0.92	0.98
MDCP 21% P	1.61	1.08
Salt	0.35	0.30
Choline Chloride 60%	0.08	0.07
Vitamin & Mineral Premix	0.50	0.50
Sodium Bicarbonate	0.50	0.60
Milk Whey Permeate	5.00	3.00
Total	100.00	100.00
Nutrients		
ME	3,400.00	3,350.00
Protein	21.00	18.50
Fat	4.00	4.00
Fiber	2.02	2.20
Calcium	0.34	0.83
Total Phosphorus	0.65	0.70
Avail. P	0.40	0.33
Salt	0.34	0.29
Lysine	1.30	1.30
Methionine + Cystine	0.98	0.71
Methionine	0.68	0.45
Phenylalanine + Tyrosine	1.65	
Phenylalanine	0.96	
Tryptophan	0.26	0.27
Threonine	0.86	0.97
Choline	500.00	400.00
Lactose	4.00	2.40

Note: Provide per kg of complete diet: Vitamin A 4 MIU, D 0.64 MIU, E 24,000 IU, K₃ 1.4 g, B₁ 0.6 g, B₂ 0.3 g, B₆ 0.75 g, B₁₂ 14 mg, Nicotinic acid 20 g, Pantothenic acid 10 g, Folic acid 0.44 g, Biotin 0.04g, Choline 60 g, Fe 45 g, Cu 40 g, Mn 15 g, Zn 40 g, Co 0.2 g, I 0.4 g, Se 0.06 g, Carrier Added to 1 kg. Kcal/kg for ME. Percentage for Lysine, Methionine + Cystine, Methionine, Tryptophan, Threonine and Lactose. Mg/kg for Choline. SBM 48% CP = Soybean meal 48% crude protein, L-Lysine HCL = L-Lysine hydrochloride, MDCP 21% P = Monodicalciumphosphate 21% phosphorus, ME = Metabolizable energy, Avail. P = Available phosphorus.

ELISA kits were used for immunoglobulin analysis including IgG (CAS#E99-131, Bethyl Laboratories, Texas, USA) and IgA (CAS#E101-102, Bethyl Laboratories, Texas, USA). The IgG and IgA assay were quantified following the manufacturer's protocol. The absorbance values were measured at 450 nm. Immunoglobulin concentrations were determined using a standard curve prepared with known concentrations of IgG and IgA.

INTESTINAL MORPHOLOGY ANALYSIS

The intestinal samples (duodenum, jejunum, and ileum) were cut under aseptic conditions (n= 12 per group), then immediately fixed in 10% buffered neutral formalin for 24 hours. After fixation, the small intestine sections were dehydrated and embedded in paraffin. Four transverse sections were cut from each embedded tissue, mounted on glass slides, and stained with hematoxylin and eosin. Morphological analysis was measured for the height of at least ten randomly oriented villi and their corresponding crypts within each section (Pluske *et al.*, 1996) using a light microscope equipped with a calibrated eyepiece graticule (Olympus FSX100; Olympus, Tokyo, Japan).

FECES SCORE AND MICROBIOLOGICAL EXAMINATIONS

Fecal scores were systematically recorded on a daily basis throughout the experimental period, specifically from days 1 to 10 and days 15 to 24. The fecal scoring system employed in this study categorized fecal samples into four distinct levels based on consistency and visual appearance, following the classification system described by (Pérez-Calvo *et al.*, 2019): 0 = normal feces, characterized by a well-formed and solid structure; 1 = soft feces, which are less firm than normal but still retain some shape; 2 = mild diarrhea, defined by a liquid and milky consistency with minimal or no form; and 3 = severe diarrhea, marked by watery feces with no discernible structure, resembling dirty water. Following each daily data collection, the fecal areas and the hindquarters of the pigs were thoroughly cleaned to remove any remaining fecal residues.

The microbiological examinations were conducted according to (Schwaiger *et al.*, 2020). Quantification of total aerobic and anaerobic bacteria in fecal samples was performed using the spatula method to ensure a detection limit of 10² CFU/g of fecal matter. Bacterial counts were determined using microscopic examination, biochemical assays, and matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS). *Lactobacillus spp.* and *E. coli* were determined according to (Hartemink and Rombouts, 1999).

STATISTICAL ANALYSIS

All data were statistically analyzed using analysis of variance (ANOVA) with the SAS software package (SAS

Institute Inc., Cary, NC, USA). Significant differences among groups were determined using Duncan's multiple comparison test within the PROC ANOVA procedure. A significance level was used at $P < 0.05$ to identify statistically significant differences. Results are represented as means. The statistical model was used as the following model:

$$Y_{ij} = \mu + \tau_i + \varepsilon_{ij}$$

Where,

Y_{ij} : The observation of the traits.

μ : The overall mean.

τ_i : Effect of treatment ($i = T1, T2, T3$ and $T4$).

ε_{ij} : The random residual error.

Table 2: Growth performance of piglets fed with diet supplementing antibiotic, plus MCFAs, plus C12 during nursery period.

Item	Group				SEM P-value	
	T1	T2	T3	T4		
Initial BW, kg	6.81	6.87	6.53	6.64	0.17	0.89
Final BW, kg	29.22	29.00	28.12	29.40	0.43	0.73
BW Gain, kg	22.42	22.11	21.59	22.76	0.31	0.61
ADG, kg	0.53	0.53	0.51	0.54	0.01	0.61
ADFI, kg	0.88	0.86	0.86	0.84	0.01	0.76
FCR	1.66	1.64	1.66	1.55	0.14	0.14

Note: T1 = a control diet without antibiotics and additives; T2 = a control diet with colistin 60 ppm and halquinol 400 ppm; T3 = a control diet with colistin 60 ppm and mixtures of medium-chain fatty acids (MCFAs) 0.2%; T4 = a control diet with colistin 60 ppm and mixtures of lauric acid (C12) 0.2%; BW = body weight; ADG = average daily gain; ADFI = average daily feed intake; FCR = feed conversion ratio.

RESULTS AND DISCUSSION

GROWTH PERFORMANCE

There was no significant difference between groups for initial body weight, final body weight, body weight gain, average daily gain (ADG), average daily feed intake (ADFI), and feed conversion ratio (FCR) ($P > 0.05$) (Table 2). The initial body weight for T1 to T4 were 6.81, 6.87, 6.53 and 6.64 kg, respectively. The final body weight for T1 to T4 were 29.22, 29.00, 28.12 and 29.40 kg, respectively. Body weight gain for T1 to T4 were 22.42, 22.11, 21.59 and 22.76 kg, respectively. ADG for T1 to T4 were 0.53, 0.53, 0.51 and 0.54 kg, respectively. ADFI for T1 to T4 were 0.88, 0.86, 0.86 and 0.84 kg, respectively. FCR for T1 to T4 were 1.66, 1.64, 1.66 and 1.55, respectively. This study observed similar trends in growth performance which is different from the previous studies (Devi and Kim, 2014; Li *et al.*, 2015; Zhang *et al.*, 2019; Świątkiewicz *et al.*, 2020). This study was not difference in growth performance be-

cause pigs fed at 0.2% MCFAs or 0.2% C12. MCFAs often used during the weaning periods and believed that MCFAs have impacted on the gut microbiota and decreases the gastrointestinal pathogen colonization (Zheng *et al.*, 2021). Growth performance and nutrient digestion have been demonstrated to be enhanced by dietary probiotic supplements and a high-nutrient diet. MCFAs and basal diet were not significantly difference on average daily gain (ADG) and gain-to-feed ratio (Devi and Kim, 2014). Pigs fed 0.3% MCFAs showed significant improvement in average daily gain (ADG) compared to those fed a basal diet. However, pigs fed with 0.3% MCFAs was similarly ADG with antibiotics supplementation (Zhang *et al.*, 2019). Piglets fed with MCFAs in diet grew more quickly than controls given soybean oil in the first two weeks. Piglets in the experimental and control groups have the same weight gain. This might be due to the absorption and solubility properties of MCFAs that are responsible for performance improvement (Li *et al.*, 2015). Sows fed coconut oil had higher development rate of piglets. They may have been more acclimated to MCFAs and consumed coconut oil until 28th day of age while still with the sows. Moreover, they able to utilize MCFAs (Świątkiewicz *et al.*, 2020).

BLOOD HEMATOLOGY AND IMMUNOLOGY

At 28 days of trial, blood hematology including neutrophil, lymphocyte, monocyte, eosinophil, basophil, IgA and IgG were not significantly different between groups ($P > 0.05$) (Table 3). However, there were significant differences between groups in WBC ($P < 0.05$). T4 was lower WBC than T2 (15,937.50 VS 20,563.98 cell/mm³). At 42 days of trial, WBC, neutrophil, lymphocyte, monocyte, eosinophil, basophil, IgA and IgG were not significantly different between groups ($P > 0.05$). Monoglycerides and medium-chain fatty acids (MCFAs) have become a viable alternative because of important molecular features and functions, such as ability to prevent bacterial and viral infections by enhancing immunity (Jackman *et al.*, 2020). White blood cells or Leucocytes are the body's defense against invasive pathogens. They serve as the body's primary defense against illness, preventing pathogen damage and eliminating poisons and damaged cells (Glenn and Armstrong, 2019). This study represented that MCFAs and lauric acid have improved in immune of piglets as well. Lauric acid has been shown to have great potential to increase immunity (Lauridsen, 2020). Dietary supplementation of lauric acid demonstrated diarrhea disease, which, after being appropriately treated, recovered rapidly. In contrast to the control group pigs, which died in weaned piglets (Rebucci *et al.*, 2021). MCFAs supplementation increased the proportion of lymphocytes, decreased total leukocytes and the percentage of neutrophils. Moreover, MCFAs was significantly increased serum immunoglobulin M (IgM) and immunoglobulin G (IgG) levels on days 14 and 21 (Han *et al.*, 2020).

Table 3: Blood profiles and immunology of piglets fed with diet supplementing antibiotic, plus MCFAs, plus C12 during nursery period.

Item	Group				SEM	P- value
	T1	T2	T3	T4		
Day – 28						
WBC (cell/mm ²)	18,379.94 ^{ab}	20,563.98 ^a	17,893.97 ^{ab}	15,937.50 ^b	684.90	<0.05
Neutrophill (%)	34.50	33.83	28.33	38.25	1.76	0.27
Lymphocyte (%)	64.16	60.33	60.00	56.25	1.77	0.51
Monocyte (%)	4.50	4.00	3.83	3.50	0.49	0.93
Eosinophill (%)	1.50	1.67	2.67	1.50	0.34	0.55
Basophil (%)	0.50	0.33	0.33	0.00	0.10	0.53
IgA (g/L)	0.06	0.08	0.07	0.08	0.003	0.37
IgG (g/L)	1.97	1.86	1.70	1.28	0.18	0.30
Day - 42						
WBC (cell/mm ²)	23,965.00	31,291.49	26,312.50	18,660.00	2,390.86	0.25
Neutrophill (%)	20.83	16.97	16.50	18.00	1.53	0.19
Lymphocyte (%)	78.25	76.50	76.43	75.33	1.58	0.94
Monocyte (%)	4.67	4.50	3.75	3.33	0.63	0.88
Eosinophill (%)	1.53	1.25	0.50	0.50	0.22	0.19
Basophil (%)	0.00	0.39	0.25	0.50	0.10	0.36
IgA (g/L)	0.07	0.08	0.10	0.10	0.005	0.17
IgG (g/L)	2.04	2.42	2.05	1.33	0.24	0.50

^{a-b} Within a row, means without a common superscript differ ($P < 0.05$). Note: T1 = a control diet without antibiotics and additives; T2 = a control diet with colistin 60 ppm and halquinol 400 ppm; T3 = a control diet with colistin 60 ppm and mixtures of medium-chain fatty acids (MCFAs) 0.2%; T4 = a control diet with colistin 60 ppm and mixtures of lauric acid (C12) 0.2%; WBC = White blood cell.

INTESTINAL MORPHOLOGY

Intestinal morphology was analyzed for villous height, crypt depth and V/C ratio in duodenum, jejunum and ileum (Figure 2). The villous height and crypt depth in duodenum, jejunum and ileum were significantly different between groups ($P < 0.05$) (Table 4). V/C ratio in duodenum was no significant differences between groups ($P > 0.05$) but significant difference between groups in V/C ratio of jejunum and ileum ($P < 0.01$). In duodenum, T4 had the highest villous height and crypt depth. In jejunum, T3 had the highest villous height. Crypt depth was the highest in T4. The V/C ratio was the highest in T2 and T3. In ileum, villous height was the highest in group 2. Crypt depth in T1 and T4 was higher than T3. The V/C ratio was the highest in T3. Among the biggest obstacles of pig production is decreased in antibiotic use. Medium-chain fatty acids are among the many compounds that the scientific community has investigated as alternatives to antibiotics because of their antibacterial and protective effects on the gut health of piglets (Rebucci *et al.*, 2021). In addition to improving piglets' energy supply and performance, medium-chain triglycerides and medium-chain fatty acids may also stabilise the intestinal flora and promote piglets' health during the post-weaning phase (Zentek *et al.*, 2011). Villi heights

and crypt depths were increased in diet supplemented with MCFAs (Hanczakowska *et al.*, 2016; Świątkiewicz *et al.*, 2020). However, there were no significant difference on the intestinal weight, ileum inflammatory infiltrates, epithelial regeneration, oedema and hyperaemia between the treatment groups (premix with or without short and medium chain fatty acids) after 7 days post-challenge (Caprarulo *et al.*, 2023). Feed basal diet supplemented with 0.2% of phytogenic premix (PHY1) and basal diet supplemented with 0.2% of phytogenic premix added with 2000 ppm of SCFAs and MCFAs (PHY2) were no significant differences in the histological analysis between the groups (Caprarulo *et al.*, 2022).

FECES SCORE AND MICROBIOLOGICAL EXAMINATIONS

Feces score in pre-start period and start period were no significant differences between groups ($P > 0.05$) (Table 5). Bacteria was analyzed including aerobic bacteria, anaerobic bacteria, *Lactobacillus spp.* and *E. coli*. There were no significant differences with anaerobic bacteria and *Lactobacillus spp.* ($P > 0.05$) (Table 5). The aerobic bacteria and *E. coli* were significantly different between groups ($P < 0.01$). Aerobic bacteria were the lowest in T4. T2 and T4 had higher *E. coli* than T1. Medium-chain fatty acids (MCFAs; C6–

12) have shown antibacterial properties against a variety of pathogens, including *Escherichia coli* (Kovanda *et al.*, 2019).

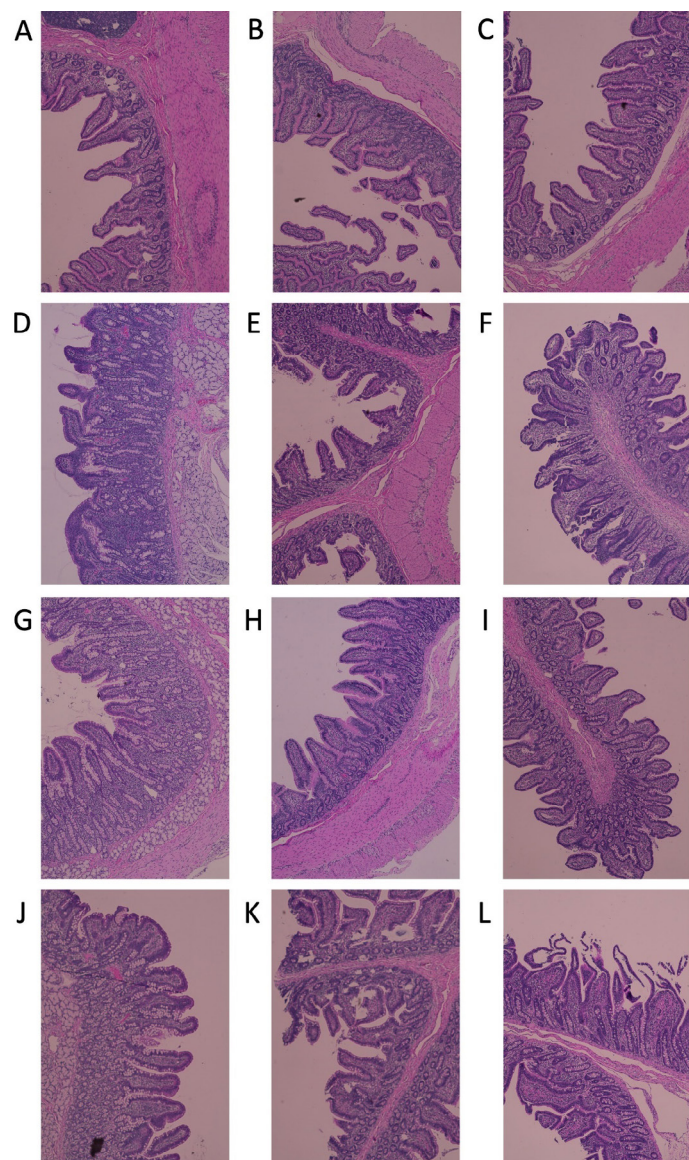


Figure 2: Intestinal histology of piglets fed with diet supplementing antibiotic, plus MCFAs, plus C12 during nursery period. T1 = a control diet without antibiotics and additives; T2 = a control diet with colistin 60 ppm and halquinol 400 ppm; T3 = a control diet with colistin 60 ppm and mixtures of medium-chain fatty acids (MCFAs) 0.2%; T4 = a control diet with colistin 60 ppm and mixtures of lauric acid (C12) 0.2%. (A) = Duodenum: T1; (B) = Jejunum: T1; (C) = Ileum: T1; (D) = Duodenum: T2; (E) = Jejunum: T2; (F) = Ileum: T2; (G) = Duodenum: T3; (H) = Jejunum: T3; (I) = Ileum: T3; (J) = Duodenum: T4; (K) = Jejunum: T4; (L) = Ileum: T4.

MCFAs are utilized for piglet feeding because they are more absorbed and metabolized. MCFAs influence on the gut microbiota and have an inhibitory effect on the bacterial concentrations in the digesta, especially on *coliforms* and *Salmonella* (Zentek *et al.*, 2011). MCFAs have become an alternative to replace antibiotics because of their function

as ability to prevent bacterial and viral infections. Monoglycerides and MCFAs demonstrate potential in feed to stop the spread of disease. In pig production, MCFAs and monoglycerides may emerge as a feed additive for enhancing gut health and reducing feed pathogens (Jackman *et al.*, 2020).

Table 4: Intestinal morphology of piglets fed with diet supplementing antibiotic, plus MCFAs, plus C12 during nursery period.

Item	Group				SEM	P-value
	T1	T2	T3	T4		
Duodenum						
Villous Hight	593.02 ^d	690.68 ^c	707.08 ^c	749.15 ^c	15.8	<0.01
Crypt Depth	295.34 ^b	294.93 ^b	339.18 ^{ab}	352.45 ^a	9.12	<0.05
V/C ratio	2.02	2.35	2.11	2.13	0.05	0.10
Jejunum						
Villous Hight	635.21 ^d	667.31 ^{cd}	698.93 ^c	641.21 ^d	7.62	<0.01
Crypt Depth	268.73 ^{ab}	240.26 ^{ab}	232.10 ^b	277.43 ^a	6.62	<0.05
V/C ratio	2.36 ^d	2.78 ^c	3.07 ^c	2.34 ^d	0.08	<0.01
Ileum						
Villous Hight	532.73 ^d	587.17 ^c	558.16 ^{cd}	560.31 ^{cd}	7.13	<0.01
Crypt Depth	257.93 ^a	247.75 ^{ab}	223.69 ^b	259.94 ^a	4.68	<0.05
V/C ratio	2.07 ^d	2.37 ^{cd}	2.57 ^c	2.15 ^d	0.06	<0.01

^{a-b} Within a row, means without a common superscript differ ($P < 0.05$). ^{c-d} Within a row, means without a common superscript differ ($P < 0.01$). Note: T1 = a control diet without antibiotics and additives; T2 = a control diet with colistin 60 ppm and halquinol 400 ppm; T3 = a control diet with colistin 60 ppm and mixtures of medium-chain fatty acids (MCFAs) 0.2%; T4 = a control diet with colistin 60 ppm and mixtures of lauric acid (C12) 0.2%; V/C ratio = Villous Height/ Crypt Depth ratio.

Table 5: Feces score and bacteria of piglets fed with diet supplementing antibiotic, plus MCFAs, plus C12 during nursery period.

Item	Group				SEM	P-value
	T1	T2	T3	T4		
Feces score Pre start	0.54	1.01	0.96	0.64	0.09	0.17
Feces score Start	0.31	0.22	0.24	0.34	0.03	0.58
Aerobic (Log CFU/g)	6.55 ^c	6.73 ^c	6.72 ^c	5.96 ^d	0.32	<0.01
Anaerobic (Log CFU/g)	7.61	7.40	7.29	7.77	0.03	0.41
<i>Lactobacillus spp.</i> (Log CFU/g)	6.41	6.35	6.25	6.31	0.03	0.41
<i>E. coli</i> (Log CFU/g)	2.83 ^d	5.50 ^c	5.47 ^c	4.02 ^{cd}	0.32	<0.01

^{c-d} Within a row, means without a common superscript differ ($P < 0.01$). Note: T1 = a control diet without antibiotics and additives; T2 = a control diet with colistin 60 ppm and halquinol 400 ppm; T3 = a control diet with colistin 60 ppm and mixtures of medium-chain fatty acids (MCFAs) 0.2%; T4 = a control diet with colistin 60 ppm and mixtures of lauric acid (C12) 0.2%.

CONCLUSIONS AND RECOMMENDATIONS

The finding in this study revealed that supplements with MCFAs and C12 were improved in piglet health as represented in WBC, villous height and crypt height. Supplement with C12 was decreased in *E. Coli* in feces. The results represent the role of MCFAs and C12 that they might be the alternatives to reduced using antibiotics in piglet diets. Further research should focus into MCFAs and biological mechanism related to animal health and production efficiency.

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NOVELTY STATEMENTS

This study's goal was to reduce antibiotics in animal feed by supplement with medium-chain fatty acids (MCFAs) and lauric acid (C12) to promote piglet health.

AUTHOR'S CONTRIBUTIONS

Kaweenuch Chiengsorn and Theerawit Poeikhampha: Conducted the study, data analysis, and drafted and revised the manuscript.

Sombat Prasongsook, Chaiyapoom Bunchasak and Autchara Kayan: Provided technical assistance during the experiments.

Theerawit Poeikhampha: Study conception and design and review the manuscript.

All authors have read and approved the final manuscript.

CONFLICT OF INTEREST

The authors declare that they have no competing interests.

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