

Growth Performance Responses and Indicators of Gastrointestinal Health in Early Weaned Pigs Fed Chinese Herbal Medicine Additives-Supplemented Diets

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Abstract: The effects of Chinese Herbal Medicine Additives (CHMD) supplemented diets on weaned piglets growth performance, incidence of diarrhea, visceral organ mass, digesta pH, luminal microbial population and small intestinal morphology were studied in a 3 weeks trial. Total of 144 crossbred (Duroc x Landrace x Yorkshire) weaning piglets (BW = 5.86±0.24 kg) from 18 L with an age of 21 days were selected and divided randomly into 4 groups balanced for sex, weight and litter origin. In each group, the piglets were divided randomly into 3 pens (replicates, 12 pigs per pen), a corn-soybean meal-expanded soybean basal diet without antibiotics or probiotics was used as control and the other 3 groups were fed the control diet supplemented with the CHMD at ratios of 0.5, 1 and 1.5% (wt/wt). In the 3 weeks trial, the data showed that. Compared to the control group, supplementation with 1% CHMD increased ($p<0.01$) Final Body Weight (FBW), Average Daily Weight Gain (ADWG) ($p<0.01$), Average Daily Feed Intake (ADFI) ($p<0.05$) and lower ($p<0.01$) F/G (total feed consumed (kg) by per kg gain in weight). Piglets fed the diet containing 1.5% CHMD had greater ($p<0.05$) ADWG, greater ($p>0.05$) ADFI, compared to the control group but without affecting F/G ($p>0.05$). There were no differences in ADFI, ADWG and F/G between the 0.5% CHMD group and the control group ($p>0.05$). Piglets fed the diet containing 1% CHMD and 1.5% CHMD both had lower ($p<0.01$) rate of diarrhoea, lower ($p<0.01$) incidence of diarrhoea and lower ($p<0.01$) index of diarrhoea compared to the control group. Piglets fed the diet containing 0.5% CHMD had lower ($p<0.01$) incidence of diarrhoea and lower ($p>0.05$) index of diarrhoea compared to the control group. Compared to the control group, piglets fed the diet containing 1% CHMD had higher ($p<0.01$) stomach weight and longer ($p>0.05$) small intestine, the 0.5% CHMD group and the 1.5% CHMD group both had no effect ($p>0.05$) on stomach weight and small intestine length. Compared to the control group, the 1% CHMD group and 1.5% both reduced the digesta pH of stomach ($p>0.05$), the 1% CHMD group reduced the digesta pH of duodenum ($p>0.05$), the 1% CHMD group and 1.5% both reduced the digesta pH of jejunum ($p>0.05$), the 1% CHMD group and 1.5% both reduced the digesta pH of ileum ($p>0.05$). Compared to the control group, the 1% CHMD group had a higher cecal and colonic lactobacilli count ($p<0.01$), a lower cecal and colonic *Escherichia coli* count ($p>0.05$) and had a higher LAB: *E. coli* ratio in the middle caecum ($p<0.01$), in the middle colon ($p>0.05$), pigs fed the diets with 1.5% CHMD had a higher cecal and colonic lactobacilli count ($p>0.05$) and had a higher LAB: *E. coli* ratio in the middle caecum ($p>0.05$). Compared to the control group, diets with 1% CHMD resulted in a greater VH in the duodenum ($p<0.01$), in the jejunum ($p<0.01$) and in the ileum ($p>0.05$), a lower CD in the duodenum ($p<0.01$), in the jejunum ($p<0.01$) and in the ileum ($p>0.05$), a greater ($p<0.01$) calculated VH:CD ratio in the duodenum in the jejunum and in the ileum, diets with 1.5% CHMD resulted in a greater ($p>0.05$) VH, a lower ($p>0.05$) CD and a greater VH:CD ratio ($p>0.05$) in the duodenum. The results show that the CHMD used in this study as a dietary additive could enhance indicators of gastrointestinal health, improve growth performance in weaned piglets, additionally imply that the dose of 1% CHMD supplement is the most ideal concentration to achieve the most beneficial effects.

Key words: Weaned piglets, chinese herbal medicine additive, growth performance, gastrointestinal health

INTRODUCTION

Frequently, several hours can pass before piglets consume feed for the first time after weaning. In addition to separation from the mother and confrontation with

piglets from another litter, the abrupt change in feed associated with weaning places great demands on the ability of the animals to adapt (Mormede and Hay, 2003). After weaning, piglet feed is abruptly changed from mainly sow's milk to solid feed only which usually reduces

the daily energy and nutrient intake (Pluske *et al.*, 1996). As a result of the reduction in feed intake there is often a regression of the intestinal villi with simultaneous deepening of the crypts which are thought to cause a decrease in the digestive and absorptive capacity of the small intestine (Hampson, 1986; Li *et al.*, 2001; Pluske, 2001). In addition, the change of diet may alter the gastrointestinal microbial balance which increases the opportunities for pathogens to colonize the gut (Fuller, 1989; Jensen, 1998). In the 1st week after weaning there is frequently a depression in growth which has wide-ranging consequences for the subsequent course of the nursery and finishing period (King and Pluske, 2003).

To optimize performance of piglets at this stage, in-feed antibiotics have been used as growth promotants and for therapeutic treatment of gastroin-testinal diseases (Verstegen and Williams, 2002).

Because of concerns with residues in meat products and bacterial resistance to antibiotics, investigating alternatives to in-feed antibiotics is required (Cromwell, 2002). Thus there is an urgent need to remove antibiotics from the list of feed additives allowed for food-producing animals. In order to improve pig performance as well as to help pigs resist the common diseases occurring in early-weaned period, the feed regulations in China allow the use of Chinese herbal medicine as an alternative to the use of in-feed antibiotics.

The aim of this research was to study the impact of supplementing a piglet diet with different dose of one complex additive of Chinese herbal medicines, selected for their pharmacological properties on growth performance, gut environment and health status in post-weaning piglets. Researchers hypothesized that supplementation of a typical piglet diet with Chinese herbal medicines will improve growth performance and reduce the incidence of diarrhoea in weaned piglets.

MATERIALS AND METHODS

The study was carried out in accordance with the Chinese guidelines for animal welfare and experimental protocol (Yin *et al.*, 2004).

Composition of the Chinese herbal medicines additive: The Chinese Herbal Medicines additive (CHMD) used in this study is consisted of seven dried Chinese herbs including *Astragalus Membranaceus*, *Scutellaria malt*, *Glycyrrhiza uralensis*, *Codonopsis pilosula*, *Poria cocos*, *Atractylodes macrocephala* and the rate is 2:1:1:1:2:2:2. Before inclusion in the feed, they were mixed according

the aforesaid ratio and crumbled to an ultra-fine powder with an average granule diameter of 30 μm , packed in hermetical plastic bags and stored at room temperature. All the medicinal herbs were purchased from WeiMing pharmacy in Hefei city of China.

Animals, diets and experimental design: At weaning, at 21 days of age, 144 crossbred (Duroc x Landrace x Yorkshire) piglets (72 females and 72 males) with a body weight of 5.86 ± 0.24 kg were selected from 18 L (among the original 21 L) that were healthy and had not been treated by any antibiotics and were divided randomly into 4 groups balanced for sex, weight and litter origin. In each group, the piglets were divided randomly into 3 pens (12 animals per pen) and each group was fed one of 4 diets for 3 weeks.

The pens had concrete floors with no litter and each pen was equipped with a feeder and nipple drinker. The nursery had a temperature of 27.0°C in the 1st week after weaning. From week 2 until the end of the nursery, the temperature was decreased weekly by 0.5°C . The photoperiod was controlled to provide 12 h of light and 12 h of dark in the stall. The ventilation also was provided to ensure good air quality. All piglets were vaccinated against pasteurellosis, paratyphoid, asthma and hog cholera.

A basal diet without antibiotics or probiotics was used as control and was fed to one group (C). The other 3 groups were fed the control diet supplemented with 0.5, 1 and 1.5% (wt/wt) CHMD (Treatment 1, T2 and T3). The basal diet mainly contained maize, soyabean meal, expanded soybean, milk replacer, whey powder, soybean oil and a premix of vitamins and minerals and the nutrient contents met or exceeded nutrient requirements recommended by National Research Council (1998). The piglets were fed *ad libitum* and had free access to water. The diets were fed in meal form and the CHMD (Sealed, placed in the cool dry place in separate bags) was mixed into the basal diet every day.

Performance monitoring: The feed offered and refused was weighed daily to calculate daily feed intake. The piglets were weighed at the beginning of the research (at weaning) on day 21 and Body Weight (BW), Average Daily Feed Intake (ADFI), Average Daily Weight Gain (ADWG), Feed/Gain ratio (F/G, total feed consumed (kg) by per kg gain in weight) were measured in the whole period were calculated.

Assessment of severity of diarrhea: To ascertain the health status of the pigs, faecal consistency was evaluated daily during 21 days after weaning as follows:

score 0: firm, dry faeces; score 1: pasty faeces; score 2: thick, fluid faeces and score 3: watery faeces (De Cupere *et al.*, 1992; Kelly *et al.*, 1990). The incidence of diarrhoea (%) was calculated as the sum of the total number of diarrhoeal piglets over the period divided by the number of piglet days in the period multiplied by 100. The rate of diarrhoea (%) was calculated as the sum of the total number of diarrhoeal piglets over the period by the sum of the total number of piglets multiplied by 100. The index of diarrhoea was calculated as the sum of the diarrhoea score over the period divided by the sum of the total number of piglets.

Digesta collection and histological measurements and bacterial enumerations: At the end of the 3 weeks study period, 1 pig selected at random from each pen was held under halothane general anesthesia and killed by an intra-cardiac injection of sodium pentobarbital. Stomach, small intestine and caecum, colon were removed and flushed with ice-cold physiological saline solution containing phenylmethyl sulfonyl fluoride (2 L of 0.9% saline, pH 7.4+2 mL of 100 mM phenylmethyl sulfonyl fluoride) to remove any excess blood and 20 mL each of digesta from the stomach and the small intestine were obtained for pH measurement. All pH measurements were made with an electronic pH meter (Accumet Basic, Fisher Scientific) which was standardized with certified pH 4 and 7 buffer solutions.

Approximately 1 g each of digesta sample from the caecum and the colon were obtained for microbial counts. The bacterial flora in digesta samples were estimated by culture methods using selective media. Digesta samples were dissolved in sterile normal saline (0.85%) in a 1:10 dilution. Secondary dilutions were from 10^{-4} - 10^{-5} for the digesta to estimate the *E. coli* population. For estimation of the lactobacilli population, the secondary dilutions were from 10^{-5} - 10^{-6} for samples. *E. coli* was cultured in MacConkey agar (MAC) and LAB was cultured in MRS agar (Mann, Rogosa and Sharpe). Each dilution was performed in duplicate and the result was the average of 2 dilutions. The digesta microbial enumerations were expressed as log₁₀ Colony Forming Units (CFU) in fresh matter.

After blotting the organs with an absorbent study, weight and length (small intestine) were determined and 10 cm segments of the duodenum, jejunum and ileum were taken and stored in 10% formalin to fix the villi and the crypts for subsequent histological measurement according to the procedures described by Owusu-Asiedu *et al.* (2003). Briefly, 6 cross-sections were obtained from each formalin-fixed segment and processed for histological examination using the standard

hematoxylin and eosin method. Villous height was measured from the tip to the crypt-villus junction and crypt depth was measured from the crypt-villus junction to the base on 10 well-oriented villi per specimen using a Zeiss photomicroscope equipped with a Sony 3 chip CCD color camera (Carl Zeiss Canada Ltd., Toronto, Ontario, Canada). Villus: crypt ratio was calculated by dividing villus height by crypt depth. Duodenum sample was taken at 30 cm away from the stomach. Jejunum sample was taken at 2 m before the ileal-cecal junction. Ileum was sampled at 30 cm before the ileal-cecal junction. The images were captured using Northern Eclipse Image Processing Software (Empix Imaging, Inc., Mississauga, Ontario, Canada).

Statistical analyses: Data are presented as arithmetic means with standard deviation of the mean (Mean±SD). Differences among groups were compared by SPSS18.0 statistics software using one way ANOVA and LSD method test and $p < 0.05$ was selected as significant standard, $p < 0.01$ was selected as remarkably significant standard.

RESULTS

Growth performance: The performance data measured during 3 weeks period in experiment are shown in Table 1. The initial BW was similar among diets at 5.86 ± 0.24 kg (mean±SD). In the 3 weeks trial, piglets fed the diet containing 1% CHMD had greater ($p < 0.01$) Final Body Weight (FBW), greater ($p < 0.01$) Average Daily Weight Gain (ADWG), greater ($p > 0.05$) Average Daily Feed Intake (ADFI) and lower ($p < 0.01$) F/G compared to the control group. Piglets fed the diet containing 1.5% CHMD had greater ($p > 0.05$) ADWG, greater ($p > 0.05$) ADFI, compared to the control group but without affecting F/G ($p > 0.05$). There were no differences in ADFI, ADWG and F/G between the 0.5% CHMD group and the control group ($p > 0.05$).

Diarrhoea: During the 3 weeks post-weaning, some piglets in all groups showed diarrhoea symptoms (Table 2). However, none of the pigs had a diarrhoea score of 3 (watery faeces). In the 3 weeks trial, piglets fed the diet containing 1% CHMD and 1.5% CHMD both had lower ($p < 0.01$) rate of diarrhoea, lower ($p < 0.01$) incidence of diarrhoea and lower ($p < 0.01$) index of diarrhoea compared to the control group. Piglets fed the diet containing 0.5% CHMD had lower ($p < 0.01$) incidence of diarrhoea and lower ($p > 0.05$) index of diarrhoea compared to the control group. Compared to the 1.5% CHMD group, the 1% CHMD had lower ($p > 0.05$) rate of diarrhoea, lower ($p > 0.05$) incidence of diarrhoea and lower ($p < 0.01$) index of diarrhoea. Compared to the 0.5% CHMD group, the

Table 1: Effects of CHMD on the growth performance parameters of weaning piglets

Parameters	CHMD dose in wt/wt (%)			
	0	0.5	1	1.5
Initial BW (kg)	5.973±0.083	5.803±0.266	5.840±0.462	5.830±0.026
Final BW (kg)	9.910±0.356 ^{ab}	10.040±0.147 ^{bb}	10.990±0.308 ^{aa}	10.350±0.200 ^{ABb}
ADWG (kg)	0.188±0.016 ^{bc}	0.202±0.019 ^{bbc}	0.245±0.010 ^{aa}	0.215±0.011 ^{ABb}
ADFI (kg)	0.387±0.005 ^b	0.392±0.002 ^{ab}	0.396±0.001 ^a	0.395±0.005 ^a
F/G	2.074±0.162 ^{aa}	1.954±0.194 ^{ABa}	1.615±0.069 ^{Bb}	1.840±0.099 ^{ABab}

In the same row values with different small letter superscripts mean significant difference ($p < 0.05$) and with different capital letter superscripts mean extremely significant difference ($p < 0.01$). The same as below

Table 2: Effects of CHMD on diarrhea parameters of weaning piglets

Parameters	CHMD dose in wt/wt (%)			
	0	0.5	1	1.5
Rate of diarrhoea (%)	36.11±4.82 ^{Aa}	27.78±4.81 ^{ABab}	11.11±4.82 ^{Cc}	22.22±4.81 ^{BCb}
Incidence of diarrhoea (%)	4.89±0.23 ^{Aa}	2.91±1.01 ^{Bb}	1.32±0.61 ^{Bc}	2.78±0.40 ^{Bb}
Index of diarrhoea	3.44±0.17 ^{Aa}	2.64±0.64 ^{ABb}	1.36±0.27 ^{Cc}	2.39±0.13 ^{Bb}

Table 3: Effects of CHMD on organ weights and digesta pH of weaning piglets

Parameters	CHMD dose in wt/wt (%)			
	0	0.5	1	1.5
Organ weight or length				
Stomach (g)	69.66±2.19 ^{Bb}	73.91±2.65 ^{ABb}	82.09±2.21 ^{Aa}	74.78±4.68 ^{ABb}
Small intestine (cm)	1175.52±3.81 ^b	1181.45±8.05 ^{ab}	1190.76±6.54 ^a	1182.24±7.89 ^{ab}
Digesta pH				
Stomach	5.60±0.10 ^a	5.43±0.15 ^{ab}	5.33±0.58 ^b	5.37±0.58 ^b
Duodenum	6.23±0.21 ^a	6.10±0.10 ^a	5.83±0.58 ^b	6.07±0.58 ^a
Jejunum	6.73±0.58 ^a	6.70±0.10 ^a	6.43±0.58 ^b	6.50±0.10 ^b
Ileum	7.40±0.26 ^a	7.20±0.10 ^{ab}	6.87±0.58 ^b	7.07±0.15 ^b

Table 4: Effects of CHMD on LAB and *E. coli* counts in different intestinal segments of weaning piglets

Microorganism (log ₁₀ cfu g ⁻¹)	CHMD dose in wt/wt (%)			
	0	0.5	1	1.5
Caecum				
Lactobacilli	8.21±0.11 ^{Bc}	8.31±0.13 ^{ABbc}	8.60±0.11 ^{Aa}	8.44±0.10 ^{ABab}
<i>Escherichia coli</i>	6.56±0.08 ^a	6.45±0.12 ^{ab}	6.31±0.13 ^b	6.38±0.17 ^{ab}
LAB: <i>E. coli</i> ratio	1.25±0.01 ^{Bc}	1.29±0.04 ^{ABbc}	1.36±0.03 ^{Aa}	1.32±0.04 ^{ABab}
Colon				
Lactobacilli	8.14±0.07 ^{Bb}	8.25±0.11 ^{ABab}	8.32±0.04 ^{Aa}	8.27±0.04 ^{ABa}
<i>Escherichia coli</i>	6.40±0.05 ^a	6.38±0.07 ^{ab}	6.29±0.06 ^b	6.34±0.05 ^{ab}
LAB: <i>E. coli</i> ratio	1.27±0.01 ^b	1.29±0.03 ^{ab}	1.32±0.02 ^a	1.30±0.01 ^{ab}

1% CHMD group had lower ($p < 0.01$) rate of diarrhoea, lower ($p < 0.01$) index of diarrhoea and lower ($p > 0.05$) incidence of diarrhoea.

Organ weights and digesta pH: Visceral organ weights (length for small intestine) and digesta pH results are shown in Table 3. In the 3 weeks trial, piglets fed the diet containing 1% CHMD had higher ($p < 0.01$) stomach weight compared to the control group. Compared to the control group, the 0.5% CHMD group and the 1.5% CHMD group both had no effect ($p > 0.05$) on stomach weight. Compared to the control group, the 1% CHMD group had longer ($p > 0.05$) small intestine.

Compared to the control group, the 1% CHMD group and 1.5% both reduced the digesta pH of stomach ($p > 0.05$), the 1% CHMD group reduced the digesta pH of duodenum ($p > 0.05$), the 1% CHMD group and 1.5% both reduced the digesta pH of jejunum ($p > 0.05$), the 1%

CHMD group and 1.5% both reduced the digesta pH of ileum ($p > 0.05$). The 1% CHMD group had lower ($p > 0.05$) digesta pH of duodenum than the 0.5% CHMD group and 1.5% CHMD group had lower ($p > 0.05$) digesta pH of jejunum than the 0.5% CHMD group had lower ($p > 0.05$) digesta pH of ileum than the 0.5% CHMD group and 1.5% CHMD group.

Bacterial counts: Table 4 shows data relative to microbiota in the digestive tract characterized as the lactobacilli and *E. coli* counts in the middle caecum and in the middle colon. Pigs fed the diets with 1% CHMD had a higher cecal and colonic lactobacilli count ($p < 0.01$), a lower cecal and colonic *Escherichia coli* count ($p > 0.05$) and had a higher LAB: *E. coli* ratio in the middle caecum ($p < 0.01$) in the middle colon ($p > 0.05$) than pigs fed the diets without CHMD. Pigs fed the diets with 1.5% CHMD had a higher cecal and colonic lactobacilli count ($p > 0.05$)

Table 5: Effect of CHMD supplemented diets on intestinal morphology in early-weaned piglets

Intestinal morphology (µm)	CHMD dose in wt/wt (%)			
	0	0.5	1	1.5
Duodenum				
Villous Height (VH)	530.975±11.63 ^{Bc}	537.17±10.52 ^{Bbc}	579.11±10.12 ^{Aa}	555.70±10.49 ^{ABb}
Crypt Depth (CD)	203.25±7.3500 ^{Aa}	197.82±7.060 ^{ABb}	170.70±8.790 ^{Bc}	187.79±7.150 ^{ABb}
VH:CD	2.62±0.1400 ^{Bc}	2.17±0.120 ^{Bbc}	3.40±0.190 ^{Aa}	2.99±0.220 ^{ABb}
Jejunum				
Villous Height (VH)	453.20±17.560 ^{Bb}	460.92±11.21 ^{Bb}	515.37±14.55 ^{Aa}	476.82±13.71 ^{ABb}
Crypt Depth (CD)	188.49±9.6200 ^{Aa}	177.67±8.170 ^{ABa}	162.59±5.480 ^{Bb}	176.56±5.860 ^{ABb}
VH:CD	2.41±0.2200 ^{Bb}	2.59±0.150 ^{Bb}	3.17±0.190 ^{Aa}	2.70±0.130 ^{ABb}
Ileum				
Villous Height (VH)	431.98±14.610 ^B	439.83±15.05 ^b	471.74±16.31 ^a	449.66±15.69 ^{ab}
Crypt Depth (CD)	177.51±6.1200 ^a	173.71±5.050 ^a	162.10±5.780 ^b	168.34±7.440 ^b
VH:CD	2.44±0.1600 ^{Bb}	2.53±0.130 ^{ABb}	2.91±0.17 ^{Aa}	2.68±0.190 ^{ABb}

and had a higher LAB: *E. coli* ratio in the middle caecum ($p>0.05$) than pigs fed the diets without CHMD. Pigs fed the diets with 1% CHMD had a higher cecal lactobacilli count ($p>0.05$), a higher LAB: *E. coli* ratio in the middle caecum ($p>0.05$) than pigs fed the diets with 0.5% CHMD.

Morphology of the small intestine: Table 5 showed the structural characteristics of the mucous membrane in the small intestine segments. Among dietary treatments various interactions were observed. Diets with 1% CHMD resulted in a greater VH in the duodenum ($p<0.01$) in the jejunum ($p<0.01$) and in the ileum ($p>0.05$), a lower CD in the duodenum ($p<0.01$) in the jejunum ($p<0.01$) and in the ileum ($p>0.05$), a greater ($p<0.01$) calculated VH:CD ratio in the duodenum, in the jejunum and in the ileum, compared to the control group. Diets with 1.5% CHMD resulted in a greater ($p>0.05$) VH, a lower ($p>0.05$) CD and a greater VH:CD ratio ($p>0.05$) in the duodenum, compared to the control group. Compared to the 0.5% CHMD group, the 1% CHMD group had a greater VH in the duodenum ($p<0.01$), in the jejunum ($p<0.01$) and in the ileum ($p>0.05$), a lower CD in the duodenum ($p<0.01$) in the jejunum ($p>0.05$) and in the ileum ($p>0.05$), a greater VH:CD ratio in the duodenum ($p<0.01$) in the jejunum ($p<0.01$) and in the ileum ($p>0.05$). Compared to the 1.5% CHMD group, the 1% CHMD group had a greater VH in the duodenum ($p>0.05$), in the jejunum ($p>0.05$), a lower CD in the duodenum ($p>0.05$), a greater VH:CD ratio in the duodenum ($p>0.05$) and in the jejunum ($p>0.05$).

DISCUSSION

The inclusion of the Chinese herbal medicines additive in the current experiment increased FBW, ADWG, ADFI and decreased F/G of the pigs throughout the duration of the experiment (Table 1). This is in general agreement with previous studies in weaned piglets given

a herbal plant extract or a mixture of Chinese herbal ultra-fine powder (Kong *et al.*, 2007), a single Chinese herbal medicine BaZhen (Lien *et al.*, 2007) or a single Chinese herbal medicine *Acanthopanax senticosus* extract (Kong *et al.*, 2009). This study showed that 1% CHMD supplement in the diet was enough to achieve the most beneficial effects. This positive response to CHMD maybe due to a number of reasons.

Firstly, the improved performance may be attributable to an increase in the lactobacilli population and a reduction in *E. coli* populations in the gut of the CHMD fed pigs (Table 4). *Lactobacillus* sp. are known to produce lactic acid, proteolytic enzymes and cell-associated polysaccharide depolymerases which can enhance nutrient digestion in the gastrointestinal tract (Veizaj-Delia *et al.*, 2010; Dillon *et al.*, 2010). Lactobacilli can colonize and adhere to the gastrointestinal tract epithelium forming a protective membrane against pathogenic microorganisms while at the same time modulate immunity with stimulating epithelial lymphocytes (Yu *et al.*, 2008). *Lactobacilli* sp. are in abundance post weaning rapidly converting lactose to lactic acid (Pierce *et al.*, 2006) through fermentation in the hindgut. This further causes a reduced intestinal pH and thereby unfavorable conditions for coliform bacteria. Lactobacilli represent the largest group of microorganisms in the small intestine and are considered important to maintain good intestinal health because of their ability to control potentially pathogenic groups such as *E. coli* (Blomberg *et al.*, 1993; Canibe and Jensen, 2003) and to optimize immune response (Perdigon *et al.*, 2001). Accordingly, researchers use the LAB: *E. coli* ratio as an index of intestinal equilibrium (Hillman *et al.*, 1995). Xu *et al.* (2003) reported that Chinese herbs could reduce the density of enterotoxigenic *E. coli* and increase the density of *Bacillus acidilactici* or *Bacillus bifidus*. The results from the present study also suggest the potential of CHMD in suppressing pathogenic bacteria and enriching beneficial bacteria.

Secondly, CHMD supplementation exerted a positive influence on intestinal morphology in the weaned piglets of the experiment (Table 5). The gut plays an important role in the digestion, absorption and metabolism of nutrients. Findings from the previous (McCracken *et al.*, 1999; Fang *et al.*, 2009) study demonstrated that piglets showed a decrease in villus height and an increase in crypt depth during the weaning process, villus height and crypt depth are indirect indications of the maturity and functional capacity of enterocytes and larger villi and crypts are associated with a greater number of enterocytes (Hampson *et al.*, 1985). Both VH and CD are related to the absorptive capacity of the mucous membrane (Buddle and Bolton, 1992). The results indicated that dietary supplementation with CHMD effectively increased villus height in the duodenum, jejunum and ileum and decreased crypt depth in the gut, compared with the control group. The above findings suggest that CHMD could effectively improve the recovery of stress-induced damage to the gut morphology which might be a potential mechanism by which CHMD increases the nutrient uptake from intestine and result in improved growth performance in weaned piglets.

In addition, gastrointestinal pH was measured to provide an indication of the effect of feeding CHMD-supplemented diets on indicators of gastrointestinal health. The pH values in the current study are in close agreement with those reported previously (Owusu-Asiedu *et al.*, 2003). Decreased pH values in the stomach reduce the gastric emptying rate (Francois, 1962). Higher intestinal pH is thought to provide an optimal environment for enterotoxigenic *E. coli* to colonize the villi, leading to diarrhea (Smith and Jones, 1963). A lower pH, on the other hand may favor development of beneficial bacteria and/or inhibit development of harmful bacteria (Fuller, 1977) and has been shown to have a beneficial effect on nutrient digestibility (Canibe and Jensen, 2003; Lyberg *et al.*, 2006).

A novel and important finding from the present study is that dietary supplementation with the CHMD reduced the incidence of diarrhea, the rate of diarrhoea and the index of diarrhoea in piglets throughout the duration of the research (Table 2). The lower diarrhoea parameters in piglets fed diets supplemented with CHMD suggests a more healthy gut. Obviously, the anti-diarrhea effect of the CHMD was great which noted that the antibiotic effect of the CHMD was good. Diarrhea results from an increase in water secretion from the intestinal epithelial cells and/or a decrease in the absorption of water and nutrients from the intestinal lumen. Thus, it is likely that the CHMD regulates these two physiological processes by improving the metabolism of amino acids and glucose (the major fuels) and enhancing the anti-oxidant activity

in the small-intestinal mucosa (Wu *et al.*, 2004). Alternatively, the dietary supplementation with the Chinese herbs enhances the intestinal immune function thereby reducing inflammation in the small-intestinal mucosa that often occurs in weaning piglets (Nabuurs, 1995). Although, the precise mechanisms are not clear, the results demonstrate the feasibility of using the Chinese herbs as natural green dietary additives for early-weaned piglets to replace feed antibiotics.

CONCLUSION

In summary, these findings suggest that CHMD as a dietary additive could enhance gastrointestinal health by regulating the microbiota composition and maintaining a normal morphology in weaned piglets, thereby decreasing the incidence of diarrhea resulting from weaning stress and results in improved growth performance.

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