Effects of Saccharomyces cerevisiae Fermentation Broth on Growth Performance, Small Intestinal Development, and Mucosal Immune Function in Weaned Piglets (Postprint)

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Abstract

The present study aimed to investigate the effects of Saccharomyces cerevisiae (S. cerevisiae) fermentation broth on growth performance, small intestinal development, and small intestinal mucosal immune function in weaned piglets. Sixty 26-day-old "Landrace × Duroc" weaned piglets with an average body weight of (6.57$±$0.13) kg (half male and half female) were selected and randomly divided into 3 groups according to body weight and sex, with 4 replicates per group and 5 piglets per replicate. The three groups were: control group (basal diet + 300 mL/kg blank culture medium), Saccharomyces cerevisiae fermentation broth group (basal diet + 300 mL/kg Saccharomyces cerevisiae fermentation broth), and antibiotic group (basal diet + 20 mg/kg colistin sulfate + 40 mg/kg zinc bacitracin). The experimental period lasted 21 days. The results showed: 1) Compared with the control group, the average daily gain (ADG) and average daily feed intake (ADFI) in the Saccharomyces cerevisiae fermentation broth group and antibiotic group were significantly or extremely significantly increased ($P<0.05$ or $P<0.01$), and the feed-to-gain ratio was extremely significantly decreased ($P<0.01$); however, there were no significant differences in the above growth performance indices between the Saccharomyces cerevisiae fermentation broth group and antibiotic group ($P>0.05$). 2) Compared with the control group, the total protein, DNA, and RNA contents in the duodenal, jejunal, and ileal mucosa of the Saccharomyces cerevisiae fermentation broth group and antibiotic group were significantly or extremely significantly increased ($P<0.05$ or $P<0.01$); however, there were no significant differences in the above indices between the Saccharomyces cerevisiae fermentation broth group and antibiotic group ($P>0.05$). 3) Compared with the control group, the villus height in the duodenum and ileum of the Saccharomyces cerevisiae fermentation broth

group and antibiotic group was significantly increased (P<0.05), and the villus height/crypt depth (V/C) ratio in the duodenum, jejunum, and ileum was significantly or extremely significantly increased (P<0.05 or P<0.01); however, there were no significant differences in the above indices between the Saccharomyces cerevisiae fermentation broth group and antibiotic group (P>0.05). 4) Compared with the control group, the contents of immunoglobulin A, immunoglobulin G, and immunoglobulin M in the duodenal, jejunal, and ileal mucosa of the Saccharomyces cerevisiae fermentation broth group and antibiotic group were significantly or extremely significantly increased (except for jejunal mucosal immunoglobulin G) (P<0.05 or P<0.01); however, there were no significant differences in the above indices between the Saccharomyces cerevisiae fermentation broth group and antibiotic group (P>0.05). The results demonstrated that dietary supplementation with Saccharomyces cerevisiae fermentation broth could improve growth performance, promote small intestinal development, and enhance small intestinal mucosal immune function in weaned piglets, achieving effects comparable to those of antibiotics. This suggests that Saccharomyces cerevisiae fermentation broth can effectively alleviate weaning stress, reduce or replace antibiotic use in weaned piglet diets, and provides strong theoretical basis and data support for the development of antibiotic-free diets.

Full Text

Effects of Saccharomyces cerevisiae Fermentation Broth on Growth Performance, Small Intestine Development, and Small Intestinal Mucosal Immune Function of Weaned Piglets

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Abstract

This study investigated the effects of Saccharomyces cerevisiae (S. cerevisiae) fermentation broth on growth performance, small intestine development, and small intestinal mucosal immune function in weaned piglets. Sixty 26-day-old "Landrace × Duroc" weaned piglets (half male and half female) with an average body weight of (6.57$±$0.13) kg were randomly allocated into three groups based on body weight and sex, with four replicates per group and five piglets per replicate. The three groups were: a control group (basal diet + 300 mL/kg blank culture medium), an S. cerevisiae fermentation broth group (basal diet

+ 300 mL/kg S. cerevisiae fermentation broth), and an antibiotics group (basal diet + 20 mg/kg colistin sulfate + 40 mg/kg bacitracin zinc). The experimental period lasted 21 days.

The results showed: (1) Compared with the control group, the average daily gain (ADG) and average daily feed intake (ADFI) in the S. cerevisiae fermentation broth and antibiotics groups were significantly or extremely significantly increased ($P<0.05$ or $P<0.01$), while the feed-to-gain ratio was extremely significantly decreased ($P<0.01$). However, no significant differences were observed in these growth performance indices between the S. cerevisiae fermentation broth and antibiotics groups ($P>0.05$). (2) The contents of total protein, DNA, and RNA in the duodenal, jejunal, and ileal mucosa were significantly or extremely significantly elevated in the S. cerevisiae fermentation broth and antibiotics groups compared with the control group ($P<0.05$ or $P<0.01$), with no significant differences between the two treatment groups ($P>0.05$). (3) Villus height in the duodenum and ileum, as well as the villus height-to-crypt depth ratio (V/C) in the duodenum, jejunum, and ileum, were significantly or extremely significantly increased in the S. cerevisiae fermentation broth and antibiotics groups ($P<0.05$ or $P<0.01$), again with no significant differences between these groups ($P>0.05$). (4) The contents of immunoglobulin A (IgA), immunoglobulin G (IgG), and immunoglobulin M (IgM) in the duodenal, jejunal, and ileal mucosa were significantly or extremely significantly increased in the S. cerevisiae fermentation broth and antibiotics groups (except for IgG in jejunal mucosa) ($P<0.05$ or $P<0.01$), with no significant differences between the two treatment groups ($P>0.05$).

These findings demonstrate that dietary supplementation with S. cerevisiae fermentation broth can improve growth performance, promote small intestine development, and enhance small intestinal mucosal immune function in weaned piglets, achieving effects comparable to antibiotics. This suggests that S. cerevisiae fermentation broth can effectively alleviate weaning stress and reduce or replace antibiotic use in weaned piglet diets, providing a robust theoretical basis and data support for developing antibiotic-free feed formulations.

Keywords: Saccharomyces cerevisiae; weaned piglets; small intestine development; immunoglobulin; weaning stress

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Early weaning is an important technology for improving sow productivity and pig production efficiency, offering considerable economic value. However, early weaning often triggers severe stress responses in the gastrointestinal tract of piglets, leading to microbial flora imbalance, pathogen proliferation, growth retardation, and increased mortality—a condition commonly known as weaning stress syndrome. Research by McCracken et al. suggests that growth retardation in weaned piglets is primarily caused by dietary antigenic proteins triggering autoimmune responses that damage intestinal mucosa, resulting in diarrhea and reduced immunity and feed intake. While antibiotics added to weaned piglet

diets can improve growth performance and disease resistance to some extent, their extensive use causes problems including antibiotic resistance, disruption of normal intestinal flora, and environmental pollution from excretions, posing potential threats to human health. Consequently, developing antibiotic alternatives is imperative for healthy livestock production and meat safety. Yeast and its metabolites represent ideal antibiotic substitutes, exerting positive effects on growth performance, intestinal development, and immune function in weaned animals. Previous studies have shown that dietary supplementation with fermented products can increase ADFI and ADG while decreasing F/G in weaned piglets, achieving effects similar to antibiotics. As a beneficial microorganism, S. cerevisiae effectively competes with and inhibits pathogen proliferation during proliferation, protecting intestinal flora and promoting intestinal mucosal development while enhancing nutrient digestion and absorption. Approved by agricultural authorities domestically and internationally as a feed additive, S. cerevisiae produces beneficial components during fermentation—including cell wall polysaccharides, active probiotics, secondary metabolites, minerals, vitamins, and unknown growth-promoting factors—that help maintain intestinal microecological balance and enhance immune function.

Therefore, this study investigated the effects of S. cerevisiae fermentation broth on growth performance, small intestine development, and small intestinal mucosal immune function in weaned piglets to provide theoretical reference and basis for developing safe, efficient, and environmentally friendly antibiotic-free piglet diets.

1. Materials and Methods

1.1. Experimental Materials

The S. cerevisiae auxotrophic strain INVScl used in this study was purchased from Invitrogen (USA). The yeast strain was inoculated onto yeast extract peptone dextrose agar (YPD agar) plates and cultured at 30 °C for 24 h. Plate counting revealed a yeast concentration of 4$×10^{7}$ CFU/mL.

Primary materials and equipment included: enzyme-linked immunosorbent assay (ELISA) kits (Sigma, USA), BCA protein quantification kits (Beijing Zoman Biotechnology Co., Ltd.), Trizol reagent kits (Invitrogen, USA), Multiskan MK3 microplate readers (Thermo Fisher Scientific), and digital trinocular microscopes (BA400Digital, Motic Group).

1.2. Experimental Animals and Design

Sixty 26-day-old "Landrace × Duroc" weaned piglets (half male and half female) with an average body weight of (6.57$±$0.13) kg were randomly divided into three groups based on body weight and sex, with four replicates per group and five piglets per replicate. The three groups were: a control group (basal diet + 300 mL/kg blank culture medium), an S. cerevisiae fermentation broth group

(basal diet + 300 mL/kg S. cerevisiae fermentation broth), and an antibiotics group (basal diet + 20 mg/kg colistin sulfate + 40 mg/kg bacitracin zinc). The experimental period lasted 21 days.

1.3. Experimental Diets and Management

The composition and nutrient levels of the basal diet are presented in Table 1 . The experiment was conducted at the Enping Animal Testing Center in Guangdong Province. Piglets were managed according to conventional feeding systems, with five piglets per pen provided ad libitum access to feed (multiple small meals to minimize waste) and water. Routine immunization procedures were followed, and disease incidence and treatments were recorded.

Table 1. Composition and nutrient levels of the basal diet (as-fed basis), %

Items	Content
Ingredients	
Corn	
Wheat meal	
Whey powder	
Soybean meal	
Soybean oil	
Fish meal	
L-Lys · HCl	
DL-Met	
L-Thr	
Sugar	
Limestone	
NaCl	
$CaHPO_4$	
Vitamin premix[1]	
Mineral premix[2]	
Total	
Nutrient levels[3]	
ME (MJ/kg)	
CP	
EE	
AP	
L-Lys	
D-Met	
Met+Cys	
Thr	

[1]Vitamin premix provided the following per kg of diet: VA 10,000 IU, VD_3

1,500 IU, VE 50 IU, VB_1 4.50 mg, riboflavin 60.00 mg, nicotinic acid 36.00 mg, pantothenic acid 1.00 mg, VB_6 10.00 mg, folic acid 2.00 mg, cobalamin 0.01 mg, biotin 0.50 mg, VK_3 2.00 mg, VC 200 mg.

[2]Mineral premix provided the following per kg of diet: $FeSO_4$ (Fe) 100 mg, $CuSO_4$ (Cu) 6.00 mg, $MnSO_4$ (Mn) 4.00 mg, $ZnSO_4$ (Zn) 100 mg, Na_2SeO_3 (Se) 0.30 mg, KI (I) 0.30 mg, $CoCO_4$ (Co) 0.14 mg.

[3]Nutrient levels were calculated values.

1.4. Sample Collection

At the end of the experiment, piglets were fasted for 12 h. Four piglets with body weights close to the group average were randomly selected from each group and anesthetized via intravenous injection of 150.00 mg pentobarbital sodium per kg body weight. After anesthesia, piglets were slaughtered following standard procedures. Small intestine segments (2.5 cm each) were collected from the duodenum (approximately 10 cm from the pylorus), jejunum (mid-section), and ileum (approximately 5 cm from the ileocecal junction), fixed in 10% neutral formalin solution for paraffin sectioning and hematoxylin-eosin (HE) staining to assess intestinal morphology. The remaining portions of each segment were longitudinally opened, rinsed with sterile phosphate-buffered saline (PBS), and the mucosa was scraped off with glass slides and stored at -80 °C for further analysis.

1.5. Measurement Indicators

1.5.1. Growth Performance Daily feed provision and residual amounts were recorded for each pen to calculate average daily feed intake. Body weight was measured before morning feeding at the beginning and end of the experiment to calculate average daily gain. Feed-to-gain ratio was calculated as average daily feed intake divided by average daily gain.

1.5.2. Small Intestine Development HE-stained paraffin sections (cross-sections) were observed under an optical microscope to measure villus height and crypt depth in the duodenum, jejunum, and ileum. Additionally, intestinal mucosa from these segments was weighed, and total protein content was determined using BCA protein quantification kits. RNA and DNA contents were calculated based on absorbance values at 260 nm measured with a UV spectrophotometer.

1.5.3. Small Intestinal Mucosal Immunoglobulin Content Duodenal, jejunal, and ileal mucosa samples were dissolved in 2 mL PBS (pH=7.4) and centrifuged at 3,500×g for 5 min. The supernatant was collected, and IgA, IgG, and IgM contents were detected following ELISA kit instructions. Immunoglobulin analyses were completed in collaboration with Chengdu Lai Lai Biological Laboratory.

1.6. Statistical Analysis

Experimental data were initially processed using Excel 2015, and significant differences were tested using one-way ANOVA in SPSS 18.0 statistical software. Results are expressed as means ± standard deviation. $P<0.01$ was considered extremely significant, $P<0.05$ significant, and $0.05\leq P<0.10$ indicative of a trend.

2. Results

2.1. Effects of S. cerevisiae Fermentation Broth on Growth Performance of Weaned Piglets

As shown in Table 2 , compared with the control group, the S. cerevisiae fermentation broth and antibiotics groups exhibited extremely significantly higher ADFI ($P<0.01$), significantly higher ADG ($P<0.05$), and extremely significantly lower feed-to-gain ratio ($P<0.01$). No significant differences were observed in these growth performance indices between the S. cerevisiae fermentation broth and antibiotics groups ($P>0.05$). These results indicate that dietary supplementation with S. cerevisiae fermentation broth can improve growth performance in weaned piglets with efficacy comparable to antibiotics.

Table 2. Effects of S. cerevisiae fermentation broth on growth performance of weaned piglets

Items	Control	S. cerevisiae fermentation broth group	Antibiotics group	P-value
Initial body weight (kg)	6.58$±0.15	6.59±0.19	6.56±0.09	
	$Final body weight (kg)$	11.72±0.29	12.91±0.44	13.02±0.58
	$ADFI (g$			
	$sup >$			
	$Bb <$			
	$/sup >$			
	466.76±7.94 <			
	$sup >$			
	$Aa <$			
	$/sup >$			
	471.01±18.96 <			
	$sup >$			
	$Aa <$			
	$/sup >$			
	$ADG (g)$	244.52±16.76 <		
	$sup >$			
	$b <$			
	$/sup >$			
	301.31±28.18 <			
	$sup >$			
	$a <$			
	$/sup >$			
	307.62±29.98 <			
	$sup >$			
	$a <$			
	$/sup >$			
	F/G	1.83±0.09 <		
	$sup >$			
	$Aa <$			
	$/sup >$			
	1.56±0.12 <			
	$sup >$			
	$Bb <$			
	$/sup >$			
	1.54±$0.11Bb			

In the same row, values with no letter or the same letter superscripts indicate no significant difference (P>0.05), different lowercase letters indicate significant difference (P<0.05), and different capital letters indicate extremely significant difference (P<0.01). The same applies below.

2.2. Effects of S. cerevisiae Fermentation Broth on Total Protein, DNA, and RNA Contents in Small Intestinal Mucosa of Weaned Piglets

As presented in Table 3 , compared with the control group, the S. cerevisiae fermentation broth and antibiotics groups showed significantly increased total protein and DNA contents ($P<0.05$) and extremely significantly increased RNA content ($P<0.01$) in duodenal mucosa, as well as extremely significantly increased total protein, DNA, and RNA contents in jejunal and ileal mucosa ($P<0.01$). No significant differences were found in these indices between the S. cerevisiae fermentation broth and antibiotics groups ($P>0.05$). These findings suggest that dietary S. cerevisiae fermentation broth can stimulate intestinal mucosal growth and promote intestinal development in weaned piglets with efficacy comparable to antibiotics.

Table 3. Effects of S. cerevisiae fermentation broth on total protein, DNA, and RNA contents in small intestinal mucosa of weaned piglets

Items	Control	S. cerevisiae fermentation broth group	Antibiotics group	P-value
Total protein				

Items	Control	S. cerevisiae fermentation broth group	Antibiotics group	P-value
Duodenum	26.33$±1.92	20.00		
	sup >			
	b <			
	/*sup* >			
	|32.20±3.71 <			
	sup >			
	a <			
	/*sup* >			
	|32.54±2.66 <			
	sup >			
	a <			
	/*sup* >			
	|||*Jejunum*|18.54±1.75 <			
	sup >			
	Bb <			
	/*sup* >			
	|27.38±4.08 <			
	sup >			
	Aa <			
	/*sup* >			
	|27.50±1.63 <			
	sup >			
	Aa <			
	/*sup* >			
	|||*Ileum*|18.50±2.00 <			
	sup >			
	Bb <			
	/*sup* >			
	|26.56±1.75 <			
	sup >			
	Aa <			
	/*sup* >			
	|27.73±2.37 <			
	sup >			
	Aa <			
	/*sup* >			
	| <			
	0.001||∗			
	∗*DNA*∗			
	∗|||||*Duodenum*|3.59±0.43 <			
	sup >			
	b <			
	/*sup* >			
	|4.55±0.49 <			
	sup >			
	a <			
	/*sup* >			
	|4.60±0.43 <			
	sup >			
	a <			
	/*sup* >			
	|||*Jejunum*|3.39±0.13 <			
	sup >			

2.3. Effects of S. cerevisiae Fermentation Broth on Small Intestinal Morphology of Weaned Piglets

As shown in Figure 1 [Figure 1: see original paper] and Table 4 , compared with the control group, the S. cerevisiae fermentation broth and antibiotics groups exhibited significantly increased villus height in the duodenum and ileum ($P<0.05$) and significantly or extremely significantly increased villus height-to-crypt depth ratio (V/C) in the duodenum, jejunum, and ileum ($P<0.05$ or $P<0.01$). No significant differences were observed in these indices between the S. cerevisiae fermentation broth and antibiotics groups ($P>0.05$). These results indicate that dietary S. cerevisiae fermentation broth can increase the surface area of intestinal mucosa for nutrient absorption in weaned piglets.

Figure 1. Detection of small intestine morphology of weaned piglets

Table 4. Effects of S. cerevisiae fermentation broth on small intestinal morphology of weaned piglets

Items	Control	S. cerevisiae fermentation broth group	Antibiotics group	P-value
Villus height (m)				

Items	Control	S. cerevisiae fermentation broth group	Antibiotics group	P-value
Duodenum	505.25$±15.33 < sup > b < /sup >	562.25±44.92 < sup > a < /sup >	570.50±24.73 < sup > a < /sup >	
		Jejunum	485.50±19.02 < sup > Bc < /sup >	515.75±20.92 < sup > Bb < /sup >
		Ileum	495.00±36.83 < sup > b < /sup >	544.25±18.73 < sup > a < /sup >
				565.25±29.23 < sup > a < /sup >
				<
				0.001
				*
Cryptdepth(μm)				
		Duodenum	406.33±12.53	390.26±43.84
		Jejunum	401.29±26.97	372.29±9.53
Villusheight/Cryptdepth(V/C)				
		Duodenum	1.25±0.07 < sup > b < /sup >	1.47±0.07 < sup > a < /sup >
			1.46±0.18 < sup > a < /sup >	

2.4. Effects of S. cerevisiae Fermentation Broth on Immunoglobulin Contents in Small Intestinal Mucosa of Weaned Piglets

As shown in Table 5 , compared with the control group, the S. cerevisiae fermentation broth and antibiotics groups exhibited significantly increased IgA and IgG contents ($P<0.05$) and extremely significantly increased IgM content ($P<0.01$) in duodenal mucosa; significantly increased IgA and IgM contents ($P<0.05$) in jejunal mucosa; and extremely significantly increased IgA and IgM contents ($P<0.01$) and significantly increased IgG content ($P<0.05$) in ileal mucosa. No significant differences were found in these indices between the S. cerevisiae fermentation broth and antibiotics groups ($P>0.05$). These results suggest that dietary S. cerevisiae fermentation broth can enhance immune function in weaned piglets.

Table 5. Effects of S. cerevisiae fermentation broth on immunoglobulin contents in small intestinal mucosa of weaned piglets

Items	Control	S. cerevisiae fermentation broth group	Antibiotics group	P-value
IgA				

Items	Control	S. cerevisiae fermentation broth group	Antibiotics group	P-value
Duodenum	1.99$±0.20 <			

sup >
b <
/sup >
|2.30±0.06 <
sup >
a <
/sup >
|2.35±0.13 <
sup >
a <
/sup >
|||Jejunum|1.95±0.22 <
sup >
b <
/sup >
|2.39±0.13 <
sup >
a <
/sup >
|2.31±0.20 <
sup >
a <
/sup >
|||Ileum|1.81±0.22 <
sup >
Bb <
/sup >
|2.34±0.15 <
sup >
Aa <
/sup >
|2.43±0.15 <
sup >
Aa <
/sup >
||| *
IgG
*|||||Duodenum|2.12±0.06 <
sup >
b <
/sup >
|2.63±0.11 <
sup >
a <

/sup >
|2.76±0.42 <
sup >
a <
/sup >
|||Jejunum|2.16±0.12|2.58±0.29|2.84±0.61|||Ileum|2.05±0.10 <
sup >
b <
/

3. Discussion

3.1. Effects of S. cerevisiae Fermentation Broth on Growth Performance of Weaned Piglets

S. cerevisiae improves gastrointestinal environment and flora structure by providing nutritional substrates to microbial communities, stabilizing gastrointestinal pH, promoting beneficial bacteria proliferation, and increasing their concentration and activity. This enhances nutrient digestion, absorption, and utilization, increases feed intake, improves feed efficiency, and ultimately enhances growth performance. Reducing early weaning stress in piglets has long been a research focus, with antibiotics commonly added to piglet diets to mitigate negative effects. However, growing concerns about antibiotic residues and resistance have highlighted the importance of S. cerevisiae as an alternative. This study found that dietary S. cerevisiae fermentation broth significantly increased ADFI and ADG while decreasing feed-to-gain ratio in weaned piglets. Wang et al. reported similar results in weaned SD rats fed the same S. cerevisiae culture broth, showing significantly increased ADG and decreased feed-to-gain ratio comparable to antibiotic effects. Consistent results have been reported by Mathew et al., Wang et al., and Jurgens et al. in weaned piglets. The current study also found no significant differences in growth performance between the S. cerevisiae fermentation broth and antibiotics groups. Hu et al. reported that fermented feed rich in lactic acid bacteria and yeast improved growth performance in weaned piglets without significant differences compared to antibiotics. While antibiotics promote animal growth, they induce resistance and food safety concerns, making S. cerevisiae supplementation a promising alternative.

3.2. Effects of S. cerevisiae Fermentation Broth on Small Intestine Development of Weaned Piglets

As a protein source, S. cerevisiae produces beneficial nutrients during fermentation—including cell wall polysaccharides, secondary metabolites, and amino acids—and continues to secrete various metabolites in the intestinal tract that promote intestinal development. The small intestine serves as the primary site for digestion and absorption and is a key indicator of digestive function; maintaining normal intestinal mucosal structure is essential for adequate nutrient digestion and absorption. Villus height and crypt depth reflect the surface area for nutrient contact and absorption, as well as absorptive capacity. The V/C ratio comprehensively reflects small intestinal functional status, with higher values indicating more complete mucosal structure and stronger absorptive function. Early weaning abruptly eliminates milk-derived growth factors and immunoglobulins, reducing immunity, promoting pathogen proliferation, causing microbial flora imbalance, and altering small intestinal villus structure and morphology, thereby inducing diarrhea and reduced appetite. This study demonstrated that dietary S. cerevisiae fermentation broth significantly increased total protein, DNA, and RNA contents in small intestinal mucosa, as well as villus height in the duodenum and ileum and V/C ratios throughout the small intestine. In-

creased villus height enhances the number of mature enterocytes and the surface area for nutrient contact and absorption, thereby improving absorptive capacity. The precise mechanism by which yeast improves small intestinal mucosal structure remains unclear but may involve β-glucans and mannans in the yeast cell wall, which reduce antigen binding to gastrointestinal mucosa by adsorbing, phagocytosing, destroying, and absorbing invading bacteria, thereby protecting gastrointestinal mucosa—a conclusion supported by Iji et al. and Zhang et al. Other researchers suggest that yeast may promote intestinal development through polyamines, which are essential for cell growth and differentiation and can increase intestinal epithelial cell proliferation rates. Additionally, S. cerevisiae may improve small intestinal mucosal structure by producing organic acids that lower intestinal pH, inhibiting pathogen colonization by E. coli and other harmful bacteria. In this study, no significant differences were observed between S. cerevisiae fermentation broth and antibiotics regarding small intestinal development. While antibiotics effectively inhibit pathogen proliferation, they disrupt gastrointestinal microbial flora structure, whereas S. cerevisiae protects intestinal flora through competitive inhibition of harmful pathogens. Further research is needed to explore the effects of S. cerevisiae on intestinal flora structure and development in weaned piglets.

3.3. Effects of S. cerevisiae Fermentation Broth on Small Intestinal Mucosal Immune Function of Weaned Piglets

Mannan, a polysaccharide in yeast cell walls, acts as an immune activator that effectively enhances the immune system by stimulating humoral and cellular immunity, thereby increasing immunity and disease resistance. Zhou et al. reported that dietary yeast culture increased serum IgA, IgG, and IgM levels in broiler chickens without significant differences compared to antibiotics, possibly by reducing harmful bacteria and enhancing immune system responsiveness. This study found that dietary S. cerevisiae fermentation broth significantly increased IgA and IgM contents in small intestinal mucosa of weaned piglets, consistent with reports by Zhou et al. and Yue et al. As a probiotic, S. cerevisiae promotes immune organ growth and maturation, increasing T and B lymphocyte numbers to help initiate immune responses, while also metabolizing and synthesizing bacteriocins such as lactopeptides that stimulate immune responses and activate the immune system. However, Wang et al. found that dietary S. cerevisiae did not increase serum IgA, IgG, and IgM levels in weaned SD rats, possibly due to differences in animal species, S. cerevisiae dosage, or mechanisms of action, warranting further investigation.

In conclusion, dietary supplementation with S. cerevisiae fermentation broth can improve growth performance, promote small intestine development, and enhance small intestinal mucosal immune function in weaned piglets, achieving effects comparable to antibiotics. These results suggest that S. cerevisiae fermentation broth can effectively alleviate weaning stress and reduce or replace antibiotic use in weaned piglet diets, providing robust theoretical basis and data support

for developing antibiotic-free feed formulations.

References

[1] Li W, Feng PG, Wang T. Weaning stress and its nutritional regulation in piglets. *Journal of Domestic Animal Ecology*, 2007, 28(6): 1–4.

[2] Wang XR, He JH, Dai QZ, et al. Effects of early weaning stress on intestinal mucosal barrier function and its detection and repair in piglets. *Chinese Journal of Animal Nutrition*, 2014, 26(11): 3197–3202.

[3] McCracken BA, Gaskins HR, Ruwekaiser PJ, et al. Diet-dependent and diet-independent metabolic responses underlie growth stasis at weaning. *The Journal of Nutrition*, 1995, 125(11): 2838–2845.

[4] Wang SJ, Wang BX, Guo CH, et al. Application of recombinant S. cerevisiae secreting pEGF in weaned piglets. *Acta Veterinaria et Zootechnica Sinica*, 2016, 47(5): 944–954.

[5] Wang SJ, Chen HN, Zhang ZF, et al. Effects of S. cerevisiae on production performance, intestinal development, blood physicochemical indices, and immune function of weaned rats. *Journal of Domestic Animal Ecology*, 2014, 35(10): 41–45.

[6] Wang SJ, Zhou L, Chen HN, et al. Cloning and expression of recombinant porcine epidermal growth factor in S. cerevisiae and identification of its biological activity. *Acta Veterinaria et Zootechnica Sinica*, 2014, 45(12): 1971–1980.

[7] Hu XX, Zhou YH, Liu HZ, et al. Effects of antibiotic-free fermented feed on growth performance, intestinal flora, blood biochemical indices, and immune function of weaned piglets. *Chinese Journal of Animal Nutrition*, 2013, 25(12): 2989–2997.

[8] Mathew AG, Chattin SE, Robbins CM, et al. Effects of a direct-fed yeast culture on enteric microbial populations, fermentation acids, and performance of weanling pigs. *Journal of Animal Science*, 1998, 76(8): 2138–2145.

[9] Wang SJ, Guo CH, Zhou L, et al. Effects of dietary supplementation with epidermal growth factor-expressing S. cerevisiae on duodenal development in weaned piglets. *British Journal of Nutrition*, 2016, 115(9): 1509–1520.

[10] Gao J. Effects of yeast culture on broiler chickens and its mechanism. PhD dissertation. Beijing: Chinese Academy of Agricultural Sciences, 2008.

[11] Xiao M, Gao ZH, Li XH, et al. Effects of yeast culture on growth performance, intestinal mucosal structure, and intestinal flora of broiler chickens. *Chinese Journal of Animal Nutrition*, 2013, 25(7): 1624–1631.

[12] Yu XR. Application prospects of yeast and its culture. *China Feed Additive*, 2008(9): 9–12.

[13] Wang SJ, Guo CH, Zhou L, et al. Comparison of the biological activities of S. cerevisiae-expressed intracellular EGF, extracellular EGF, and tagged EGF in early-weaned pigs. *Applied Microbiology and Biotechnology*, 2015, 99(17): 7125–7135.

[14] Jurgens MH, Rikabi A, Zimmerman DR, et al. The effect of dietary active dry yeast supplement on performance of sows during gestation-lactation and their pigs. *Journal of Animal Science*, 1997, 75(3): 593–597.

[15] Wang ZX, She RP, Chen Y, et al. Effects of dietary zinc and selenium levels on small intestinal mucosal structure in broiler chickens. *Chinese Veterinary Science*, 2003, 33(7): 18–.

[16] Dong KS, Xiao ZD. Observation of effects of acid-producing probiotics on small intestinal villus morphology in newborn piglets. *Journal of Jilin Agricultural University*, 1994, 16(1): 93–96.

[17] Hampson DJ. Alterations in piglet small intestinal structure at weaning. *Research in Veterinary Science*, 1986, 40(1): 32–40.

[18] Iji PA, Saki AA, Tivey DR. Intestinal structure and function of broiler chickens on diets supplemented with a mannan oligosaccharide. *Journal of the Science of Food and Agriculture*, 2001, 81(2): 1186–1192.

[19] Zhang AW, Lee BD, Lee SK, et al. Effects of yeast (S. cerevisiae) cell components on growth performance, meat quality, and ileal mucosa development of broiler chicks. *Poultry Science*, 2005, 84(7): 1015–1021.

[20] Bontempo V, Di Giancamillo A, Savoini G, et al. Live yeast dietary supplementation acts upon intestinal morpho-functional aspects and growth in weanling piglets. *Animal Feed Science and Technology*, 2006, 129(3/4): 224–236.

[21] Costalos C, Skouteri V, Gounaris A, et al. Enteral feeding of premature infants with S. boulardii. *Early Human Development*, 2003, 74(2): 89–96.

[22] Van Heugten E, Funderburke DW, Dorton KL. Growth performance, nutrient digestibility, and fecal microflora in weanling pigs fed live yeast. *Journal of Animal Science*, 2003, 81(4): 1004–1012.

[23] Lin BQ, Yang H, Wang T. Effects of yeast cell wall and probiotics on production performance and immune function of laying hens. *Chinese Journal of Animal Nutrition*, 2014, 26(5): 1327–1332.

[24] Zhou SQ, Sun WZ. Effects of yeast culture and antibiotics on growth performance and immune function of broiler chickens. *Animal Husbandry and Veterinary Medicine*, 2004, 36(11): 9–11.

[25] Yue ZH, Zeng JM. Yeast cell wall (PR-500) and its application in animal production. *China Fisheries*, 2000, 294(5): 68–69.

[26] Wang SJ, Zhou L, Chen HN, et al. Analysis of the biological activities of S. cerevisiae expressing intracellular EGF, extracellular EGF, and tagged EGF in

early-weaned rats. *Applied Microbiology and Biotechnology*, 2015, 99(5): 2179–2189.

[27] Zhang TF. Effects of antibiotic-free fermented feed on pig immune function. *Chinese Animal Husbandry and Veterinary Abstracts*, 2011, 27(5): 180.

Note: Figure translations are in progress. See original paper for figures.