Effects of Plant Polysaccharides on Growth Performance and Intestinal Environment of Weaned Piglets (Postprint)

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Abstract

This experiment aimed to investigate the effects of individual or combined supplementation of Astragalus polysaccharide, Atractylodes polysaccharide, and Achyranthes polysaccharide in the diet of weaned piglets on their growth performance and intestinal environment.

A total of 256 Duroc × Landrace × Yorkshire (DLY) three-way crossbred weaned piglets at 35 days of age were selected and randomly divided into 8 groups with 4 replicates per group and 8 piglets per replicate. A three-factor two-level (2$×2×$2) experimental design was adopted, with supplementation levels of Astragalus polysaccharide, Atractylodes polysaccharide, and Achyranthes polysaccharide in the diet set at 0 and 800 mg/kg. The experiment lasted for 28 days.

The results showed that: 1) Individual or pairwise combined supplementation of Astragalus polysaccharide, Atractylodes polysaccharide, and Achyranthes polysaccharide in the diet of weaned piglets showed a trend toward increasing average daily gain (P>0.05), while the combined use of all three polysaccharides significantly increased the average daily gain of weaned piglets (P<0.05). There was no significant difference in feed-to-gain ratio among groups (P>0.05). Individual supplementation of Astragalus polysaccharide, Atractylodes polysaccharide, or Achyranthes polysaccharide in the diet of weaned piglets significantly reduced the diarrhea rate (P<0.05), and there were certain interaction effects among the polysaccharides.

2) Compared with the control group, there was no significant change in the pH of ileal and rectal contents in weaned piglets from the individual or combined polysaccharide supplementation groups (P>0.05), while the pH of cecal and colonic contents was significantly decreased (P<0.05). The

combined supplementation of all three polysaccharides had an interaction effect on cecal content pH (P<0.05).

3) Compared with the control group, the total volatile fatty acid content in cecal contents of weaned piglets in the individual or combined polysaccharide supplementation groups was significantly increased (P<0.05), and the effect of the triple combination was significantly higher than that of individual supplementation (P<0.05).
4) Individual or combined supplementation of plant polysaccharides in the diet of weaned piglets significantly increased the numbers of Lactobacillus and Bifidobacterium in the intestine (P<0.05). The numbers of Lactobacillus and Bifidobacterium in the pairwise combination groups showed an increasing trend compared with the individual supplementation groups, but the difference was not significant (P>0.05). Individual or combined supplementation of polysaccharides significantly reduced the number of Escherichia coli in the intestine (P<0.05).

It can be concluded that dietary supplementation with Astragalus polysaccharide, Atractylodes polysaccharide, and Achyranthes polysaccharide can improve the intestinal environment, reduce piglet diarrhea, and thereby improve the growth performance of weaned piglets, with the combined supplementation of all three showing the best effect.

Full Text

Effects of Botanical Polysaccharides on Growth Performance and Intestinal Environment of Weaned Piglets

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Abstract: This experiment was conducted to investigate the effects of Astragalus polysaccharides (APS), Atractylodes macrocephala polysaccharides (AMP), and Achyranthes bidentata polysaccharides (ABP) on growth performance and intestinal environment of weaned piglets. A total of 256 Duroc × Landrace × Yorkshire crossbred piglets at 35 days of age were randomly allocated into 8 groups with 4 replicates per group and 8 piglets per replicate. A 2$×2×$2 factorial design was employed, with APS, AMP, and ABP each added at either 0 or 800 mg/kg in the diets for a 28-day feeding trial. The results showed: (1) Individual or pairwise supplementation of APS, AMP, and ABP tended to increase average daily gain (ADG) of weaned piglets (P>0.05), while the combination of all three polysaccharides significantly improved ADG (P<0.05). No significant differences were observed in feed-to-gain ratio among all groups (P>0.05). Individual supplementation of APS, AMP, or ABP significantly reduced diarrhea rate (P<0.05), with certain interactive effects among polysaccharides. (2) Compared with the control group, dietary polysaccharide supplementation did not significantly alter pH values in ileal and rectal contents (P>0.05), but significantly decreased pH in cecal and colonic contents (P<0.05), with a significant interactive effect observed for the triple combination on cecal pH (P<0.05). (3) Total volatile fatty acid (VFA) content in cecal digesta was significantly elevated in all polysaccharide-supplemented groups compared with the control (P<0.05), with the triple combination showing significantly greater effects than individual supplementation (P<0.05). (4) Individual or combined supplementation of plant polysaccharides significantly increased intestinal Lactobacillus and Bifidobacterium counts (P<0.05), while pairwise combinations showed a numerical increase over individual supplementation without statistical significance (P>0.05). All polysaccharide treatments significantly reduced Escherichia coli populations (P<0.05). In conclusion, dietary supplementation with APS, AMP, and ABP improved intestinal environment, reduced diarrhea, and consequently enhanced growth performance of weaned piglets, with the triple combination demonstrating optimal efficacy.

Keywords: Astragalus polysaccharides; Atractylodes macrocephala polysaccharides; Achyranthes bidentata polysaccharides; growth performance; intestinal environment

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Introduction

Weaning represents a major stressor for piglets, as the dramatic changes in diet form and composition destabilize the intestinal microenvironment, altering microbial populations and diversity while modifying intestinal structure and function [1]. Suboptimal management during this period can lead to "growth depression," characterized by reduced feed intake, increased maintenance requirements, heightened susceptibility to intestinal pathogens, and elevated risk of colibacillosis [2]. For decades, antibiotics have been routinely added to weaned piglet diets to improve health, prevent disease, and enhance production performance. However, growing concerns regarding the negative consequences of antibiotic use in animal production have emerged in recent years [3], necessitating the urgent search for effective alternatives. Medicinal plant polysaccharides have garnered considerable attention among animal nutritionists as potential immunomodulators and metabolic regulators for livestock [4].

Medicinal plant polysaccharides are bioactive macromolecules extracted from natural medicinal plants, consisting of ten to tens of thousands of monosaccharides and monosaccharide derivatives linked by glycosidic bonds. Beyond serving as energy sources and structural components, these compounds actively regulate various life processes and exhibit diverse biological activities, including antiparasitic, antitumor, antibacterial, hypoglycemic, antioxidant, antiviral, antithrombotic, radioprotective, anti-aging, lipid-lowering, and immunomodulatory effects [5-6]. Numerous plant polysaccharides have been isolated and preliminary feeding trials have demonstrated their growth-promoting and immune-regulating potential in animals. Zhao et al. [7] reported that Atractylodes macrocephala polysaccharide increased microbial diversity and improved intestinal environment in weaned piglets. Zhen et al. [8] observed that dietary supplementation with 200 mg/kg Astragalus polysaccharides enriched cecal microbiota and improved feed efficiency. Ren [9] demonstrated that Achyranthes bidentata polysaccharides significantly affected E. coli and Lactobacillus populations across various intestinal segments.

The biological activity of medicinal plant polysaccharides is substantially influenced by glycosidic bond types, with certain glycosidic linkages conferring physiological activity. Additionally, polysaccharide structure, molecular weight, solubility, and viscosity all modulate their functional properties [10]. Astragalus polysaccharides are high-molecular-weight compounds extracted from Astragalus membranaceus, primarily composed of glucose, arabinose, and galactose. Atractylodes macrocephala polysaccharides are extracted from the Asteraceae plant Atractylodes macrocephala, consisting mainly of fructans and mannans. Achyranthes bidentata polysaccharides have relatively low molecular weight and are predominantly composed of glucose and fructose. While previous research has primarily focused on single polysaccharide supplementation, comparative studies among individual polysaccharides and investigations of potential synergistic effects from combined usage on weaned piglet intestinal environment remain scarce. This study examined the individual and combined effects of APS, AMP, and ABP on growth performance and intestinal environment of weaned piglets, aiming to provide theoretical and practical guidance for formulating high-quality piglet diets.

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1. Materials and Methods

1.1 Experimental Materials

Astragalus polysaccharides, Atractylodes macrocephala polysaccharides, and Achyranthes bidentata polysaccharides were extracted and prepared according to the method described by Chen et al. [11]. The polysaccharide contents were determined using the anthrone-sulfuric acid method, yielding purity levels of 65%, 60%, and 60% for APS, AMP, and ABP, respectively.

1.2 Experimental Animals and Diets

Two hundred fifty-six 35-day-old Duroc × Landrace × Yorkshire crossbred weaned piglets were selected for the trial. A corn-soybean meal basal diet was formulated according to NRC (2012) standards. The composition and nutrient levels of the basal diet are presented in Table 1 .

Table 1 Composition and nutrient levels of the basal diet (air-dry basis)

Note: The premix provided per kilogram of diet: VA 12,276 IU, VD3 1,228 IU, VE 11 IU, VK3 2.2 mg, VB1 1.3 mg, VB2 3.1 mg, VB6 1.2 mg, VB12 23 g, nicotinic acid 23 mg, pantothenic acid 13.4 mg, biotin 0.11 mg, folic acid 0.68 mg, Cu 25 mg, Fe 156 mg, Zn 100 mg, Mn 54 mg, Se 0.3 mg, I 0.5 mg. Nutrient levels were calculated values.

1.3 Experimental Design and Management

Piglets were randomly assigned to 8 groups following the principle of similar initial body weight and equal sex distribution (1:1 ratio), with 4 replicates per group and 8 piglets per replicate. A three-factor two-level (2$×2×$2) factorial arrangement was employed. Group 1 served as the control group receiving the basal diet, while treatment groups received the basal diet supplemented with various combinations of APS, AMP, and ABP as shown in Table 2 . The feeding trial was conducted at the Yiyang Agricultural Science Research Institute Pig Farm in Hunan Province for 28 days. Piglets were housed on slatted floors and fed four times daily at 06:00, 10:00, 14:00, and 18:00 with ad libitum access to water. Routine vaccination protocols were followed according to standard farm management practices.

Table 2 Experimental design (mg/kg)

1.4 Measurements

1.4.1 Growth Performance Piglets were weighed at the beginning and end of the experiment. Daily feed intake and diarrhea incidence were recorded throughout the trial. Diarrhea was defined as unformed feces with high moisture content, while normal feces were characterized by formed consistency with low moisture. Average daily gain (ADG), feed-to-gain ratio (F/G), and diarrhea rate were calculated accordingly.

1.4.2 Intestinal Content pH Determination At the conclusion of the trial, one piglet per replicate (4 piglets per group) was randomly selected for slaughter. The abdominal cavity was opened, and the ileum, cecum, and colon were ligated. Intestinal contents were collected from each segment, mixed with distilled water at a 1:5 mass-to-volume ratio, homogenized, and centrifuged at 3,500 r/min for 10 min at 4°C. The supernatant was collected and pH values were measured using a pH meter.

1.4.3 Volatile Fatty Acid (VFA) Content Analysis Immediately after opening the abdominal cavity, the ileum, cecum, and colon were ligated, and approximately 10 g of digesta from each segment was collected into sterile 10 mL centrifuge tubes. For cecal VFA analysis, 3 g of digesta was accurately weighed and diluted with an equal volume of distilled water, vortexed thoroughly, and allowed to stand for 30 min. The mixture was then centrifuged at 2,500 r/min for 10 min, and 2 mL of supernatant was collected. This supernatant was centrifuged again at 12,000 r/min for 10 min, and 1 mL of the resulting supernatant was mixed with 0.2 mL of 25% metaphosphoric acid, vortexed, and allowed to stand for 30 min. After centrifugation at 12,000 r/min for 10 min, 100 L of supernatant was mixed with 900 L methanol, vortexed, centrifuged at 12,000 r/min for 10 min, and the final supernatant was stored at -20°C for subsequent analysis. VFA concentrations were determined using gas chromatography.

1.4.4 Intestinal Microbial Enumeration In a sterile environment, 1.0 g of cecal and colonic digesta was accurately weighed and mixed with 9 mL sterile physiological saline to prepare a 1:10 dilution. Serial dilutions from 10^{-3} to 10^{-7} were prepared and plated for enumeration of E. coli, Bifidobacterium, and Lactobacillus populations. E. coli was cultured on MacConkey agar, Lactobacillus on MRS agar, and Bifidobacterium on selective Bifidobacterium medium. All microbial counts were determined using standard plate count methods.

1.5 Statistical Analysis

All data were analyzed using the General Linear Model (GLM) procedure of SAS 9.2 software. Results are expressed as means ± standard deviation. Significance was declared at $P<0.05$, and Duncan's multiple range test was used for post-hoc comparisons when significant effects were detected.

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2. Results

2.1 Effects of Botanical Polysaccharides on Growth Performance of Weaned Piglets

As shown in Table 3 , among individual polysaccharide supplementation groups, the APS group (Group 2) exhibited the highest ADG, though differences among groups were not statistically significant ($P>0.05$). Compared with the control group, the triple polysaccharide combination significantly increased ADG ($P<0.05$). Individual supplementation of APS, AMP, or ABP did not significantly affect average daily feed intake (ADFI) ($P>0.05$). The F/G ratio in APS, AMP, and ABP groups showed a decreasing trend compared with the control group, but the differences were not significant ($P>0.05$). Similarly, all polysaccharide combination groups numerically reduced the F/G ratio without achieving statistical significance ($P>0.05$). Individual supplementation of APS, AMP, or ABP significantly reduced diarrhea rate ($P<0.05$). Significant interactive effects on diarrhea rate were observed for the APS+AMP combination, AMP+ABP combination, and the triple combination ($P<0.05$).

Table 3 Effects of botanical polysaccharide on growth performance of weaned piglets

Note: In the same column, values with different letter superscripts differ significantly (P<0.05), while values with the same letter or no superscripts do not differ significantly (P>0.05). The same applies below.

2.2 Effects of Botanical Polysaccharides on Intestinal Content pH of Weaned Piglets

Table 4 shows that dietary polysaccharide supplementation tended to decrease pH values in ileal and rectal contents, but these changes were not significant ($P>0.05$). Both individual and pairwise supplementation of APS, AMP, and ABP significantly reduced cecal content pH ($P<0.05$), with no significant differences among various polysaccharide groups ($P>0.05$). The triple combination significantly lowered cecal pH compared with individual or pairwise combinations ($P<0.05$), demonstrating a highly significant interactive effect ($P<0.01$). In the colon, no significant differences in pH were observed among different polysaccharide combination groups ($P>0.05$); however, polysaccharide-supplemented groups (Groups 2 and 5-8) significantly decreased colonic content pH compared with the control group ($P<0.05$).

Table 4 Effects of botanical polysaccharide on pH of intestinal contents of weaned piglets

2.3 Effects of Botanical Polysaccharides on VFA Content in Cecal Digesta of Weaned Piglets

As presented in Table 5 , total VFA concentrations in cecal digesta were significantly higher in all polysaccharide-supplemented groups compared with the control ($P<0.05$). Except for Group 8, no significant differences were observed among various polysaccharide groups ($P>0.05$). Pairwise combinations showed a numerical increase in total VFA content compared with individual polysaccharides ($P>0.05$). The triple combination significantly elevated total VFA content compared with individual supplementation ($P<0.05$) and showed a numerical increase over pairwise combinations without statistical significance ($P>0.05$). Individual supplementation of APS, AMP, or ABP significantly increased acetic and propionic acid concentrations ($P<0.05$), with no significant differences among these groups ($P>0.05$). Pairwise combinations did not significantly differ from individual supplementation in terms of acetic and propionic acid content ($P>0.05$), whereas the triple combination significantly outperformed individual polysaccharides ($P<0.05$). Individual, pairwise, or triple supplementation of APS, AMP, and ABP significantly increased butyric acid concentration ($P<0.05$), with the triple combination showing significantly higher butyric acid levels than individual supplementation ($P<0.05$).

Table 5 Effects of botanical polysaccharide on VFA content in cecum content of weaned piglets (n=4) (mg/mL)

2.4 Effects of Botanical Polysaccharides on Intestinal Microbial Populations of Weaned Piglets

Table 6 demonstrates that individual supplementation of APS, AMP, or ABP significantly reduced E. coli populations in both cecum and colon ($P<0.05$), with no significant differences among the three groups (P>0.05). Pairwise combinations numerically decreased E. coli counts compared with individual supplementation, but the differences were not significant (P>0.05). The triple combination significantly reduced E. coli populations compared with pairwise combinations (P<0.05) and exhibited a significant interactive effect (P<0.05). For intestinal Lactobacillus counts, individual supplementation of APS, AMP, or ABP significantly increased populations compared with the control group (P<0.05), showing a pattern where pairwise combinations outperformed individual supplementation and the triple combination outperformed pairwise combinations, though these differences were not statistically significant (P>0.05). Individual supplementation of AMP and ABP significantly increased cecal Bifidobacterium counts (P<0.05), while APS showed a numerical increase without statistical significance (P>0.05). Pairwise combinations numerically enhanced cecal Bifidobacterium counts compared with individual supplementation (P>0.05). The triple combination significantly increased cecal Bifidobacterium counts compared with individual supplementation (P<0.05), but did not differ significantly from pairwise combinations (P>0.05). All three polysaccharides numerically increased colonic Bifidobacterium counts (P>0.05), with the triple combination significantly outperforming individual supplementation (P<0.05).

Table 6 Effects of botanical polysaccharide on intestinal microorganism number of weaned piglets (lg CFU/g)

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3. Discussion

3.1 Effects of Botanical Polysaccharides on Growth Performance of Weaned Piglets

Botanical polysaccharides exhibit diverse biological activities, including antimicrobial, antiviral, antioxidant, anti-inflammatory, anti-stress, and lipid-lowering properties [5-6]. Previous research has demonstrated that plant polysaccharides can function as growth promoters when incorporated into animal diets. Yuan et al. [12] reported that different doses of Astragalus polysaccharides significantly improved growth performance in weaned piglets by enhancing immune function. Yin et al. [13] found that dietary APS supplementation improved growth performance by promoting amino acid digestion, absorption, and metabolism. Mao et al. [14] observed that 500 mg/kg APS alleviated lipopolysaccharide-induced immune suppression and prevented declines in ADFI and ADG. Chen et al. [15] demonstrated that 1,000 mg/kg ABP significantly improved ADG and feed conversion ratio in piglets from 28 to 60 days of age. Yuan [16] reported that 500 mg/kg APS significantly increased ADG in weaned piglets. The current study showed that individual polysaccharide supplementation tended to increase ADG compared with the control group, though the difference was not significant, which may be attributed to dosage differences. Notably, the triple polysaccharide combination significantly increased ADG. Individual supplementation of

APS, AMP, or ABP significantly reduced diarrhea rate, with cumulative effects observed when polysaccharides were combined, the triple combination showing optimal efficacy. The varying interactive effects among the three polysaccharides on diarrhea reduction may be related to the complexity of polysaccharide structures, animal physiological characteristics, and the relatively simple supplementation approach, warranting further investigation.

3.2 Effects of Botanical Polysaccharides on Intestinal Content pH and VFA Concentrations

The hindgut represents the most metabolically active region for microbial fermentation in pigs, where intestinal microorganisms ferment carbohydrates to produce various VFAs, including lactic, formic, acetic, and butyric acids, consequently reducing intestinal pH [17]. Zhang et al. [18] investigated the effects of four yam polysaccharides on cecal fermentation products in Ningxiang pigs, demonstrating that yam polysaccharides produced substantial short-chain fatty acids during in vitro fermentation, decreased intestinal pH, and altered microbial populations. The present study showed that individual supplementation of APS, AMP, or ABP reduced pH in cecal and colonic contents, with pairwise combinations significantly decreasing pH in these segments. Polysaccharide supplementation significantly increased cecal VFA content, with combinations of two or more polysaccharides showing superior effects compared with individual supplementation. As large macromolecular compounds lacking corresponding digestive enzymes in the foregut, plant polysaccharides serve as substrates for hindgut microbial fermentation, generating substantial VFA production and consequently reducing pH in the lower gastrointestinal tract, thereby modulating intestinal environment.

3.3 Effects of Botanical Polysaccharides on Intestinal Microbial Populations of Weaned Piglets

Weaned piglets possess underdeveloped digestive enzyme systems, immature regulatory mechanisms, high metabolic rates, and incomplete immune system development, making them highly susceptible to stress-induced reductions in Lactobacillus and Bifidobacterium populations and concurrent increases in E. coli and Salmonella [19]. This microbial imbalance can transform E. coli into potential pathogens [20], with post-weaning diarrhea primarily caused by E. coli [21]. Weaning disrupts the established microbial ecosystem, characterized by increased E. coli populations and subsequent diarrhea [22]. Modern research indicates that plant polysaccharides actively modulate intestinal microecological balance and exhibit prebiotic activity. These polysaccharides are fermented by hindgut microorganisms, selectively stimulating beneficial bacteria growth while inhibiting harmful bacteria, thereby reducing diarrhea incidence [23]. Cao et al. [24] demonstrated through in vitro experiments that Polygonatum polysaccharides significantly inhibited E. coli and Staphylococcus aureus proliferation. Lactobacillus and Bifidobacterium, as resident intestinal bacteria, regulate microecological balance, participate in nutrient metabolism, and establish immune defense systems, playing crucial roles in disease prevention. These beneficial bacteria produce various organic acids through carbohydrate fermentation, enhancing digestive function, promoting nutrient absorption, and competitively inhibiting pathogen proliferation [25]. Tang et al. [26] reported that oligosaccharides from saccharicterpenin increased beneficial Bifidobacterium and Lactobacillus populations while inhibiting harmful microorganisms in weaned piglets. Li and Zhao [27] demonstrated that Astragalus polysaccharides promoted beneficial microbial colonization and significantly increased Lactobacillus and Bifidobacterium counts while reducing E. coli in chicks. Xu et al. [28] found that burdock polysaccharides significantly increased Lactobacillus and Bifidobacterium populations in mice without significantly affecting E. coli. Bäckhed et al. [29] reported that probiotics such as Lactobacillus and Bifidobacterium could utilize undegraded polysaccharides to inhibit pathogen proliferation. Sun et al. [30] demonstrated that fermented Yupingfeng polysaccharides enhanced microbial diversity, increased Bifidobacterium and Lactobacillus counts, and significantly reduced E. coli populations in rabbits. The current study corroborates these findings, showing that individual supplementation of APS, AMP, or ABP significantly reduced E. coli populations while increasing Lactobacillus and Bifidobacterium counts in the cecum and colon, with the triple combination showing optimal effects. Plant polysaccharides can be utilized by Lactobacillus and Bifidobacterium, promoting proliferation of beneficial bacteria. Furthermore, the substantial VFA production by these bacteria lowers intestinal pH, inhibiting acid-intolerant pathogens such as E. coli and Salmonella, thereby rapidly restoring microbial balance [25]. Additionally, plant polysaccharides may exert antibacterial effects through immune system modulation [31], while polysaccharide components compete with pathogenic E. coli for intestinal binding sites, inhibiting pathogen colonization and promoting beneficial Bifidobacterium growth [32].

4. Conclusion

Dietary supplementation with Astragalus polysaccharides, Atractylodes macrocephala polysaccharides, and Achyranthes bidentata polysaccharides individually or in combination reduced E. coli populations, increased beneficial bacteria counts, improved intestinal microecological balance, decreased pH across intestinal segments, elevated short-chain fatty acid concentrations in the cecum and colon, reduced diarrhea incidence, and enhanced intestinal environment, consequently improving growth performance of weaned piglets. The triple combination demonstrated superior efficacy compared with individual supplementation.

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