Effects of Attapulgite Nano-Zinc Oxide on Growth Performance, Organ Indices, and Blood Biochemical Parameters in Weaned Piglets (Postprint)

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Abstract

This experiment aimed to investigate the effects of dietary supplementation with attapulgite-loaded nano zinc oxide (attapulgite nano zinc oxide) on growth performance, organ indices, and blood biochemical parameters in weaned piglets. The experiment selected 210 healthy Duroc × Landrace × Large White weaned piglets at 21 days of age with similar body weight [(6.30±0.51) kg], which were randomly divided into 7 groups with 6 replicates per group and 5 piglets per replicate. The control group (CON group) was fed the basal diet, the antibiotic group (ANT group) was fed the basal diet + 100 g/kg 50% olaquindox + 150 g/kg 15% chlortetracycline + 50 g/kg 10% colistin sulfate, the zinc oxide group (ZO group) was fed the basal diet + 3,000 mg/kg zinc oxide, the nano zinc oxide group (NZO group) was fed the basal diet + 800 mg/kg nano zinc oxide, the low attapulgite nano zinc oxide group (LA-ZO group) was fed the basal diet + 700 mg/kg attapulgite nano zinc oxide, the medium attapulgite nano zinc oxide group (MA-ZO group) was fed the basal diet + 1,000 mg/kg attapulgite nano zinc oxide, and the high attapulgite nano zinc oxide group (HA-ZO group) was fed the basal diet + 1,300 mg/kg attapulgite nano zinc oxide. The experiment consisted of a 3-day pre-trial period and a 9-day formal trial period. The results showed: 1) Compared with the CON group, dietary supplementation with attapulgite nano zinc oxide significantly increased the average daily feed intake and average daily gain of weaned piglets ($P<0.05$). 2) The diarrhea rate in the LA-ZO group was significantly lower compared with the CON and ANT groups ($P<0.05$), and the diarrhea index in the LA-ZO group was significantly lower compared with the CON, ANT, and NZO groups ($P<0.05$). 3) The pancreas index in the LA-ZO and MA-ZO groups was significantly higher compared with the NZO group ($P<0.05$). 4) Compared with the CON group,

the MA-ZO group showed significantly decreased blood total cholesterol (TC) and triglyceride (TG) contents (P<0.05), and significantly increased blood high-density lipoprotein (HDL) content (P<0.05), which was also significantly higher than that in the ANT and NZO groups (P<0.05); the HA-ZO group had significantly higher blood HDL content than the CON and ANT groups (P<0.05). The results demonstrated that dietary supplementation with attapulgite nano zinc oxide in weaned piglets could improve growth performance, reduce diarrhea rate, and enhance piglet growth and development, while also decreasing blood TG and TC contents, increasing blood HDL content, strengthening lipid metabolism and pancreatic organ development, and could replace the use of antibiotics and high-level zinc.

Full Text

Effects of Attapulgite Nano Zinc Oxide on Growth Performance, Organ Indices and Blood Biochemical Indices of Weaned Piglets

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Abstract: This experiment was conducted to investigate the effects of dietary attapulgite nano zinc oxide on growth performance, organ indices, and blood biochemical indices of weaned piglets. A total of 210 healthy Duroc × Landrace × Yorkshire piglets at 21 days of age with similar body weight [(6.30±0.51) kg] were randomly allocated into 7 groups with 6 replicates per group and 5 piglets per replicate. The control group (CON) was fed a basal diet, the antibiotic group (ANT) received the basal diet supplemented with 100 g/kg 50% olaquindox, 150 g/kg 15% chlortetracycline, and 50 g/kg 10% colistin sulfate, the zinc oxide group (ZO) received the basal diet plus 3,000 mg/kg zinc oxide, the nano zinc oxide group (NZO) received the basal diet plus 800 mg/kg nano zinc oxide, and the low, medium, and high attapulgite nano zinc oxide groups (LA-ZO, MA-ZO, and HA-ZO) received the basal diet supplemented with 700, 1,000, and 1,300 mg/kg attapulgite nano zinc oxide, respectively. The experiment lasted for 9 days following a 3-day adaptation period. The results showed that: 1) Compared with the CON group, dietary attapulgite nano zinc oxide significantly increased the average daily feed intake and average daily gain of weaned piglets (P<0.05). 2) The diarrhea rate in the LA-ZO group was significantly lower than that in the CON and ANT groups (P<0.05), and the diarrhea index in the LA-ZO group was significantly lower than that in the CON, ANT, and NZO groups (P<0.05). 3) The pancreas index in the LA-ZO and MA-ZO groups was significantly higher than that in the NZO group (P<0.05). 4) Compared with the CON group, the MA-ZO group exhibited significantly decreased blood total cholesterol (TC)

and triglyceride (TG) contents (P<0.05) and significantly increased blood high-density lipoprotein (HDL) content (P<0.05), which was also significantly higher than that in the ANT and NZO groups (P<0.05). The HDL content in the HA-ZO group was significantly higher than that in the CON and ANT groups (P<0.05). These results indicate that dietary attapulgite nano zinc oxide can improve growth performance, reduce diarrhea rate, and promote growth and development in weaned piglets, while decreasing blood TG and TC contents, increasing blood HDL content, enhancing lipid metabolism and pancreatic organ development, thus demonstrating potential to replace antibiotics and high-dose zinc supplementation.

Keywords: attapulgite nano zinc oxide; weaned piglets; growth performance; organ index; blood biochemical index

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Piglets experience incomplete intestinal development and low immune function at weaning, which can trigger weaning stress syndrome, leading to intestinal flora disorder and diarrhea that severely impairs their growth and development [1]. Antibiotics and high-dose zinc have been widely used in production practice to effectively alleviate these adverse effects of weaning stress [2]. However, researchers have identified numerous drawbacks to their use, including antibiotic residues and bacterial resistance, as well as the excretion of large amounts of zinc in feces due to low digestibility, causing resource waste and environmental pollution [3]. Consequently, the European Union, the Netherlands, the United States, and other countries have already restricted zinc supplementation levels in piglet feed. Therefore, exploring rational zinc utilization methods and actively seeking antibiotic alternatives to effectively alleviate weaning stress in piglets is imperative.

Among known approaches, nano zinc oxide represents a potentially feasible alternative to conventional zinc oxide, demonstrating more pronounced effects than traditional zinc oxide [4]. Nano zinc oxide features small particle size, large specific surface area, and high absorption rate, with particle diameters of 1–100 nm, and can regulate animal immunity and reproductive performance [5]. Studies have confirmed that dietary supplementation with 200–1,000 mg/kg nano zinc oxide exerts varying degrees of growth-promoting effects in livestock and poultry, improving intestinal mucosal morphology, reducing diarrhea rates, stimulating cellular, humoral, and non-specific immune functions, and enhancing disease resistance [6-8]. Zhao et al. [9] found that feeding broilers 60 mg/kg nano zinc oxide increased later-stage daily weight gain. Long et al. [10] reported that adding 500 mg/kg nano zinc oxide to weaned piglet diets significantly increased daily weight gain and feed intake while reducing diarrhea rates, with effects superior to 3,000 mg/kg zinc oxide. However, high doses of nano zinc oxide can have toxic side effects [11].

Attapulgite, also known as palygorskite, is a clay mineral primarily composed of silicate [theoretical chemical formula: Si Mg O (OH) · 4H O] that is non-

toxic, tasteless, non-irritating, and inexpensive. It exhibits porous surfaces, large specific surface area, and cation exchange properties, with good adsorption capacity for lead ions (Pb^2), copper ions (Cu^2), and antibiotics [12-15]. Furthermore, research indicates that clay minerals serve as controlled-release carriers for bioactive molecules, drugs, and nutrients [16-18]. Since 2011, attapulgite has been listed as a feed additive [19]. Studies have found that adding 2,000 mg/kg attapulgite to weaned piglet diets significantly increased average daily feed intake, improved feed conversion ratio, and effectively enhanced intestinal health [20]. Tang et al. [21] discovered that dietary supplementation with 1,800 mg/kg attapulgite significantly increased average daily gain, reduced diarrhea, promoted piglet growth, and improved economic benefits. However, numerous studies have shown that excessive attapulgite supplementation can negatively affect piglet growth and development [21-23].

Previous research has focused on high-dose attapulgite supplementation, with no reported effects of low-dose attapulgite in piglets. Meanwhile, the efficacy of 500–1,000 mg/kg nano zinc oxide supplementation in piglet diets remains controversial. Therefore, based on previous findings and considering production costs, this study combined low-dose attapulgite with varying levels of nano zinc oxide particles to form attapulgite nano zinc oxide. By supplementing weaned piglet diets with this compound, we explored the appropriate supplementation level of this low-dose attapulgite and nano zinc oxide combination during the early feeding stage of weaned piglets and its potential to replace antibiotics and high-dose zinc as a new green and healthy additive.

1 Materials and Methods

1.1 Experimental Materials

The zinc oxide (white powder), nano zinc oxide (white powder, purity 99%, average particle size 45 nm), and attapulgite nano zinc oxide (purity 85%, composed of 80% nano zinc oxide and 20% attapulgite, with the same physicochemical properties of nano zinc oxide as described above, and attapulgite primarily composed of silicate with a double-chain structure) used in this experiment were provided by the College of Chemical Engineering at Yangzhou University. The 50% olaquindox, 15% chlortetracycline, and 10% colistin sulfate were provided by the Yangzhou University Feed Mill. The experimental piglets were 21-day-old Duroc × Landrace × Yorkshire weaned piglets provided by Suzhou Taicang Jinzhu Agricultural Development Co., Ltd.

The colistin sulfate used in this experiment was produced in September 2016. According to the Ministry of Agriculture Announcement No. 2428, its use was permitted until April 30, 2017, and the experimental period complied with this standard. All materials were used exclusively for experimental research aimed at replacing antibiotic usage.

1.2 Experimental Design

A total of 210 healthy Duroc × Landrace × Yorkshire weaned piglets at 21 days of age with similar body weight [(6.30±0.51) kg] were randomly divided into 7 groups: control group (CON, fed basal diet), antibiotic group (ANT, fed basal diet + 100 mg/kg 50% olaquindox + 150 mg/kg 15% chlortetracycline + 50 mg/kg 10% colistin sulfate), zinc oxide group (ZO, fed basal diet + 3,000 mg/kg zinc oxide), nano zinc oxide group (NZO, fed basal diet + 800 mg/kg nano zinc oxide), low attapulgite nano zinc oxide group (LA-ZO, fed basal diet + 700 mg/kg attapulgite nano zinc oxide), medium attapulgite nano zinc oxide group (MA-ZO, fed basal diet + 1,000 mg/kg attapulgite nano zinc oxide), and high attapulgite nano zinc oxide group (HA-ZO, fed basal diet + 1,300 mg/kg attapulgite nano zinc oxide). Each group had 6 replicates with 5 piglets per replicate. The experiment was conducted at Taicang Jinzhu Pig Farm in Jiangsu Province in April 2017, with a 3-day adaptation period followed by a 9-day formal experimental period.

1.3 Animal Management and Experimental Diets

The pig farm was a large-scale production facility with complete equipment and comprehensive epidemic prevention and management measures. All experimental piglets were housed in the same building and fed powdered diets ad libitum (four times daily at 06:30, 10:30, 14:30, and 18:30, with each feeding amount adjusted to ensure slight remaining feed in the trough). Fresh water was available at all times. Pens were cleaned daily to maintain hygiene, with natural ventilation. Routine management, disinfection, and epidemic prevention followed standard farm procedures. The basal diet was formulated according to NRC (2012) nutrient requirements for piglets combined with production practice, and its composition and nutrient levels are shown in Table 1 .

1.4 Measurements

1.4.1 Growth Performance Feed intake was recorded by replicate throughout the experiment. All piglets were weighed after overnight fasting at 08:00 on day 1 and the final day of the experiment to calculate average daily gain (ADG), average daily feed intake (ADFI), and feed-to-gain ratio (F/G).

1.4.2 Diarrhea Rate and Diarrhea Index Feces were observed daily at 07:00 during the experimental period and scored according to Castillo et al. [24]: 0 points for strip-shaped or granular feces; 1 point for soft but formed feces; 2 points for thick, unformed feces without water separation; and 3 points for liquid, unformed feces with water separation. Diarrhea was defined as a fecal score of 2 or above. Diarrhea rate and diarrhea index were calculated using the following formulas:

Diarrhea rate (%) = 100 × (total diarrhea incidents per group) / (experimental days × number of piglets per group)

Diarrhea index = 100 × (total diarrhea scores per group) / (number of piglets per group)

1.4.3 Organ Indices At the end of the experiment, piglets were slaughtered after blood collection. The heart, liver, spleen, lungs, kidneys, thymus, and pancreas were collected, excess surface blood was removed with absorbent paper, and organs were immediately weighed to calculate organ indices.

Organ index (g/kg) = organ wet weight (g) / live body weight (kg)

1.4.4 Blood Biochemical Indices At 08:00 on the final experimental day, one piglet with body weight close to the replicate average was randomly selected from each replicate, and 3 mL of blood was collected from the anterior vena cava into anticoagulant tubes. Blood alkaline phosphatase (ALP) and lactate dehydrogenase (LDH) activities, as well as total protein (TP), albumin (ALB), globulin (GLB), triglyceride (TG), total cholesterol (TC), high-density lipoprotein (HDL), and low-density lipoprotein (LDL) contents were measured, and the albumin-to-globulin ratio (A/G) was calculated.

1.5 Statistical Analysis

Experimental data were initially organized using Excel 2010 and then subjected to one-way ANOVA using SPSS 20.0 software. Duncan's multiple range test was used for pairwise comparisons among groups, with $P<0.05$ indicating significant differences.

2 Results

2.1 Effects of Attapulgite Nano Zinc Oxide on Growth Performance of Weaned Piglets

As shown in Table 2 , dietary supplementation with antibiotics, zinc oxide, nano zinc oxide, and attapulgite nano zinc oxide had no significant effects on final body weight or feed-to-gain ratio of weaned piglets ($P>0.05$). Compared with the CON group, dietary attapulgite nano zinc oxide significantly increased ADFI and ADG of weaned piglets ($P<0.05$). Additionally, ADFI in the HA-ZO group was 34.78% higher than that in the ANT group ($P<0.05$).

2.2 Effects of Attapulgite Nano Zinc Oxide on Diarrhea of Weaned Piglets

As shown in Table 3 , the diarrhea rate in the LA-ZO and HA-ZO groups was significantly lower than that in the CON group ($P<0.05$), with the LA-ZO group showing significantly lower diarrhea rate than the ANT group ($P<0.05$). The diarrhea index in all attapulgite nano zinc oxide groups was significantly reduced compared with the CON group ($P<0.05$), and the LA-ZO group exhibited significantly lower diarrhea index than the ANT and NZO groups ($P<0.05$).

2.3 Effects of Attapulgite Nano Zinc Oxide on Organ Indices of Weaned Piglets

As shown in Table 4 , dietary supplementation with different levels of attapulgite nano zinc oxide had no significant effects on heart, liver, spleen, lung, kidney, or thymus indices of weaned piglets compared with the CON group ($P>0.05$). Heart index tended to increase with attapulgite nano zinc oxide supplementation compared with the ANT and NZO groups ($P>0.05$). The pancreas index in the LA-ZO and MA-ZO groups was significantly higher than that in the NZO group ($P<0.05$).

2.4 Effects of Attapulgite Nano Zinc Oxide on Blood Biochemical Indices of Weaned Piglets

As shown in Table 5 , dietary supplementation with different levels of attapulgite nano zinc oxide had no significant effects on blood ALP and LDH activities or TP, ALB, GLB, and LDL contents, nor on A/G ratio of weaned piglets ($P>0.05$). The TG content in the LA-ZO and MA-ZO groups was significantly lower than that in the CON group ($P<0.05$). The TC content in the MA-ZO group was reduced by 23.04% compared with the CON group ($P<0.05$) and by 25.94% compared with the ZO group ($P<0.05$). The HDL content in the LA-ZO group showed no significant difference compared with the CON, ANT, and NZO groups ($P>0.05$) but was significantly lower than that in the ZO group ($P<0.05$). The HDL content in the MA-ZO group was increased by 54.55% compared with the CON and ANT groups ($P<0.05$), while the HA-ZO group showed 16.36% higher HDL content than the CON and ANT groups ($P<0.05$).

3 Discussion

3.1 Effects of Attapulgite Nano Zinc Oxide on Growth Performance of Weaned Piglets

Early weaning in piglets leads to incomplete intestinal development, low nutrient digestion and absorption efficiency, and high diarrhea frequency, which severely affects their growth and development. Feed intake and feed conversion ratio are the primary factors determining growth rate in weaned piglets [25-26]. Studies have shown that attapulgite and nano zinc oxide can effectively alleviate weaning stress and improve piglet growth performance. Wang et al. [27] reported that adding 1,200 mg/kg nano zinc oxide to weaned piglet diets improved intestinal barrier function and protected the intestine compared with a colistin sulfate-zinc oxide mixture, thereby reducing diarrhea rates. They also found lower zinc content in piglet tissues and excreta, indicating reduced zinc accumulation and improved zinc absorption and utilization, thus achieving the effects of high-dose conventional zinc oxide with smaller amounts of nano zinc oxide. Long et al. [10] demonstrated that dietary supplementation with 500 mg/kg nano zinc oxide significantly increased ADG, ADFI, and feed conversion ratio in weaned piglets, with effects comparable to 3,000 mg/kg zinc oxide. Meng [28] found that adding

3,000 mg/kg attapulgite to piglet diets during days 1-21 and 22-42 increased intestinal digestive enzyme activity, promoted carbohydrate conversion, and accelerated nutrient digestion, thereby improving ADG and ADFI. Our results suggest that reduced attapulgite usage can achieve similar outcomes, though the specific efficacy of low-dose attapulgite requires further investigation as our additive was not a single substance.

This study examined the application effects of attapulgite nano zinc oxide, a combination of attapulgite and nano zinc oxide particles. We found that compared with the CON group, attapulgite nano zinc oxide significantly increased ADG and ADFI in weaned piglets, with the LA-ZO and MA-ZO groups showing effects comparable to the ZO group. The LA-ZO group also outperformed the ANT group in improving growth performance and numerically surpassed the NZO group, while achieving the lowest F/G ratio. These findings indicate that reduced zinc and attapulgite usage can maintain similar effects, though feed conversion ratio gradually decreased with increasing attapulgite nano zinc oxide doses, differing from previous studies. Research has shown that attapulgite possesses viscosity, rheological properties, and ion exchange capacity, forming colloidal films that increase intestinal retention time and improve villus growth [18,20,28-29]. We hypothesize that attapulgite may form colloidal films covering the intestinal mucosal surface, reducing mucosal damage and contact with harmful substances while ensuring mucosal integrity. As attapulgite maintains close contact with the mucosa, nano zinc oxide particles carried by attapulgite are gradually absorbed by the intestine. Additionally, ions carried by attapulgite may undergo exchange reactions with ions in the digestive tract, and the exchanged ions may be absorbed by the intestine to promote the release of various digestive enzymes, accelerating feed conversion and absorption, thereby improving weaned piglet growth performance. However, as attapulgite nano zinc oxide dosage increases, excessive coverage of the intestinal mucosal surface may hinder nutrient digestion and absorption, reducing feed conversion ratio, though not significantly. Furthermore, nano zinc oxide can increase the proliferation rate of Lgr5 and Bmil stem cells in the ileum, and accelerated proliferation of these cells promotes intestinal epithelial cell renewal and improves intestinal nutrient absorption and utilization [30-31]. Therefore, the combined action of attapulgite and nano zinc oxide on the intestine promotes piglet growth and development.

3.2 Effects of Attapulgite Nano Zinc Oxide on Diarrhea in Weaned Piglets

After weaning, piglets have incomplete defense systems and poor resistance to adverse environmental conditions, making them susceptible to stress-induced intestinal flora disorder. This reduces the inhibition of dominant flora on harmful bacteria such as Escherichia coli, allowing their extensive colonization and resulting in diarrhea [32]. Studies have found that attapulgite effectively treats diarrhea in humans and ruminants [33-35], though its application in piglets is

limited. Nano zinc oxide has been widely used in weaned piglets with good efficacy in effectively reducing diarrhea frequency [25-26]. Hu et al. [1] found that adding 500 and 750 mg/kg montmorillonite-zinc oxide mixture (containing 25% zinc) to weaned piglet diets significantly reduced diarrhea index in piglets aged 4-14 days, with 500 mg/kg montmorillonite-zinc oxide mixture showing effects comparable to 2,000 mg/kg zinc oxide. They suggested that 500 and 750 mg/kg montmorillonite-zinc oxide mixture could improve intestinal mucosal integrity and increase digestive enzyme activity in the pancreas and small intestine, accelerating nutrient breakdown and reducing continuous stimulation of solid diets on the intestine, thereby decreasing piglet diarrhea. Han [31] compared the effects of adding 2,000 mg/kg conventional zinc oxide and 500 mg/kg nano zinc oxide to weaned piglet diets and found that both groups significantly reduced diarrhea incidence with no difference between nano zinc oxide and conventional zinc oxide groups, primarily because nano zinc oxide significantly reduced intestinal permeability and increased villus height and villus-to-crypt ratio in the ileum and colon, improving the intestinal environment.

Our results indicate that dietary supplementation with three levels of attapulgite nano zinc oxide effectively reduced diarrhea rate and index in weaned piglets, with the 700 mg/kg dosage showing the lowest diarrhea rate and index, lower than the 3,000 mg/kg zinc oxide and 800 mg/kg nano zinc oxide groups, and significantly superior to the ANT group. This demonstrates that low-dose attapulgite nano zinc oxide has good anti-diarrheal effects. The 140 mg/kg attapulgite (in the 700 mg/kg attapulgite nano zinc oxide) may achieve the same anti-diarrheal effect as 200 mg/kg attapulgite due to its properties and combination with nano zinc oxide. The 1,000 mg/kg dosage showed a diarrhea index similar to 3,000 mg/kg zinc oxide, consistent with Hu et al. [1]. Therefore, dietary supplementation with 700 mg/kg attapulgite nano zinc oxide provides optimal anti-diarrheal effects.

The mechanism by which attapulgite nano zinc oxide reduces piglet diarrhea may involve nano zinc oxide carried by attapulgite increasing the expression of intestinal mucosal proteins Occludin and Claudin, which facilitate repair of the intestinal physical barrier. Nano zinc oxide can also kill Gram-negative bacteria such as E. coli and high-temperature/high-pressure resistant spores, assisting beneficial bacteria colonization and dominance [36]. Additionally, attapulgite' s adsorptive properties enable it to adsorb mycotoxins and protect the intestinal barrier. The synergistic action of both components maintains intestinal integrity and flora stability, reducing diarrhea [37]. During the experiment, we observed that the CON and ANT groups primarily exhibited mild diarrhea and soft feces, resulting in higher diarrhea rates. However, antibiotics promoted piglet growth and improved feed utilization, resulting in lower F/G than the CON group. High-dose zinc oxide can alleviate diarrhea but may impair nutrient absorption and conversion, thereby reducing feed conversion ratio.

3.3 Effects of Attapulgite Nano Zinc Oxide on Organ Indices of Weaned Piglets

Visceral organs are essential components of the animal body, maintaining normal growth and development and resisting environmental changes and pathogen invasion. Organ indices reflect the growth, development, metabolism, function, and health status of various organs, thereby indicating animal growth performance and physiological condition. The pancreas is an important component of the gastrointestinal tract, and its endogenous digestive enzyme activity affects nutrient absorption and utilization [38]. Good organ development is crucial for animals to cope with stress and can influence piglet growth and nutrient absorption. Our results suggest that compared with 800 mg/kg nano zinc oxide, dietary supplementation with 700 and 1,000 mg/kg attapulgite nano zinc oxide significantly increased pancreas weight, slightly higher than the CON group, but pancreas index decreased with increasing attapulgite nano zinc oxide dosage. Previous studies have found that high-dose nano zinc oxide exerts toxic effects on organ growth and affects mouse development [12]. We hypothesize that increased zinc supplementation may inhibit the activity and content of certain anti-stress-related enzymes in the pancreas or the differentiation of some stem cells, exacerbating pancreatic oxidative stress and negatively affecting organ growth, leading to impaired pancreatic development. Attapulgite may mitigate this damage to some extent, though the specific mechanism requires further investigation. Heart index tended to increase with attapulgite nano zinc oxide supplementation and was higher than in the ANT group, but showed no significant difference from the CON group, indicating that attapulgite nano zinc oxide had no significant effect on heart development or other organ growth in weaned piglets.

3.4 Effects of Attapulgite Nano Zinc Oxide on Blood Biochemical Indices of Weaned Piglets

Blood is an important component of the internal environment, and changes in blood biochemical indices can indirectly reflect alterations in organ tissues and metabolism. Good metabolism promotes nutrient absorption and enzyme release, maintaining nutritional balance, improving growth performance, and increasing production efficiency. TG, derived from fat decomposition or carbohydrate conversion, provides energy for the body. Serum TG content reflects fat decomposition and body fat changes; decreased TG content indicates enhanced fat decomposition or conversion to body fat, and appropriate TG levels benefit internal environment homeostasis [39]. TC is a lipid substance and a raw material for various steroid hormones, and its content reflects lipid absorption and metabolism. Meng [28] reported that adding 3,000 mg/kg attapulgite to weaned piglet diets had no significant effect on blood TG content. Xu et al. [41] found that adding 40 mg/kg nano zinc oxide to broiler diets significantly reduced serum TC content after 28 days of age. Our results in piglets supplemented with 1,000 mg/kg attapulgite nano zinc oxide showed similar serum

TC content to the latter study but differed from the former, possibly because attapulgite has no significant effect on blood TG and TC content, while nano zinc oxide can affect their production and conversion, appropriately reducing TG and TC content to promote fat decomposition and lipid conversion and absorption, improving energy utilization and growth performance.

HDL has important transport functions, transferring cholesterol to tissues such as the adrenal gland for metabolism and from peripheral tissues to the liver for excretion as bile acids, alleviating atherosclerosis [42]. Wang et al. [43] found that adding 150 and 300 mg/kg nano zinc oxide to diets increased serum HDL content in weaned piglets, with no significant difference from the high-zinc group, suggesting that novel nano zinc oxide could improve crude protein absorption and utilization, promoting piglet growth. Our results indicate that 1,000 mg/kg attapulgite nano zinc oxide supplementation significantly increased blood HDL content, significantly higher than in the ANT group but not significantly different from the ZO group, demonstrating that attapulgite nano zinc oxide can increase blood HDL synthesis rate, accelerate cholesterol conversion, enhance inter-tissue substance transport, and promote nutrient metabolism and absorption, thereby improving animal production performance.

4 Conclusion

Dietary attapulgite nano zinc oxide supplementation can significantly increase ADG and ADFI, reduce diarrhea rate, and improve growth performance in weaned piglets, though F/G gradually increases with supplementation level.

Dietary supplementation with 1,000 mg/kg attapulgite nano zinc oxide can significantly reduce blood TG and TC contents, significantly increase blood HDL content, promote fat decomposition, accelerate cholesterol and other lipid conversion, and enhance lipid metabolism.

Based on our results, 700 mg/kg attapulgite nano zinc oxide can replace antibiotics, 800 mg/kg nano zinc oxide, and 3,000 mg/kg zinc oxide in improving growth performance and reducing diarrhea rate in weaned piglets. Using low-dose zinc in weaned piglet diets can improve production efficiency. Therefore, we recommend an appropriate attapulgite nano zinc oxide supplementation level of 700 mg/kg.

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