

Effects of Compound Microencapsulated Organic Acids and Essential Oils on Growth Performance, Immune Organ Indices, Slaughter Performance, Meat Quality, and Serum Biochemical Parameters in Broiler Chickens (Postprint)

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Abstract

This experiment aimed to investigate the effects of microencapsulated organic acids and essential oils composite (MOAEO) on growth performance, immune organ indices, slaughter performance, meat quality, and serum biochemical indices of broiler chickens. A total of 300 one-day-old yellow-feathered broiler roosters were selected and randomly divided into 5 groups with 5 replicates per group and 12 birds per replicate. Group I was the control group fed the basal diet; Group II was the antibiotic group supplemented with 30 mg/kg bacitracin zinc and 6 mg/kg colistin sulfate in the basal diet; Groups III, IV, and V were experimental groups supplemented with 150, 200, and 250 mg/kg MOAEO in the basal diet, respectively. The experimental period lasted 70 days. The results showed that: 1) At 1-21 days of age, compared with the control group, Group II had significantly higher average daily gain ($P < 0.05$), Group IV had significantly lower feed conversion ratio ($P < 0.05$), Groups IV and V had significantly higher spleen index ($P < 0.05$), and Group V had significantly lower serum uric acid content ($P < 0.05$). 2) At 1-70 days of age, compared with the control group, Group IV had significantly lower feed conversion ratio ($P < 0.05$), Group IV had significantly higher spleen index ($P < 0.05$), Groups II and IV had significantly higher serum total protein content, Groups II and IV had significantly lower serum uric acid content ($P < 0.05$), Group IV had significantly higher breast muscle percentage and leg muscle percentage ($P < 0.05$), and Group IV had significantly lower breast muscle drip loss ($P < 0.05$). 3) Compared with the antibiotic group, dietary supplementation of MOAEO had no significant effects on growth performance, immune organ indices, slaughter performance, and serum biochemical indices of broiler chickens ($P > 0.05$). It can be

concluded that dietary supplementation of MOAEO in broiler chickens can improve growth performance, immune organ indices, slaughter performance, meat quality, and serum biochemical indices, and the appropriate supplementation level of MOAEO is 200 mg/kg.

Full Text

Effects of Microencapsulated Organic Acids and Essential Oils on Growth Performance, Immune Organ Indexes, Slaughter Performance, Meat Quality, and Serum Biochemical Indexes of Broilers

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Abstract

This experiment was conducted to investigate the effects of microencapsulated organic acids and essential oils (MOAEO) on growth performance, immune organ indexes, slaughter performance, meat quality, and serum biochemical indexes of broilers. A total of 300 one-day-old yellow-feathered male broilers were randomly divided into 5 groups with 5 replicates per group and 12 broilers per replicate. Broilers in group (control group) were fed a basal diet, broilers in group (antibiotics group) were fed the basal diet supplemented with 30 mg/kg bacitracin zinc and 6 mg/kg colistin sulfate, and those in groups , , and were fed the basal diet supplemented with 150, 200, and 250 mg/kg MOAEO, respectively. The experiment lasted for 70 days. The results showed as follows: 1) At 1 to 21 days of age, compared with the control group, the average daily gain in group was significantly increased ($P < 0.05$), the feed-to-gain ratio in group was significantly decreased ($P < 0.05$), the spleen index in groups and was significantly increased ($P < 0.05$), and the serum uric acid content in group was significantly decreased ($P < 0.05$). 2) At 1 to 70 days of age, compared with the control group, the feed-to-gain ratio in group was significantly decreased ($P < 0.05$), the spleen index in group was significantly increased ($P < 0.05$), the serum total protein content in groups and was significantly increased ($P < 0.05$), the serum uric acid content in groups and was significantly decreased ($P < 0.05$), the breast muscle rate and leg muscle rate in group were significantly increased ($P < 0.05$), and the water loss rate of breast muscle in group was significantly decreased ($P < 0.05$). 3) Compared with the antibiotics group, dietary MOAEO supplementation did not significantly affect the growth performance, immune organ indexes, slaughter performance, or serum biochemical indexes of broilers ($P > 0.05$). In conclusion, dietary supplementation of MOAEO can improve growth performance, immune organ indexes, slaughter performance, meat quality, and serum biochemical indexes of broilers,

with a suitable addition level of 200 mg/kg.

Key words: microencapsulation; essential oils; organic acids; broilers

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Introduction

Due to concerns that antibiotic supplementation in feed may lead to pathogen resistance and antibiotic residues in animal products, many countries have banned the use of antibiotics in feed. For example, the European Union banned antibiotic supplementation in feed in 2006. Therefore, finding effective antibiotic alternatives to promote animal growth and health is of great significance. Plant essential oils (EO) and organic acids (OA) have good antioxidant and antibacterial effects, can improve animal intestinal health, and promote nutrient absorption, making them commonly used antibiotic alternatives. Studies have shown that EO and OA can improve animal growth performance, reduce pathogen adhesion to the intestine, and improve intestinal morphology. However, the application of EO and OA as feed additives in production faces several problems: EO is prone to volatile oxidation, has unclear components, and has a stimulating effect; OA easily stimulates the digestive tract mucosa and cannot pass through the stomach, preventing EO and OA from exerting better efficacy. Microencapsulation technology can enable nutrients to be slowly released in the intestine while reducing irritating odors and eliminating effects on animal palatability, thus overcoming the defects of uncoated EO and OA. Currently, there are few reports in the literature on the use of microencapsulated EO and OA to improve broiler growth performance. Therefore, this experiment aimed to study the effects of different doses of microencapsulated organic acids and essential oils (MOAEO) on growth performance, immune organ indexes, slaughter performance, meat quality, and serum biochemical indexes of broilers, thereby providing a reference basis for the application of MOAEO in broiler production.

Materials and Methods

1.1 Test Materials

MOAEO was produced by Vetagro Company in Italy (trade name AVIplus P), with component contents of 25% citric acid, 16.7% sorbic acid, 1.7% thymol, 1.0% vanillin, and a carrier of fat (55.6%). Product quality standard number: SJK120058; production batch number: 8902011.

1.2 Experimental Design

A total of 300 healthy one-day-old yellow-feathered male broilers (purchased from Putian Wenshi Poultry Co., Ltd.) were randomly divided into 5 groups with 5 replicates per group and 12 broilers per replicate. Group 1 was the control group fed a basal diet; group 2 was the antibiotics group fed the basal diet supplemented with 30 mg/kg bacitracin zinc and 6 mg/kg colistin sulfate; and

groups , , and were experimental groups fed the basal diet supplemented with 150, 200, and 250 mg/kg MOAEO, respectively. The basal diet was formulated according to China's "Feeding Standard of Broilers" (NY/T 33–2004). The composition and nutrient levels of the basal diet are shown in Table 1 . Broilers were manually fed twice daily with free access to feed and water. Immunization and disinfection were carried out according to conventional broiler procedures. The experimental period lasted 70 days. On days 21 and 70 of the experiment, 2 broilers close to the average body weight were selected from each replicate for blood collection and slaughter sampling.

1.3 Measurements

1.3.1 Growth Performance The initial body weight of each broiler was recorded at the start of the experiment, and all broilers were weighed individually at 08:00 on days 21 and 70 after 12 hours of fasting to calculate the average daily gain for each stage. On a replicate basis, the daily feed amount, residual feed amount, and loss amount of each replicate were accurately weighed and recorded to calculate the average daily feed intake for each stage. The feed-to-gain ratio for each stage was then calculated based on average daily feed intake and average daily gain. Meanwhile, the number of dead broilers was recorded daily to calculate the survival rate for each stage.

1.3.2 Immune Organ Indexes On days 21 and 70, after slaughter, the spleen, thymus, and bursa of Fabricius were collected. Blood on each organ was absorbed with filter paper, fat was stripped, and the organs were weighed. The indexes were calculated using the following formulas: Spleen index (%) = (spleen weight/live weight) \times 100; Thymus index (%) = (thymus weight/live weight) \times 100; Bursa of Fabricius index (%) = (bursa of Fabricius weight/live weight) \times 100.

1.3.3 Slaughter Performance After slaughter on day 70, broilers were bled and slaughtered, feathers were removed by wet plucking, and carcass weight was weighed after draining water. The semi-eviscerated weight was obtained by removing the trachea, esophagus, crop, intestines, spleen, pancreas, and reproductive organs from the carcass weight while retaining the heart, liver (without gallbladder), lungs, kidneys, glandular stomach, gizzard (without contents and cuticle), and abdominal fat. The eviscerated weight was obtained by removing the heart, liver, glandular stomach, gizzard, abdominal fat, and the weight of head, neck, and feet from the semi-eviscerated weight. The abdominal fat, breast muscle, and leg muscle were collected and weighed. The following formulas were used: Slaughter rate (%) = (carcass weight/pre-slaughter weight) \times 100; Semi-eviscerated rate (%) = (semi-eviscerated weight/pre-slaughter weight) \times 100; Eviscerated rate (%) = (eviscerated weight/pre-slaughter weight) \times 100; Abdominal fat rate (%) = [abdominal fat weight/(eviscerated weight + abdominal fat weight)] \times 100; Breast muscle rate (%) = (both sides of breast muscle

weight/eviscerated weight) \times 100; Leg muscle rate (%) = (both sides of leg muscle weight/eviscerated weight) \times 100.

1.3.4 Serum Biochemical Indexes On days 21 and 70 at 08:00, after individual weighing, 5 mL of blood was collected from the wing vein using vacuum blood collection tubes. After tilting and standing to allow serum exudation, the samples were centrifuged at 3,000 r/min for 15 minutes. The supernatant was collected, aliquoted into EP tubes, and stored at -80 °C. Serum biochemical indexes were determined using kits from Nanjing Jiancheng Bioengineering Institute. The measured indexes included serum total protein, albumin, and uric acid content, which were determined using the Coomassie brilliant blue method, bromocresol green method, and colorimetric method, respectively.

1.3.5 Meat Quality Determination After slaughter on day 70, the pH, shear force, and water loss rate of the left breast muscle were determined. The pH was measured at three different points on the inner side of the breast muscle at 45 minutes (pH45 min) and 24 hours (pH24 h) according to the method specified in NY/T 1333-2007 “Determination of Livestock and Poultry Meat Quality.” Shear force was determined according to the method specified in NY/T 1180-2006 “Determination of Meat Tenderness—Shear Force Determination Method.” Water loss rate was determined according to the method specified in NY/T 1333-2007 “Determination of Livestock and Poultry Meat Quality.”

1.4 Data Analysis

SPSS 16.0 statistical software was used for one-way ANOVA, and multiple comparisons were performed using the LSD method. Experimental results were expressed as mean \pm SD, with $P < 0.05$ considered as a significant difference.

Results

2.1 Effects of MOAEO on Growth Performance of Broilers

As shown in Table 2, during the periods of 1-21 days, 22-70 days, and 1-70 days of age, there were no significant differences in average daily feed intake and survival rate among groups ($P > 0.05$). The average daily gain in group during 1-21 days of age was significantly higher than that in group ($P < 0.05$), while no significant differences were observed among groups in other stages ($P > 0.05$). The feed-to-gain ratio in group during 1-21 days and 1-70 days of age was significantly lower than that in group ($P < 0.05$), while no significant differences were observed among groups in other stages ($P > 0.05$).

2.2 Effects of MOAEO on Immune Organ Indexes of Broilers

As shown in Table 3, at 21 and 70 days of age, there were no significant differences in thymus index and bursa of Fabricius index among groups ($P > 0.05$).

The spleen index in group at both 21 and 70 days of age was significantly higher than that in group ($P < 0.05$), and the spleen index in group at 21 days of age was also significantly higher than that in group ($P < 0.05$).

2.3 Effects of MOAEO on Slaughter Performance and Breast Meat Quality of Broilers

As shown in Table 4, at 70 days of age, there were no significant differences in slaughter rate, semi-eviscerated rate, eviscerated rate, or abdominal fat rate among groups ($P > 0.05$). The breast muscle rate and leg muscle rate in group were significantly higher than those in group ($P < 0.05$).

As shown in Table 5, the pH45 min and pH24 h of breast muscle in group were significantly lower than those in group ($P < 0.05$). The water loss rate of breast muscle in group was significantly lower than that in group ($P < 0.05$). There were no significant differences in shear force among groups ($P > 0.05$).

2.4 Effects of MOAEO on Serum Biochemical Indexes of Broilers

As shown in Table 6, at 21 and 70 days of age, there were no significant differences in serum albumin content among groups ($P > 0.05$). The serum total protein content in groups and at 70 days of age was significantly higher than that in group ($P < 0.05$). The serum uric acid content in group at 21 days of age was significantly lower than that in group ($P < 0.05$), and the serum uric acid content in groups and at 70 days of age was significantly lower than that in group ($P < 0.05$).

Discussion

3.1 Effects of MOAEO on Growth Performance of Broilers

This study demonstrated that dietary supplementation with appropriate amounts of MOAEO can help reduce the feed-to-gain ratio and improve feed conversion efficiency in broilers. Some studies have also shown that combinations of EO and OA can improve average daily gain and feed conversion ratio in livestock and poultry. For example, dietary supplementation with thymol and benzoic acid in turkeys increased average daily gain and feed conversion ratio; dietary supplementation with formic acid, propionic acid, oregano oil, clove oil, and fennel oil in broilers improved average daily gain and feed conversion ratio without significantly affecting average daily feed intake; and dietary supplementation with citric acid, sorbic acid, thymol, and vanillin in weaned piglets increased average daily gain. For broiler chickens, the effects of EO and OA combinations (thymol, eugenol, benzoic acid) were better in male chicks than in female chicks, possibly related to the faster growth rate of male chicks. The improvement in feed conversion ratio may be related to EO and OA promoting nutrient absorption in the intestine. Microencapsulation can protect EO and OA from light, storage, feed processing, and various physicochemical

factors in the intestine, while also enabling slow and targeted release in the intestine. Therefore, microencapsulation can improve the stability of EO and OA, allowing them to function more persistently and enhancing their effects, thereby promoting broiler growth performance.

3.2 Effects of MOAEO on Immune Organ Indexes of Broilers

The main immune organs in poultry are the thymus, spleen, and bursa of Fabricius. The thymus and bursa of Fabricius are central immune organs, while the spleen participates in systemic cellular and humoral immunity and is the largest peripheral immune organ in poultry. Rivas et al. pointed out that the immune status of chicks can be evaluated by the weights of the thymus, spleen, and bursa of Fabricius. Immune organs are the producers and maintainers of immune function, and their development, maturation, and structural changes can enhance the overall immune function of broilers. In this experiment, dietary supplementation with MOAEO increased the spleen index of broilers. The increase in immune organ index indicates that MOAEO can stimulate the immune response and enhance immune function in broilers, thereby strengthening their overall immune function. Huang et al. reported that dietary supplementation with compound acidifiers (lactic acid, citric acid, and fumaric acid) in broilers significantly increased the spleen index of chicks. Walter et al. demonstrated that dietary supplementation with oregano EO in growing pigs improved the immune function of growing-finishing pigs.

3.3 Effects of MOAEO on Slaughter Performance and Breast Meat Quality of Broilers

Meat quality mainly includes the chemical composition, histological and histochemical characteristics, muscle physical properties, and sensory quality of meat. The pH, water-holding capacity, and tenderness of meat are its physicochemical properties. Muscle pH is positively correlated with muscle sourness; as muscle pH decreases, muscle sourness becomes stronger, affecting meat quality, and it is one of the most important indicators for meat quality determination. Water-holding capacity, typically measured by muscle water loss rate or drip loss, refers to the ability of muscle to retain water when subjected to external forces. Low water-holding capacity means water loss and reduced muscle weight, which can cause economic losses. Tenderness is an important indicator for evaluating meat quality and is commonly expressed by shear force.

This study found that in terms of slaughter performance, dietary supplementation with 200 mg/kg MOAEO significantly increased the breast muscle rate and leg muscle rate of 70-day-old broilers. Studies by Jin et al. and Zhang et al. also showed that OA or EO significantly increased the breast muscle rate and leg muscle rate of broilers. Zhu et al. reported that thymol EO had a certain degree of improvement on slaughter performance. Wang et al. found that organic acids had no effect on broiler slaughter performance. These different results may be related to the types and doses of EO and OA used. In terms of meat quality,

MOAEO decreased the pH_{45 min}, pH_{24 h}, and water loss rate of breast muscle, showing a certain improvement effect on meat quality. Similar results have been reported; Liu et al. found that rosemary EO could improve broiler meat quality by reducing breast muscle water loss rate, shear force, and leg muscle color. Wang et al. reported that organic acids significantly reduced the drip loss of breast and leg muscles and showed a trend toward reducing muscle shear force.

3.4 Effects of MOAEO on Serum Biochemical Indexes of Broilers

Serum total protein and albumin contents can reflect the nutritional, metabolic, and health status of animals. When nutritional status is good, protein synthesis in the body increases, and total protein and albumin contents rise. Conversely, insufficient protein intake can cause a decrease in total protein and albumin contents. The results of this experiment showed that dietary supplementation with MOAEO and antibiotics increased serum total protein content, indicating that MOAEO and antibiotics can promote protein deposition in broilers and thus promote broiler growth, which is consistent with the report by Wang et al. Uric acid is an important product of protein metabolism in poultry and can directly reflect the nutritional and protein metabolic status of animals. It is the most sensitive external indicator for measuring changes in protein metabolism in the body. Uric acid has low solubility in blood, and if purine metabolism is disordered (excessive uric acid production or long-term retention in the body) or uric acid excretion is blocked, the result is excessive uric acid content in the blood, forming hyperuricemia. Long-term high uric acid content can easily lead to gout and arthritis. This study showed that MOAEO and antibiotics reduced serum uric acid content, indicating that MOAEO and antibiotics can improve protein utilization efficiency and promote protein deposition in the body. Other studies have also shown that plant extracts or OA can reduce serum uric acid content in broilers.

In conclusion, MOAEO improved broiler growth performance, immune organ indexes, slaughter performance, meat quality, and serum biochemical indicators to a certain extent, with a suitable addition level of 200 mg/kg.

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