Effects of β-alanine on Growth Performance, Meat Quality, and Malondialdehyde and Carnosine Contents in Breast Muscle of Broiler Chickens (Postprint)

Authors: Qi Bo, Wu Shugeng, Wang Jing, Qi Guanghai, Zhang Haijun

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Abstract

This experiment aimed to investigate the effects of dietary β-alanine supplementation on growth performance, meat quality, and the contents of malondialdehyde and carnosine in breast muscle of broiler chickens. A total of 180 one-day-old Arbor Acres (AA) male broiler chicks were selected and randomly divided into 3 groups with 6 replicates per group and 10 birds per replicate. The control group was fed a basal diet, while the experimental groups were fed the basal diet supplemented with 500 (βA5 group) and 1,000 mg/kg β-alanine (βA10 group), respectively. The experimental period lasted for 42 days and was divided into two phases: the starter phase (1–21 days of age) and the grower phase (22–42 days of age). The results showed that: 1) Compared with the control group, the average body weight at 21 days of age in both βA5 and βA10 groups was significantly increased ($P<0.05$), and the average body weight at 42 days of age was increased by 30 g and 72 g, respectively ($P>0.05$); compared with the control group, the βA5 group showed an increasing trend in average daily gain (ADG) during the grower phase and the overall period (0.05

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Full Text

Effects of β-Alanine on Growth Performance, Meat Quality, and Breast Muscle Malondialdehyde and Carnosine Contents in Broilers

Qi Bo, Wu Shugeng, Wang Jing, Qi Guanghai, Zhang Haijun*

Feed Research Institute, Chinese Academy of Agricultural Sciences, Key Laboratory of Feed Biotechnology of Ministry of Agriculture, National Engineering Research Center of Biological Feed, Beijing 100081, China

Abstract

This experiment was conducted to investigate the effects of dietary β-alanine supplementation on growth performance, meat quality, and malondialdehyde (MDA) and carnosine contents in breast muscle of broilers. A total of 180 one-day-old Arbor Acres (AA) male broilers were randomly allocated into 3 groups with 6 replicates of 10 birds each. The control group was fed a basal diet, while the experimental groups were fed the basal diet supplemented with 500 mg/kg β-alanine (βA5 group) or 1,000 mg/kg β-alanine (βA10 group). The 42-day trial period was divided into two phases: early phase (1–21 days) and late phase (22–42 days). The results showed that: 1) Compared with the control group, the average body weight at 21 days was significantly increased in both βA5 and βA10 groups ($P<0.05$), and the average body weight at 42 days was increased by 30 g and 72 g, respectively ($P>0.05$). The average daily gain (ADG) during the late phase and overall period tended to increase in the βA5 group ($0.05<P<0.10$). The feed conversion ratio (FCR) in the βA5 group was reduced during the early phase, late phase, and overall period compared with the control group, with significant differences observed during the early phase and overall period ($P<0.05$). 2) Drip loss of breast muscle was significantly decreased in the βA5 group ($P<0.05$), and the redness value (a*) at 24 h was significantly increased ($P<0.05$). 3) The MDA content in breast muscle showed decreasing trends in the βA5 group at 2 and 4 days post-slaughter ($P=0.091$ and $P=0.059$, respectively), and was significantly decreased at 6 days post-slaughter in both βA5 and βA10 groups ($P<0.05$). 4) Carnosine content in breast muscle was significantly increased in βA5 and βA10 groups compared with the control group ($P<0.05$), with increases of 19.5% and 14.4%, respectively. In conclusion, dietary supplementation with 500 mg/kg β-alanine improved growth performance, reduced drip loss and MDA content, and increased carnosine content in breast muscle, thereby enhancing meat quality in broilers.

Keywords: β-alanine; broilers; growth performance; meat quality; carnosine

Introduction

With the development of animal husbandry and increasing consumer health awareness, there is growing demand for nutritional and functional animal products. Poultry meat is favored by consumers due to its high protein, low fat, and low cholesterol characteristics, as well as its rich content of functional peptides. Recent research has identified chicken as the richest source of imidazole dipeptides (carnosine and anserine) among animal-derived foods, with studies reporting that chicken consumption may enhance memory function. This has further promoted chicken consumption and stimulated research interest in muscle-derived bioactive peptides. β-Alanine is the limiting amino acid for carnosine synthesis and the only naturally occurring β-amino acid. It does not participate in protein synthesis but exerts its functions in humans and mammals primarily through carnosine, which possesses biological activities including enhanced exercise performance, anti-fatigue effects, antioxidant capacity, and improved muscle buffering capacity. Previous studies have shown that dietary β-alanine supplementation can improve broiler production performance, increase carnosine content in muscle tissue, reduce malondialdehyde (MDA) content in tissues and serum, enhance muscle antioxidant capacity, and improve sensory characteristics such as flavor and satiety. However, current research on β-alanine has primarily focused on production performance and muscle carnosine content, with inconsistent results. Few studies have investigated the practical effects and optimal dosage of β-alanine in broiler production. Therefore, this study aimed to examine the effects of β-alanine on growth performance, meat quality, and carnosine and MDA contents in breast muscle, and to explore the efficacy of low-dose β-alanine supplementation in broilers, providing a scientific basis for its application in livestock and poultry production.

Materials and Methods

1.1 Experimental Materials One-day-old Arbor Acres (AA) broiler chicks were purchased from Beijing Huadu Broiler Company. β-Alanine was used as the experimental supplement.

1.2 Experimental Design and Diets A single-factor design was employed. One hundred eighty healthy one-day-old AA male broilers were randomly divided into 3 groups with 6 replicates of 10 birds each. The trial lasted 42 days and was divided into two phases: early phase (1–21 days) and late phase (22–42 days). Basal diets were formulated according to NRC (1994) nutrient requirements, the Chinese Chicken Feeding Standard (NY/T 33-2004), and the Arbor Acres Broiler Management Guide. The composition and nutrient levels of the basal diets are shown in Table 1 . The control group received the basal diet, while the experimental groups (βA5 and βA10) received the basal diet supplemented with 500 mg/kg and 1,000 mg/kg β-alanine, respectively.

1.3 Management Birds were provided ad libitum access to feed and water under 24-hour lighting. The room temperature was maintained at 33°C for the first 3 days, then reduced by 2°C weekly until reaching 24°C, which was maintained for the remainder of the trial. Management followed the AA Broiler Management Guide, with routine vaccination and disinfection. The chicken house was well-ventilated. Temperature and humidity were recorded daily, and mortality was recorded throughout the experiment.

1.4.1 Growth Performance At the start of the experiment and on days 21 and 42, birds were weighed by replicate after fasting to calculate average body weight (ABW) and average daily gain (ADG). Feed consumption was recorded by replicate to calculate average daily feed intake (ADFI) and feed conversion ratio (F/G) for the early, late, and overall periods. Mortality was recorded daily to calculate mortality rate.

1.4.2 Meat Quality At 42 days, one bird per replicate with body weight close to the replicate average was selected, slaughtered by jugular venipuncture, and the right breast muscle was completely excised for determination of pH, meat color, drip loss, cooking loss, and shear force.

pH: pH was measured at 45 min and 24 h post-slaughter (stored at 4°C) using a pH meter (cyberScan pH 310 waterproof pen type, EUTECH, Singapore). The probe was inserted approximately 1 cm deep into the breast muscle. The electrode head was completely embedded in the meat sample, and each sample was measured three times with the average recorded as pH45min and pH24h.

Meat Color: At 45 min and 24 h post-slaughter (stored at 4°C), meat color was evaluated using the CIE-Lab system with a TCP2 colorimeter (Shanghai Precision Scientific Instrument Co., Ltd., Shanghai). Lightness (L*), redness (a*), and yellowness (b*) values were measured three times per sample and averaged.

Drip Loss: Within 45 min post-slaughter, approximately 30 g of breast muscle with similar texture and regular shape was weighed (W1), placed in a ziplock bag, inflated with nitrogen to minimize contact with the bag interior, suspended in a 4°C refrigerator, and reweighed after 24 h (W2) after gently blotting surface moisture with filter paper. Drip loss (%) = [(W1-W2)/W1] × 100.

Cooking Loss: The meat sample after 24 h drip loss measurement was placed in a new ziplock bag, heated in an 80°C water bath until the core temperature reached 75°C, cooled to room temperature under running water, and reweighed (W3) after gently blotting surface moisture. Cooking loss (%) = [(W2-W3)/W2] × 100.

Shear Force: After cooking loss measurement, meat samples were trimmed into two strips (2 cm × 2 cm × 1 cm) along the muscle fiber direction. Shear force was measured with the muscle fiber direction perpendicular to the blade using a TMS-Pro tenderness analyzer (Food Technology Corporation, Virginia, USA) with a maximum load of 100 N, crosshead speed of 150 mm/min, and gap width of 6 mm. Each sample was tested three times, and the average of six measurements from two strips was recorded as the final shear force value.

1.4.3 Carnosine Content Instrumentation: High-performance liquid chromatography (Agilent LC-15, USA), carnosine standard (98%, Sigma), homogenizer (QL-901/QL-861, Haimen Qilinbeier), high-speed refrigerated centrifuge (10 mL, Hunan Hexi), and methanol (HPLC grade).

Standard Preparation: Ten milligrams of carnosine standard was accurately weighed, dissolved in ultrapure water, and diluted to 10 mL to prepare a 1 mg/mL stock solution. Standard working solutions of 10, 20, 50, 80, and 100 g/mL were prepared by diluting with 0.1% trifluoroacetic acid (TFA).

Sample Preparation: At 42 days, one bird per replicate with body weight close to the replicate average was selected, slaughtered, and breast muscle was collected and stored at -20°C. Half a gram of breast muscle was minced, mixed with 4.5 mL physiological saline, and homogenized three times at 4,000 r/min for 30 s. The homogenate was centrifuged at 3,500 r/min for 10 min, and the supernatant was mixed with three volumes of methanol, frozen at -20°C for 15 min, and centrifuged at 11,000 r/min. The supernatant was diluted 10-fold for analysis.

HPLC Conditions: Separation was performed on an Agilent B8 column (150 mm × 4.6 mm, id: 5 m) using 0.1% TFA as the mobile phase and acetonitrile as the organic phase (95.5% organic phase) at a flow rate of 1.0 mL/min. Injection volume was 30 L, and detection wavelength was 220 nm.

1.4.4 MDA Content MDA content was determined using a commercial kit (Nanjing Jiancheng Bioengineering Institute) following the manufacturer' s instructions.

Sample Preparation: At 42 days, one bird per replicate with body weight close to the average was slaughtered by jugular venipuncture, and approximately 200 g of right breast muscle was collected and stored at -20°C. MDA content in muscle homogenate was measured at 2, 4, and 6 days post-slaughter. For homogenization, 0.50 g ($±$0.005 g) of breast muscle was placed in a centrifuge tube with 9 volumes of physiological saline and homogenized three times at 4,000 r/min for 30 s on ice. The homogenate was centrifuged at 3,000 r/min and 4°C for 15 min, and the supernatant was used for analysis.

1.5 Statistical Analysis Data are expressed as means ± standard deviation. One-way ANOVA was performed using SPSS 16.0 software, followed by F-test and Duncan' s multiple comparison test. Differences were considered significant at $P<0.05$.

Results

2.1 Effects of β-Alanine on Growth Performance of Broilers The effects of β-alanine on growth performance are presented in Table 2 . Compared with the control group, average body weight at 21 days was significantly increased in both βA5 and βA10 groups ($P<0.05$). Average body weight at 42 days was increased by 30 g and 72 g in βA5 and βA10 groups, respectively, though the differences were not significant ($P>0.05$). The ADG during the late phase and overall period tended to increase in the βA5 group ($0.05<P<0.10$), while ADG in the βA10 group was numerically higher during all phases but not significantly different ($P>0.05$). The FCR in βA5 and βA10 groups was reduced during the early and overall periods compared with the control group, with significant differences observed in the βA5 group ($P<0.05$). The FCR during the late phase was also reduced in the βA5 group ($P>0.05$). No significant differences were observed in ADFI or mortality among groups during any phase ($P>0.05$).

2.3 Effects of β-Alanine on Meat Quality of Broilers The effects of β-alanine on meat quality are shown in Table 3 . Drip loss of breast muscle was significantly decreased in the βA5 group compared with the control group ($P<0.05$), while the βA10 group showed a numerical reduction without statistical significance ($P>0.05$). The a* value at 24 h was significantly increased in the βA5 group ($P<0.05$) and numerically increased in the βA10 group. No significant differences were observed among groups in cooking loss, shear force, pH, or other meat color parameters ($P>0.05$).

2.4 Effects of β-Alanine on MDA Content in Breast Muscle of Broilers The effects of β-alanine on MDA content in breast muscle are presented in Table 4 . MDA content was significantly decreased in the βA5 group at 2 and 4 days post-slaughter (t-test, $P<0.001$ and $P=0.001$, respectively). At 6 days post-slaughter, MDA content was significantly decreased in both βA5 and βA10 groups ($P<0.05$). The βA5 group demonstrated lower MDA content and superior efficacy compared with the βA10 group. MDA content in the control and βA5 groups increased with storage time post-slaughter.

2.4 Effects of β-Alanine on Carnosine Content in Breast Muscle of Broilers The effects of β-alanine on carnosine content in breast muscle are shown in Table 5 . Carnosine content was significantly increased in both βA5 and βA10 groups compared with the control group ($P<0.05$), with increases of 19.5% and 14.4%, respectively.

Discussion

3.1 Effects of β-Alanine on Growth Performance of Broilers Limited research has been conducted on the effects of β-alanine on broiler growth performance in China. Early foreign studies demonstrated that dietary supplementation with 2.5% β-alanine significantly reduced ADFI and improved F/G in broilers. Oral administration of 22 mmol/kg (0.2%) β-alanine twice daily for 5 days starting at 2 days of age significantly improved feed conversion efficiency. The present study showed that low-dose β-alanine supplementation (500 and 1,000 mg/kg) improved feed conversion ratio, consistent with the findings of Tomonaga et al. and Hu Xinxu. Considering ADG, ADFI, and FCR comprehensively, the 500 mg/kg dose demonstrated superior effects. The effective dose identified in this study is relatively low, which is beneficial for resource utilization and feed cost control. However, excessive β-alanine supplementation (2%) has been reported to inhibit daily gain and feed intake. Therefore, appropriate dosage is crucial for practical application in broiler production.

Furthermore, Tomonaga et al. found that the administration method of β-alanine (drinking water vs. feed) affected growth performance. This study demonstrated that feed supplementation improved F/G by increasing ADG, while other research suggested that β-alanine improved F/G by reducing ADFI. This discrepancy may be attributed to β-alanine' s role as a neurotransmitter in regulating hormone secretion, thereby influencing feed intake or daily gain to modulate animal growth. The pathways and mechanisms by which β-alanine regulates growth performance require further investigation.

3.2 Effects of β-Alanine on Carnosine Content in Broilers Carnosine is an endogenous bioactive peptide predominantly found in animal muscle tissue, with functions including free radical scavenging, muscle pH buffering, anti-aging, and memory enhancement. As a substrate for carnosine synthesis, β-alanine exerts its functions in animals (as a neurotransmitter and performance enhancer) primarily through carnosine synthesis. Therefore, changes in tissue carnosine content are crucial for understanding β-alanine' s effects. Previous studies have shown that dietary β-alanine supplementation (0.5% and 1.0%) significantly increased carnosine content in broiler breast muscle, while other research found that β-alanine increased carnosine content in brain tissue but not in breast muscle. This study demonstrated that supplementation with 500 and 1,000 mg/kg β-alanine significantly increased carnosine content in broiler breast muscle by 14.3% and 19.5%, respectively.

Research has shown that exogenous addition of labeled β-alanine to chicken embryonic cells resulted in detectable radiolabeled dipeptides in cell extracts, with dipeptide content increasing significantly during myoblast differentiation. In vivo, β-alanine content in muscle is lower than histidine relative to the Km value of carnosine synthase; therefore, the rate of carnosine synthesis in muscle is primarily limited by β-alanine availability. Studies have indicated that animal sex, genotype, expression of β-alanine transporters PAT1 and TAUT genes, and carnosine transporters PAT and PEPT genes can influence carnosine synthesis and content. β-Alanine may increase carnosine content by upregulating the expression of β-alanine and carnosine transporters, though relevant research in broilers remains scarce. Further in-depth studies are needed to elucidate the pathways and mechanisms by which β-alanine increases carnosine content.

3.3 Effects of β-Alanine on Meat Quality of Broilers Meat quality is an important indicator of meat appearance. Post-slaughter, muscle enters rigor mortis, muscle fibers contract, water-holding capacity decreases, and moisture exudes to the surface, increasing L* values. Previous research has shown that 1% β-alanine reduced shear force and L* values in 42-day-old broilers, while 0.5% β-alanine decreased drip loss and cooking loss and increased a* values. This study demonstrated that β-alanine reduced drip loss and increased 24 h a* values, thereby improving meat quality. These results are consistent with Kralik et al., though the dosage used in this study was lower.

Additionally, pre-slaughter heat stress has been shown to reduce a* values (below normal range) and increase L* values (above normal range) in broiler breast muscle, predisposing to pale, soft, and exudative (PSE) meat. Dietary β-alanine supplementation increased a* values and reduced PSE meat incidence. Meat color is primarily determined by muscle pigment content; therefore, β-alanine supplementation may affect pigment content, though this was not measured in the present study and warrants further investigation.

3.4 Effects of β-Alanine on MDA Content in Breast Muscle of Broilers β-Alanine possesses strong antioxidant properties, enhancing superoxide dismutase (SOD) activity, reducing MDA content, and improving muscle antioxidant capacity. Previous studies have shown that dietary β-alanine supplementation (0.5% and 1.0%) tended to reduce MDA content in broiler breast muscle, while 0.12% β-alanine significantly decreased MDA content and increased glutathione peroxidase activity. This study demonstrated that β-alanine reduced MDA content in breast muscle at 2, 4, and 6 days post-slaughter, possibly due to increased carnosine content, as carnosine is an antioxidant that reduces lipid peroxidation and enhances oxidative stability.

Conclusion

Dietary supplementation with low doses of β-alanine (500 and 1,000 mg/kg) promoted growth, improved meat quality, enhanced antioxidant capacity, and increased carnosine content in broiler breast muscle. Considering the comprehensive results on growth performance, meat quality, and carnosine content, supplementation with 500 mg/kg β-alanine demonstrated optimal efficacy.

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