

Effects of Dietary L-Arginine Content on Amino Acid Digestibility, Serum Amino Acid Content, and Serum Biochemical Parameters in Female Mink during the Winter Fur Period: Postprint

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Abstract

This experiment aimed to investigate the effects of dietary L-arginine content on amino acid digestibility, serum amino acid concentrations, and serum biochemical indices in female mink during the winter fur period. Sixty-three healthy female mink aged (150 ± 3) days were selected and randomly divided into 7 groups (9 replicates per group, 1 mink per replicate). They were fed experimental diets supplemented with 0, 0.20%, 0.40%, 0.60%, 0.80%, 1.00%, and 1.20% L-arginine to the basal diet, resulting in dietary L-arginine contents of 1.70%, 1.90%, 2.10%, 2.30%, 2.50%, 2.70%, and 2.90%, respectively. The pre-trial period lasted 7 days, and the formal trial period lasted 60 days. The results showed that: 1) Dry matter intake, dry matter excretion, and dry matter digestibility did not differ significantly among groups ($P > 0.05$). 2) Lysine digestibility in the 1.70% group was significantly higher than that in the 2.50%, 2.70%, and 2.90% groups ($P < 0.05$). Arginine digestibility in the 2.70% and 2.90% groups was significantly higher than that in the 1.70% and 1.90% groups ($P < 0.05$). 3) Serum serine and proline concentrations in the 2.30% group were significantly higher than those in the 1.70%, 1.90%, 2.10%, and 2.90% groups ($P < 0.05$). Serum alanine concentration in the 2.30% and 2.50% groups was significantly higher than that in the 1.70%, 1.90%, 2.10%, and 2.90% groups ($P < 0.05$). Serum phenylalanine concentration in the 2.30% group was significantly higher than that in the 1.70% and 2.90% groups ($P < 0.05$). Serum arginine concentration in the 2.30% group was significantly higher than that in the 1.70%, 1.90%, 2.10%, and 2.90% groups ($P < 0.05$). 4) Serum aspartate aminotransferase activity in the 2.30% group was significantly higher than that in the 1.70% group ($P < 0.05$). Serum lactate dehydrogenase activity in the 2.30% and 2.50% groups was significantly higher than that in the 1.70%, 1.90%, 2.10%, 2.70%, and 2.90% groups

($P < 0.05$). 5) Serum immunoglobulin M concentration in the 2.30% and 2.50% groups was significantly higher than that in the 1.70%, 1.90%, 2.10%, 2.70%, and 2.90% groups ($P < 0.05$). Serum immunoglobulin G concentration in the 2.30% group was significantly higher than that in the 1.70% and 2.90% groups ($P < 0.05$). Serum complement 4 concentration in the 2.30% and 2.50% groups was significantly higher than that in the 1.70% and 2.90% groups ($P < 0.05$), and serum complement 4 concentration in the 2.10% group was significantly higher than that in the 1.70% group ($P < 0.05$). Based on these results, when dietary L-arginine content was 2.30%, female mink during the winter fur period achieved better protein and amino acid metabolism, while immune function was enhanced.

Full Text

Effects of Dietary L-Arginine Content on Amino Acid Digestibility, Serum Amino Acid Contents and Serum Biochemical Indices of Female Mink in the Winter Fur-Growing Period

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Abstract

This study investigated the effects of dietary L-arginine content on amino acid digestibility, serum amino acid contents, and serum biochemical indices in female mink during the winter fur-growing period. Sixty-three healthy female mink aged (150 ± 3) days were randomly allocated into seven groups (nine replicates per group, one mink per replicate). The animals were fed experimental diets supplemented with 0%, 0.20%, 0.40%, 0.60%, 0.80%, 1.00%, and 1.20% L-arginine, resulting in dietary L-arginine concentrations of 1.70%, 1.90%, 2.10%, 2.30%, 2.50%, 2.70%, and 2.90%, respectively. The pre-trial period lasted 7 days, followed by a 60-day formal experimental period. The results showed: 1) No significant differences in dry matter intake, dry matter output, or dry matter digestibility among all groups ($P > 0.05$). 2) Lysine digestibility in the 1.70% group was significantly higher than in the 2.50%, 2.70%, and 2.90% groups ($P < 0.05$). Arginine digestibility in the 2.70% and 2.90% groups was significantly higher than in the 1.70% and 1.90% groups ($P < 0.05$). 3) Serum serine and proline contents in the 2.30% group were significantly higher than in the 1.70%, 1.90%, 2.10%, and 2.90% groups ($P < 0.05$). Serum alanine content in

the 2.30% and 2.50% groups was significantly higher than in the 1.70%, 1.90%, 2.10%, and 2.90% groups ($P < 0.05$). Serum phenylalanine content in the 2.30% group was significantly higher than in the 1.70% and 2.90% groups ($P < 0.05$). Serum arginine content in the 2.30% group was significantly higher than in the 1.70%, 1.90%, 2.10%, and 2.90% groups ($P < 0.05$). 4) Serum aspartate aminotransferase activity in the 2.30% group was significantly higher than in the 1.70% group ($P < 0.05$). Serum lactate dehydrogenase activity in the 2.30% and 2.50% groups was significantly higher than in the 1.70%, 1.90%, 2.10%, 2.70%, and 2.90% groups ($P < 0.05$). 5) Serum immunoglobulin M content in the 2.30% and 2.50% groups was significantly higher than in the 1.70%, 1.90%, 2.10%, 2.70%, and 2.90% groups ($P < 0.05$). Serum immunoglobulin G content in the 2.30% group was significantly higher than in the 1.70% and 2.90% groups ($P < 0.05$). Serum complement 4 content in the 2.30% and 2.50% groups was significantly higher than in the 1.70% and 2.90% groups ($P < 0.05$), while serum complement 4 content in the 2.10% group was significantly higher than in the 1.70% group ($P < 0.05$). These findings indicate that a dietary L-arginine content of 2.30% supports optimal protein and amino acid metabolism while enhancing immune function in female mink during the winter fur-growing period.

Keywords: L-arginine; winter fur-growing period; female mink; amino acid digestibility; serum amino acid contents; serum biochemical indices

Introduction

Arginine, a conditionally essential amino acid, serves not only as a crucial substrate for protein synthesis but also plays vital roles in physiological regulation. Endogenous arginine synthesis is key to maintaining arginine homeostasis; however, dietary arginine supplementation can antagonize the absorption of tryptophan, lysine, and histidine, thereby affecting the digestibility of multiple amino acids and serum amino acid profiles. Serum biochemical indices are important indicators of immune function, enzyme activity, and organ integrity, and can reflect protein utilization efficiency. Consequently, measuring changes in serum biochemical indices helps elucidate nutrient metabolism in animals. Our research group previously investigated the effects of dietary L-arginine levels on growth performance, nutrient digestibility, and nitrogen metabolism in growing and winter fur-growing mink. However, research on arginine-related amino acid digestibility, serum amino acid contents, and serum biochemical indices in mink remains limited. This study examined the effects of dietary arginine levels on these parameters in winter fur-growing mink to provide scientific evidence for refining feeding standards and to establish a foundation for understanding arginine's nutritional mechanisms in mink.

Materials and Methods

1.1 Experimental Animals Sixty-three healthy female mink aged (150 ± 3) days with similar body weights were randomly selected from the fur animal production base at the Changbai Mountain Wildlife Resources Field Scientific Observation Station of the Ministry of Agriculture.

1.2 Experimental Diets As no unified feeding standard exists for mink in China, the basal diet was formulated according to NRC (1982) nutrient requirements for mink. The composition, nutrient levels, and amino acid profile of the basal diet are presented in and . L-arginine was supplemented at 0%, 0.20%, 0.40%, 0.60%, 0.80%, 1.00%, and 1.20% to create seven experimental diets with final L-arginine concentrations of 1.70%, 1.90%, 2.10%, 2.30%, 2.50%, 2.70%, and 2.90%, respectively.

1.3 Experimental Design The 63 mink were randomly divided into seven groups with nine replicates each (one mink per replicate). Group mean body weights were adjusted through analysis of variance to ensure no significant differences ($P>0.05$). Each group received one of the seven experimental diets with L-arginine contents ranging from 1.70% to 2.90%.

1.4 Management Prior to the trial, all mink were vaccinated against canine distemper and parvovirus. Animals were housed individually in cages and fed twice daily at 07:30 and 15:30 with ad libitum access to feed and water. Daily feed intake was recorded. The pre-trial period lasted 7 days, followed by a 60-day formal trial period from September 14 to November 20, 2014.

1.5 Digestion-Metabolism Trial On day 42 of the trial, six mink per group with similar body weights were selected for a digestion-metabolism study conducted from October 27 to 29, 2014 (3 days). The total collection method was employed, with management consistent with daily practices. Daily fecal collections were weighed, and 10% sulfuric acid solution (5% of fresh weight) was added with a small amount of toluene as preservative before storage at -20°C . The three-day fecal samples were pooled, sterilized at 80°C for 2 hours, dried to constant weight at 65°C , ground through a 40-mesh sieve, and prepared as air-dried samples for laboratory analysis.

1.6 Serum Preparation At the end of the feeding trial, six mink per group were selected for cardiac blood collection (5 mL per animal). Blood was placed in coagulation-promoting tubes, allowed to clot, then centrifuged at 3,500 r/min for 10 minutes at 4°C . Serum was aliquoted into 1.5 mL Eppendorf tubes and stored at -80°C .

1.7 Measurements Dry matter content in diets and feces was determined using the 105°C oven-drying method according to GB/T 6435-2006. Amino acid

contents were measured using hydrochloric acid hydrolysis per GB/T 5009.124-2003. Serum amino acids were analyzed with a Hitachi L-8900 amino acid analyzer after protein precipitation with trichloroacetic acid. Serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), and lactate dehydrogenase (LDH) activities, along with total protein (TP), albumin (ALB), immunoglobulin A (IgA), immunoglobulin G (IgG), immunoglobulin M (IgM), complement 3 (C3), and complement 4 (C4) concentrations were measured using a VITAL-E automatic biochemical analyzer with commercial kits (Beijing Zhongsheng Beikong Biotech Co., Ltd.). Globulin (GLOB) concentration was calculated as the difference between TP and ALB.

Dry matter digestibility (%) = [(DM intake - DM output) / DM intake] × 100;
Amino acid digestibility (%) = [(Amino acid intake - Amino acid output) / Amino acid intake] × 100.

Data are expressed as “mean ± standard deviation.” Statistical analysis was performed using SPSS 17.0 software with one-way ANOVA for significance testing, where P<0.05 indicated significant difference and P<0.01 indicated extremely significant difference.

Results

2.1 Effects of Dietary L-Arginine Content on Dry Matter Digestibility

As shown in , no significant differences were observed in dry matter intake, dry matter output, or dry matter digestibility among groups (P>0.05).

2.2 Effects of Dietary L-Arginine Content on Amino Acid Digestibility

reveals that lysine digestibility in the 1.70% group was significantly higher than in the 2.50%, 2.70%, and 2.90% groups (P<0.05). Arginine digestibility in the 2.70% and 2.90% groups was significantly higher than in the 1.70% and 1.90% groups (P<0.05). No significant differences were detected for other amino acids among groups (P>0.05).

2.3 Effects of Dietary L-Arginine Content on Serum Amino Acid Contents

According to , serum serine and proline contents in the 2.30% group were significantly higher than in the 1.70%, 1.90%, 2.10%, and 2.90% groups (P<0.05). Serum alanine content in the 2.30% and 2.50% groups was significantly higher than in the 1.70%, 1.90%, 2.10%, and 2.90% groups (P<0.05). Serum phenylalanine content in the 2.30% group was significantly higher than in the 1.70% and 2.90% groups (P<0.05). Serum arginine content in the 2.30% group was significantly higher than in the 1.70%, 1.90%, 2.10%, and 2.90% groups (P<0.05). No significant differences were observed for other serum amino acids among groups (P>0.05).

2.4 Effects of Dietary L-Arginine Content on Serum Routine Biochemical Indices

shows that serum aspartate aminotransferase activity in

the 2.30% group was significantly higher than in the 1.70% group ($P < 0.05$). Serum lactate dehydrogenase activity in the 2.30% and 2.50% groups was significantly higher than in the 1.70%, 1.90%, 2.10%, 2.70%, and 2.90% groups ($P < 0.05$). No significant differences were detected for other routine biochemical indices among groups ($P > 0.05$).

2.5 Effects of Dietary L-Arginine Content on Serum Immune Biochemical Indices demonstrates that serum immunoglobulin M content in the 2.30% and 2.50% groups was significantly higher than in the 1.70%, 1.90%, 2.10%, 2.70%, and 2.90% groups ($P < 0.05$). Serum immunoglobulin G content in the 2.30% group was significantly higher than in the 1.70% and 2.90% groups ($P < 0.05$). Serum complement 4 content in the 2.30% and 2.50% groups was significantly higher than in the 1.70% and 2.90% groups ($P < 0.05$), while serum complement 4 content in the 2.10% group was significantly higher than in the 1.70% group ($P < 0.05$). No significant differences were observed for other immune indices among groups ($P > 0.05$).

Discussion

3.1 Effects on Dry Matter and Amino Acid Digestibility In this study, dietary L-arginine content did not significantly affect dry matter intake, output, or digestibility in winter fur-growing mink. However, supplementation significantly influenced the digestibility of certain amino acids. Wu et al. reported that arginine absorption antagonizes lysine utilization. High dietary arginine levels affect lysine absorption, degradation, synthesis, and reabsorption, primarily because both are basic amino acids that share the same transport system, creating competitive antagonism during absorption. This study demonstrated that as dietary arginine content increased, arginine digestibility gradually increased while lysine digestibility decreased, suggesting antagonistic absorption between arginine and lysine in winter fur-growing mink. Thus, enhanced arginine absorption and utilization apparently inhibited lysine absorption and utilization.

3.2 Effects on Serum Amino Acid Contents Serum free amino acid concentrations partially reflect amino acid metabolic function in animals; low serum free amino acid levels indicate inadequate dietary amino acid supply. In vivo, arginine is utilized primarily through the arginase pathway, yielding proline, ornithine, and urea as major metabolites. Wu et al. demonstrated that proline is the main precursor for arginine synthesis in porcine enterocytes, establishing a close link between serum proline levels and arginine metabolism. In this study, increasing dietary arginine significantly altered serum serine, proline, alanine, phenylalanine, and arginine concentrations. Notably, the 2.30% L-arginine group exhibited the highest values for multiple amino acids, indicating superior amino acid metabolism.

3.3 Effects on Serum Routine Biochemical Indices Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are predominantly localized

in hepatocytes, with ALT mainly in the cytoplasm and AST in both cytoplasm and mitochondria. Serum ALT and AST activities reflect hepatic protein synthesis capacity and liver function status. Additionally, alkaline phosphase (ALP) and lactate dehydrogenase (LDH) activities indicate liver and kidney function, while AST and ALT activities reflect protein metabolism and amino acid utilization. Pan et al. reported that dietary arginine supplementation in weaned piglets increased serum total protein, albumin, ALP, and ALT activities compared to controls. In this study, appropriate arginine supplementation elevated serum AST and LDH activities in winter fur-growing mink, with maximal activity observed at 2.30% L-arginine. Increased AST and LDH activities suggest enhanced protein metabolic function and kidney function, indicating that 2.30% dietary arginine optimizes protein metabolism in winter fur-growing mink.

3.4 Effects on Serum Immune Biochemical Indices Immunoglobulins, components of serum total protein, directly reflect immune capacity. Complement comprises a group of serum and tissue fluid proteins that acquire enzymatic activity upon activation. Immunoglobulins are the primary immune molecules mediating humoral immunity, and their combination with complement can kill bacteria and viruses. Arginine improves humoral immune function. Li et al. reported that arginine supplementation in 7-day-old weaned piglets significantly increased serum immunoglobulin content by day 14. Yang found that 1% dietary arginine effectively elevated serum immunoglobulin levels in pregnant sows. Ma demonstrated that L-arginine supplementation significantly increased serum immunoglobulin A content and enhanced humoral immunity in growing rabbits. In this study, supplementation with 0.60% L-arginine (achieving 2.30% dietary content) increased serum IgM, IgG, and complement C4 levels in winter fur-growing mink, confirming enhanced immune function at this arginine concentration.

Based on these results, a dietary arginine content of 2.30% supports optimal protein and amino acid metabolism while enhancing immune function in female mink during the winter fur-growing period.

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