Optimal Supplementation Level and Method of Cysteamine in Diets for Growing Mink (Postprint)

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Abstract

This experiment aimed to determine the appropriate supplementation level and method of cysteamine in diets by investigating its effects on growth performance, nutrient digestibility, and nitrogen metabolism in growing minks. A two-factor experimental design was adopted, selecting 56 healthy female short-haired black minks of similar body weight at (86$±$5) days of age, randomly divided into 7 groups with 8 replicates per group and 1 mink per replicate. Group I (control group) was fed the basal diet; Groups II, III, and IV were fed the basal diet supplemented with 60, 90, and 120 mg/kg cysteamine, respectively, with continuous supplementation; Groups V, VI, and VII were fed the basal diet supplemented with 60, 90, and 120 mg/kg cysteamine, respectively, with intermittent supplementation (continuous supplementation for 1 week, followed by a 1-week interval). The pre-trial period lasted 7 days, and the formal trial period lasted 53 days. The results showed that: 1) Dietary cysteamine supplementation extremely significantly affected the average daily feed intake and feed-to-gain ratio of minks ($P<0.01$), but had no significant effect on average daily gain ($P>0.05$). The feed-to-gain ratio of Group V was extremely significantly lower than that of Groups I, II, and IV ($P<0.01$); the average daily gain was highest in Group V, and the intermittent supplementation groups were higher than the continuous supplementation groups. 2) Dietary cysteamine supplementation significantly affected the crude protein digestibility and crude fat digestibility of minks ($P<0.05$), and extremely significantly affected dry matter digestibility ($P<0.01$), with all these indices being highest in Group V. The cysteamine supplementation method extremely significantly affected the dry matter digestibility of minks ($P<0.01$), with the intermittent supplementation groups being extremely significantly higher than the continuous supplementation groups ($P<0.01$). 3) Dietary cysteamine supplementation significantly or extremely significantly affected the nitrogen metabolism indices of minks (P<0.05 or P<0.01). The nitrogen retention of Groups V, VI, and VII was significantly higher than that of Group I (P<0.05), and the net protein utilization and protein biological value of Groups V, VI, and VII were both extremely significantly higher than those of Group I (P<0.01), with Group V being the highest. The cysteamine supplementation method extremely significantly affected the nitrogen intake, urinary nitrogen excretion, net protein utilization, and protein biological value of minks (P<0.01). The nitrogen intake and urinary nitrogen excretion of minks in the intermittent supplementation groups were extremely significantly lower than those in the continuous supplementation groups (P<0.01); the nitrogen retention of the intermittent supplementation groups was significantly lower than that of the continuous supplementation groups (P<0.05); the net protein utilization and protein biological value of the intermittent supplementation groups were extremely significantly higher than those of the continuous supplementation groups (P<0.01). The cysteamine supplementation level significantly affected the fecal nitrogen excretion of minks (P<0.05), with the fecal nitrogen excretion of the 90 mg/kg group being significantly lower than that of the 60 mg/kg group (P<0.05). Based on comprehensive analysis of all indices, the appropriate supplementation level of cysteamine in diets for growing minks was 60 mg/kg, and the appropriate supplementation method was intermittent supplementation.

Full Text

Study on the Appropriate Supplemental Level and Method of Cysteamine in Diets for Growing Minks

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Abstract: This experiment was conducted to investigate the effects of dietary cysteamine (CS) on growth performance, nutrient digestibility, and nitrogen metabolism in growing minks, and to determine the appropriate supplemental level and method. A two-factor experimental design was adopted. Fifty-six healthy black short-haired female minks at (86$±$5) days of age with similar body weight were randomly divided into 7 groups with 8 replicates per group and 1 mink per replicate. Group I (control) was fed a basal diet; groups II, III, and IV were fed the basal diet supplemented with 60, 90, and 120 mg/kg CS, respectively, with continuous supplementation; groups V, VI, and VII were fed the basal diet supplemented with 60, 90, and 120 mg/kg CS, respectively, with interval supplementation (continuous supplementation for 1 week, followed by a 1-week interval). The preliminary period lasted 7 days, and the formal experimental period lasted 53 days.

The results showed: 1) Dietary CS supplementation extremely significantly affected average daily feed intake and feed-to-gain ratio ($P<0.01$), but had no significant effect on average daily gain ($P>0.05$). The feed-to-gain ratio in group V was extremely significantly lower than that in groups I, II, and IV ($P<0.01$); average daily gain was highest in group V, and the interval supplementation group was higher than the continuous supplementation group. 2) Dietary CS supplementation significantly affected crude protein digestibility and ether extract digestibility ($P<0.05$), and extremely significantly affected dry matter digestibility ($P<0.01$), with all values being highest in group V. The CS supplementation method extremely significantly affected dry matter digestibility ($P<0.01$), with the interval supplementation group being extremely significantly higher than the continuous supplementation group ($P<0.01$). 3) Dietary CS supplementation significantly or extremely significantly affected nitrogen metabolism indices ($P<0.05$ or $P<0.01$). Nitrogen deposition in groups V, VI, and VII was significantly higher than that in group I ($P<0.05$); net protein utilization and protein biological value in groups V, VI, and VII were extremely significantly higher than those in group I ($P<0.01$), with the highest values observed in group V. The CS supplementation method extremely significantly affected nitrogen intake, urinary nitrogen excretion, net protein utilization, and protein biological value ($P<0.01$), and significantly affected nitrogen deposition ($P<0.05$). Nitrogen intake and urinary nitrogen excretion in the interval supplementation group were extremely significantly lower than those in the continuous supplementation group ($P<0.01$); nitrogen deposition in the interval supplementation group was significantly higher than that in the continuous supplementation group ($P<0.05$); net protein utilization and protein biological value in the interval supplementation group were extremely significantly higher than those in the continuous supplementation group ($P<0.01$). The CS supplemental level significantly affected fecal nitrogen excretion ($P<0.05$), with the 90 mg/kg group being significantly lower than the 60 mg/kg group ($P<0.05$). Based on comprehensive analysis of all indicators, the appropriate supplemental level of cysteamine in diets for growing minks is 60 mg/kg, and the appropriate supplementation method is interval supplementation.

Keywords: minks; cysteamine; growth performance; nutrient digestibility; nitrogen metabolism

With the rapid development of China' s fur animal industry, feed resource shortages in major fur animal production areas have become increasingly severe. Therefore, developing new feed raw materials, promoting full utilization of feed resources, and reducing production costs remain major challenges in fur animal production. Feed additives are one of the three pillars of compound feed ingredients, with multiple functions including improving feed conversion rate, promoting normal animal growth and development, and enhancing immunity [1]. Cysteamine (CS), as a feed additive, has various biological functions. Numerous studies have shown that CS can effectively inhibit the immunological activity and biological functions of somatostatin (SS) in the hypothalamus, stomach, and intestines of animals, thereby relieving the inhibitory effects on growth hormone (GH) and other hormones, subsequently promoting animal growth and improving feed efficiency [2-3]. Additionally, CS can promote digestive enzyme secretion, increase digestive enzyme activity, enhance small intestine digestive capacity, and thus promote nutrient digestion and absorption [4]. CS can also improve body immunity and antioxidant capacity [5]. Research has found that CS exhibits certain dose- and time-dependent effects, with high-dose supplementation causing digestive tract ulcers, while low-dose supplementation has a protective effect on digestive tract mucosa [6].

Due to its lack of species specificity, low cost, and wide application range, CS has been widely used in livestock and poultry production. However, research on the effects of CS on fur animals is limited. This study used minks as experimental subjects to investigate the effects of CS on growth performance, nutrient digestibility, and nitrogen metabolism, determine the appropriate supplemental level and method of CS in mink diets, and provide a theoretical basis for the rational use of CS in mink production.

1.1 Experimental Materials

The cysteamine used in this experiment was purchased from Shanghai Huakuoda Biotechnology Co., Ltd. Its main components were cysteamine hydrochloride, β-cyclodextrin, tocopherol, and chromium polynicotinate. The product was processed using supramolecular technology, multi-layer biomimetic membrane technology, and physiological homeostasis maintenance technology, providing certain stability with a purity of 99%.

1.2 Experimental Design and Management

A two-factor experimental design was adopted, involving different supplemental levels and methods. Fifty-six healthy black short-haired female minks at (86$±5)*daysofagewithsimilarbodyweight*[(795.70±$63.96) g] were randomly divided into 7 groups with 8 replicates per group and 1 mink per replicate. Group I (control) was fed a basal diet formulated based on commonly used fresh animal feed ingredients for minks, with composition and nutrient levels shown in Table 1 . Groups II, III, and IV were fed experimental diets supplemented with 60, 90, and 120 mg/kg CS, respectively, with continuous supplementation. Groups V, VI, and VII had the same supplementation levels as groups II, III, and IV, respectively, but with interval supplementation (continuous supplementation for 1 week, followed by a 1-week interval). The preliminary period lasted 7 days, and the formal experimental period lasted 53 days. Formal trial began with fasting weight measurement every 15 days in the morning. Experimental animals were housed individually in cages and managed by dedicated personnel, fed once daily at 08:00 and 15:00 with ad libitum access to feed and water.

1.3 Sample Collection

A digestion and metabolism trial was conducted from day 40 to day 43 of the formal experimental period. Six healthy minks with similar body weight and normal feed intake were selected from each group. Management during the digestion and metabolism trial was identical to daily management. Feces and urine were collected for 4 days using the total collection method. Before urine collection, 20 mL of 10% sulfuric acid was added to collection buckets to fix nitrogen. After the digestion and metabolism trial, urine was filtered and collected in 10 mL centrifuge tubes, then stored at -20°C for later use. All fecal samples were thoroughly mixed, and 200-300 g was taken, treated with 10% sulfuric acid, dried at 65°C, ground, and passed through a 40-mesh sieve for storage.

1.4 Measurements

The specific methods for determining dry matter, crude protein, ether extract, crude ash in experimental diets and fecal samples, and crude protein in urine samples followed the methods described by Zhang Liying [7]. Nutrient digestibility, nitrogen deposition, net protein utilization, and protein biological value were calculated using the following formulas:

Dry matter digestibility (%) = [(Dry matter intake - Dry matter excretion) / Dry matter intake] × 100
Crude protein digestibility (%) = [(Crude protein intake - Crude protein excretion) / Crude protein intake] × 100
Ether extract digestibility (%) = [(Fat intake - Fat excretion) / Fat intake] × 100
Nitrogen deposition (g/d) = Nitrogen intake - Fecal nitrogen - Urinary nitrogen
Net protein utilization (%) = (Deposited nitrogen / Nitrogen intake) × 100
Protein biological value (%) = [Nitrogen deposition / (Nitrogen intake - Fecal nitrogen)] × 100

1.5 Data Processing

Data were analyzed using SAS 9.1 statistical software for one-way ANOVA. Duncan' s multiple comparison test was used for significance analysis. GLM procedure was then used for multi-factor statistical analysis. Data are expressed as mean ± standard deviation, where $P>0.05$ indicates no significant difference, $P<0.05$ indicates significant difference, and $P<0.01$ indicates extremely significant difference.

2.1 Effects of CS on Growth Performance of Growing Minks

As shown in Table 2 , dietary CS supplementation had no significant effect on average daily gain ($P>0.05$). However, compared with group I, average daily gain in groups II-VII increased, with group V showing the highest value; the interval supplementation group had higher average daily gain than the continuous supplementation group. Dietary CS supplementation extremely significantly affected average daily feed intake and feed-to-gain ratio ($P<0.01$). Specifically, average daily feed intake in group V was extremely significantly lower than that in group II ($P<0.01$), and group VI was extremely significantly lower than groups I, II, and III ($P<0.01$). Group V had the lowest feed-to-gain ratio, which was extremely significantly lower than that of groups I, II, and IV ($P<0.01$). CS supplementation method also had an extremely significant effect on average daily feed intake and feed-to-gain ratio ($P<0.01$), with the interval supplementation group being extremely significantly lower than the continuous supplementation group ($P<0.01$). There was no significant interaction between CS supplementation method and level on growth performance ($P>0.05$).

2.2 Effects of CS on Nutrient Digestibility of Growing Minks

As shown in Table 3 , dietary CS supplementation significantly improved crude protein digestibility and ether extract digestibility ($P<0.05$), and extremely significantly improved dry matter digestibility ($P<0.01$). Specifically, dry matter digestibility in group V was extremely significantly higher than that in groups I and II ($P<0.01$). Crude protein digestibility in groups III, IV, V, VI, and VII was significantly higher than that in group I ($P<0.05$). Ether extract digestibility in groups III and V was significantly higher than that in groups I and II ($P<0.05$), with no significant differences from other groups ($P>0.05$). CS supplementation method significantly affected dry matter digestibility ($P<0.05$), with the interval supplementation group being extremely significantly higher than the continuous supplementation group ($P<0.01$). CS supplementation level had no significant effect on nutrient digestibility ($P>0.05$), and there was no significant interaction between CS supplementation method and level on nutrient digestibility ($P>0.05$).

2.3 Effects of CS on Nitrogen Metabolism of Growing Minks

As shown in Table 4 , dietary CS supplementation significantly or extremely significantly affected nitrogen metabolism indices ($P<0.05$ or $P<0.01$). Nitrogen intake in groups V, VI, and VII was extremely significantly lower than that in group II ($P<0.01$). Fecal nitrogen excretion in groups III, VI, and VII was extremely significantly lower than that in group I ($P<0.01$). Urinary nitrogen excretion in groups V, VI, and VII was extremely significantly lower than that in groups I, II, III, and IV ($P<0.01$). Nitrogen deposition in groups V, VI, and VII was significantly higher than that in group I ($P<0.05$); net protein utilization and protein biological value in groups V, VI, and VII were extremely significantly higher than those in group I ($P<0.01$), with the highest values observed in group V. CS supplementation method extremely significantly affected nitrogen intake, urinary nitrogen excretion, net protein utilization, and protein biological value ($P<0.01$), and significantly affected nitrogen deposition ($P<0.05$). Specifically, nitrogen intake and urinary nitrogen excretion in the interval supplementation group were extremely significantly lower than those in the continuous supplementation group (P<0.01); net protein utilization and protein biological value in the interval supplementation group were extremely significantly higher than those in the continuous supplementation group (P<0.01); nitrogen deposition in the interval supplementation group was significantly higher than that in the continuous supplementation group (P<0.05). CS supplementation level significantly affected fecal nitrogen excretion (P<0.05), with the 90 mg/kg group being significantly lower than the 60 mg/kg group (P<0.05). There was no significant interaction between CS supplementation method and level on nitrogen metabolism indices (P>0.05).

3.1 Effects of CS on Growth Performance of Growing Minks

Animal growth is a process influenced by multiple factors including genetic background, environmental conditions, nutritional supply, and metabolic status. These internal and external factors ultimately feed back to the hypothalamus and act on the growth axis to regulate animal growth [8]. As a growth regulator, CS promotes animal growth mainly through two pathways: first, by altering the molecular conformation of SS and reducing its immunological activity, thereby increasing GH levels in animals; second, by inhibiting dopamine-β-hydroxylase activity, causing dopamine accumulation in the body, which subsequently promotes GH synthesis in the hypothalamus. The combined effects of these two pathways improve animal growth performance [9]. The results of this experiment showed that although dietary CS supplementation did not significantly affect average daily gain, the CS-supplemented groups showed increased average daily gain compared with the control group, with group V showing the highest value—a 22.34% increase compared with group I. Average daily gain tended to decrease with increasing CS supplementation level, possibly because high supplementation levels caused digestive tract mucosal injury in minks, inhibiting growth performance. Cao Weiwei [10] found that appropriate or low levels of dietary CS could promote the development of glandular stomach and duodenum in Huainan partridge ducks, while high supplementation levels had no significant effect on duodenum development and even caused ulcers in the glandular stomach wall. Secondly, in this experiment, average daily gain in the interval supplementation group was 7.08% higher than that in the continuous supplementation group, which may be related to the time-dependent effects of CS, though the specific mechanism requires further investigation.

The results also showed that dietary CS supplementation could reduce average daily feed intake in minks, with the continuous supplementation group being extremely significantly lower than the interval supplementation group (8.46% reduction). Feed-to-gain ratio in all CS groups decreased to varying degrees compared with the control group, with group V showing the lowest value—a 16.19% reduction compared with the control group. Additionally, the interval supplementation group had lower feed-to-gain ratio than the continuous supplementation group, consistent with previous research on fur animals. Wang Zhongyan [11-12] reported that dietary CS supplementation could extremely significantly reduce feed intake and improve feed conversion rate in minks and silver foxes. Huang Hui et al. [13] showed that appropriate CS supplementation levels could increase average daily gain, reduce average daily feed intake, and improve feed utilization efficiency in raccoon dogs. The interval supplementation method was superior to continuous supplementation, and low supplementation levels were better than high levels, possibly because continuous high-level stimulation caused CS residues in animals, and as a chemical substance, excessive CS concentration might be toxic to animals, reducing growth performance.

3.2 Effects of CS on Nutrient Digestibility of Growing Minks

The results showed that dry matter digestibility in minks increased initially and then decreased with increasing CS supplementation level, with the 90 mg/kg level showing the best effect, and the interval supplementation group being extremely significantly higher than the continuous supplementation group. This trend was basically consistent with the reduction in dry matter intake. To meet their own needs, minks might improve dietary dry matter digestibility while reducing feed intake to ensure adequate nutrient acquisition from the diet. Yang et al. [14] found that low dietary CS supplementation levels could significantly increase the secretion and activity of protease and lipase in the small intestine and pancreas of broiler chicks, while high levels had inhibitory effects. Khomenoko et al. [15] found that CS could directly deplete SS, increase GH levels, and simultaneously relieve the inhibitory effects on various digestive enzymes, accelerate gastrointestinal motility, and enhance digestion, absorption, and anabolic metabolism. Du et al. [16] showed that dietary CS supplementation could promote gastrin secretion in weaned piglets. Gastrin is the main regulatory hormone for gastric acid secretion and also has a growth-promoting effect on gastric mucosa [17], ensuring gastrointestinal safety while increasing pepsin activity and promoting nutrient digestion and utilization. The results showed that crude protein digestibility and ether extract digestibility in CS-supplemented groups were higher than those in the control group to varying degrees, with group V showing the highest values—7.52% higher in crude protein digestibility and 2.20% higher in ether extract digestibility than the control group. The interval supplementation method was superior to continuous supplementation. Therefore, dietary CS supplementation may promote nutrient digestion and absorption by regulating intestinal endocrine function, improving gastrointestinal digestive enzyme activity, and promoting gastrointestinal motility, though the related mechanisms require further investigation.

3.3 Effects of CS on Nitrogen Metabolism of Growing Minks

Nitrogen metabolism can reflect protein metabolism status in the body, which is an important indicator affecting animal growth and development. The results showed that differences in feed intake among groups led to differences in nitrogen intake. Si Guoli [18] and Liu Yueqin et al. [19] found in sheep studies that dietary CS supplementation could significantly reduce fecal and urinary nitrogen excretion, increase nitrogen deposition, and high supplementation levels had inhibitory effects on nitrogen deposition. The results of this experiment also showed that CS-supplemented groups tended to have reduced fecal and urinary nitrogen excretion and increased nitrogen deposition compared with the control group, with group V showing the highest nitrogen deposition. Nitrogen deposition decreased with increasing CS supplementation level, and the interval supplementation method was superior to continuous supplementation, showing a 20% improvement, indicating that appropriate CS supplementation levels could promote protein synthesis and reduce protein degradation in minks. Protein synthesis in animals is influenced by multiple factors. Eisemann et al. [20] reported that GH could increase the rate of protein synthesis from amino acids and reduce amino acid oxidative decomposition, thereby increasing protein deposition. As an active substance in organisms, CS can reduce SS levels in the digestive tract and blood, increase the secretion of hormones such as GH, insulin-like growth factors (IGFs), and gastrin, as well as digestive enzymes such as pancreatic enzymes and pepsin, promoting enhanced anabolic metabolism [21]. Insulin-like growth factor I (IGF-I) and thyroid hormones [triiodothyronine (T3) and thyroxine (T4)] are also important factors affecting protein synthesis. Xu Yinxue et al. [22] showed that dietary CS supplementation could increase blood GH and IGF-I levels in nutrias. Bai Shiping et al. [23] reported that dietary CS supplementation could increase blood IGF-I, T3, and T4 levels in minks. Therefore, dietary CS supplementation may promote nitrogen metabolism by regulating the endocrine system. Net protein utilization and protein biological value are indicators measuring the degree of dietary protein utilization. The results showed that net protein utilization and protein biological value in CS-supplemented groups were higher than those in the control group, with group V showing the highest values. The interval supplementation group was superior to the continuous supplementation group, with net protein utilization 31.51% higher and protein biological value 29.82% higher than the continuous supplementation group, indicating that interval supplementation of 60 mg/kg CS in mink diets could promote dietary protein utilization. Research on the effects of CS on nitrogen metabolism is limited, and the mechanism of action requires further exploration. Researchers have conducted experiments on the application effects of CS in animal husbandry using yellow-feathered chickens, Northeast fine-wool sheep, lactating dairy cows, and beef cattle, obtaining appropriate CS levels and effect evaluations for different animals [24-27]. This experiment, for the first time in the growing stage of black short-haired minks and considering the advantages and disadvantages of supplementation methods, screened out appropriate supplementation levels, providing theoretical support and guidance for the rational use of CS in the fur animal field.

Conclusion

Dietary CS supplementation can promote mink growth, improve nutrient digestibility, and enhance nitrogen metabolism. Under the conditions of this experiment, the appropriate supplemental level of CS in diets for growing minks is 60 mg/kg, and the appropriate supplementation method is interval supplementation.

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