

Effects of Dietary Linoleic Acid Level on Growth Performance, Nutrient Digestibility, and Nitrogen Metabolism in Growing Blue Foxes (Post-print)

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Abstract

This experiment aimed to investigate the effects of dietary linoleic acid levels on growth performance, nutrient digestibility, and nitrogen metabolism in growing blue foxes. Sixty healthy male blue foxes at (90 ± 5) days of age with similar body weight were selected and randomly divided into 6 groups, with 10 replicates per group and 1 fox per replicate. The six groups of blue foxes were fed experimental diets with linoleic acid levels of 0.11% (Group I), 0.52% (Group II), 0.92% (Group III), 1.33% (Group IV), 2.14% (Group V), and 3.36% (Group VI). The pre-trial period was 7 days, and the formal period was 63 days. The results showed that: 1) Dietary linoleic acid level had extremely significant effects on average daily gain and feed-to-gain ratio in growing blue foxes ($P < 0.01$), but had no significant effect on dry matter intake ($P > 0.05$). As dietary linoleic acid level increased from 0.92% to 3.36%, average daily gain gradually increased and feed-to-gain ratio gradually decreased, but there were no significant differences in these indices between Groups IV and V ($P > 0.05$). 2) Dietary linoleic acid level had extremely significant effects on fat, carbohydrate, and dry matter digestibility in blue foxes ($P < 0.01$), but had no significant effect on protein digestibility ($P > 0.05$). Fat and carbohydrate digestibility were both highest in Group VI, and dry matter digestibility was highest in Group V, but there were no significant differences in these three indices between Groups IV and V ($P > 0.05$). 3) Dietary linoleic acid level had significant or extremely significant effects on fecal nitrogen, urinary nitrogen, and protein biological value ($P < 0.01$ or $P < 0.05$), but had no significant effect on nitrogen intake, nitrogen retention, and net protein utilization ($P > 0.05$). Fecal nitrogen was highest in Group VI, with no significant differences from Groups II, IV, and V ($P > 0.05$). Urinary nitrogen was highest in Group II, with no significant differences from Groups

III, IV, and V ($P>0.05$). Protein biological value was highest in Group VI, significantly higher than that in Group II ($P<0.05$), with no significant differences from other groups ($P>0.05$). From the perspective of reducing environmental pollution while ensuring growth performance in growing blue foxes, a dietary linoleic acid level of 2.14% was considered appropriate.

Full Text

Effects of Dietary Linoleic Acid Level on Growth Performance, Nutrient Digestibility and Nitrogen Metabolism of Blue Foxes in Growing Period

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Abstract

This study investigated the effects of dietary linoleic acid level on growth performance, nutrient digestibility, and nitrogen metabolism in growing blue foxes. Sixty healthy male blue foxes aged (90 ± 5) days with similar body weight were randomly allocated into six groups, each consisting of 10 replicates with one fox per replicate. The six groups were fed experimental diets containing 0.11% (Group), 0.52% (Group), 0.92% (Group), 1.33% (Group), 2.14% (Group), and 3.36% (Group) linoleic acid, respectively. The experiment included a 7-day adaptation period followed by a 63-day formal experimental period. The results showed that: (1) Dietary linoleic acid level significantly affected average daily gain and feed-to-gain ratio ($P<0.01$), but had no significant effect on dry matter intake ($P>0.05$). As dietary linoleic acid increased from 0.92% to 3.36%, average daily gain gradually increased while feed-to-gain ratio decreased, though no significant differences were observed between Groups and ($P>0.05$). (2) Dietary linoleic acid level significantly affected fat, carbohydrate, and dry matter digestibility ($P<0.01$), but had no significant effect on protein digestibility ($P>0.05$). Fat and carbohydrate digestibility were highest in Group , while dry matter digestibility peaked in Group , with no significant differences between Groups and for these three indices ($P>0.05$). (3) Dietary linoleic acid level significantly or highly significantly affected fecal nitrogen, urinary nitrogen, and protein biological value ($P<0.05$ or $P<0.01$), but had no significant effect on nitrogen intake, nitrogen retention, or net protein utilization ($P>0.05$). Fecal nitrogen was highest in Group , with no significant differences compared to Groups , , and ($P>0.05$). Urinary nitrogen was highest in Group , with no significant differences compared to Groups , , and ($P>0.05$). Protein biological value was highest in Group , significantly higher than in Group ($P<0.05$),

with no significant differences compared to other groups ($P>0.05$). Considering both environmental pollution reduction and maintenance of growth performance, a dietary linoleic acid level of 2.14% is recommended for growing blue foxes.

Keywords: linoleic acid; blue fox; growth performance; nutrient digestibility; nitrogen metabolism

Introduction

The blue fox (*Alopex lagopus*), also known as the Arctic fox, is a valuable fur-bearing animal prized for its thick, lustrous pelage used in various fur garments, collars, and ornaments. With a short production cycle and increasing market demand, blue fox farming has expanded considerably. Native to cold regions, blue foxes require adequate dietary fat to provide energy for thermoregulation. Fat constitutes an essential component of tissues and organs, serves as a crucial source and storage form of energy, and supplies essential fatty acids (EFA) that cannot be synthesized endogenously and must be provided through the diet.

Linoleic acid (LA), an essential fatty acid, plays vital physiological roles in animal metabolism. It serves as a precursor for bioactive substances, and deficiency can accelerate lipolysis in adipose tissue. Linoleic acid also reduces fat deposition, modulates lipid metabolism, inhibits tumor development and atherosclerosis, and enhances immune function. Studies in humans, rodents, and poultry have demonstrated that dietary linoleic acid supplementation can reduce body weight and decrease fatty acid and triglyceride levels in liver and adipose tissue. The NRC (1988) recommended approximately 1% linoleic acid for swine under ad libitum feeding conditions, emphasizing its critical role in maintaining normal growth and lipid metabolism in mice. Ip et al. reported that linoleic acid reduces body fat content while increasing muscle mass and can alter the ratio of saturated to unsaturated fatty acids, particularly the stearic acid to oleic acid ratio. Although blue foxes can digest high-fat diets, their specific linoleic acid requirements remain unreported, and domestic and international feeding standards have not established optimal dietary levels. This study was designed to determine the appropriate linoleic acid level for growing blue foxes by examining its effects on growth performance, nutrient digestibility, and nitrogen metabolism, thereby providing theoretical parameters for precise feed formulation in blue fox production.

Materials and Methods

1.1 Experimental Animals

Sixty healthy male blue foxes aged (90 ± 5) days with similar body weight were randomly selected for the feeding trial conducted at the Fur Animal Experimental Base of the Institute of Special Animal and Plant Sciences, Chinese Academy of Agricultural Sciences.

1.2 Experimental Diets

The basal diet was formulated using extruded corn, soybean meal, fish meal, meat and bone meal, corn gluten meal, dried distillers grains with solubles (DDGS), corn oil, and palmitic acid as primary ingredients, supplemented with mineral and vitamin premixes. Dietary linoleic acid levels were adjusted by varying the proportions of corn oil and palmitic acid to create six experimental diets containing 0.11%, 0.52%, 0.92%, 1.33%, 2.14%, and 3.36% linoleic acid. To prevent oil oxidation during hot weather, diets were prepared in batches and stored in cold storage, being fed fresh daily to maintain palatability and nutritional value. The composition and nutrient levels of experimental diets are presented in Table 1 .

1.3 Experimental Design

The 60 male blue foxes were randomly divided into six groups (10 replicates per group, one fox per replicate) and fed the experimental diets with linoleic acid levels of 0.11% (Group), 0.52% (Group), 0.92% (Group), 1.33% (Group), 2.14% (Group), and 3.36% (Group). The trial consisted of a 7-day adaptation period followed by a 63-day formal experimental period.

1.4 Feeding Management

All blue foxes were housed individually in cages measuring 100 cm \times 80 cm \times 80 cm. The entire experiment was conducted under natural outdoor lighting conditions with dedicated caretakers. Foxes were fed twice daily at 07:30 and 15:30 with ad libitum access to feed and water. Daily feed intake was recorded, and body weight was measured in the morning after overnight fasting at 90, 110, 123, 138, and 153 days of age. Health status was monitored and recorded daily.

1.5 Digestion and Metabolism Trial

The digestion and metabolism trial was conducted from August 27 to August 30, 2015. Eight healthy blue foxes with normal feed intake and fecal excretion were selected from each group. The total feces and urine collection method was employed over four consecutive days, with feeding management identical to the daily routine. Urine was collected daily, with 20 mL of 10% sulfuric acid added to collection containers to preserve nitrogen. Fresh feces were weighed daily, and 10% sulfuric acid equivalent to 5% of fresh weight was added before

storage at -20 °C. After the 4-day collection period, urine and fecal samples were thoroughly mixed. Fecal samples were sterilized at 80 °C for 2 hours, then dried to constant weight at 65 °C, ground to pass through a 40-mesh sieve, and prepared as air-dried samples for laboratory analysis.

1.6 Measurements and Methods

Experimental diets and fecal samples were analyzed for dry matter, crude protein, crude fat, crude ash, calcium, and phosphorus content. Carbohydrate content and metabolizable energy were calculated. Dry matter was determined by oven-drying at 105 °C (GB/T 6435-2006). Crude protein was measured by the Kjeldahl method (GB/T 6432-1994). Crude fat was analyzed by Soxhlet extraction (GB/T 6433-2006). Crude ash was determined by combustion at 550 °C (GB/T 6432-1992). Calcium content was measured by EDTA complexometric titration (GB/T 6436-1992). Phosphorus content was determined by ammonium vanadate-molybdate colorimetry (GB/T 6437-1992). Dietary linoleic acid levels were analyzed by gas chromatography-mass spectrometry (Agilent 7890A-7000B).

1.7 Calculation Formulas

Average daily gain (g/d) = (final body weight - initial body weight) / number of trial days

Average daily feed intake (g/d) = total feed intake during trial period / number of trial days

Feed-to-gain ratio = average daily feed intake / average daily gain

Dry matter digestibility (%) = [(dry matter intake - dry matter excretion) / dry matter intake] × 100

Protein digestibility (%) = [(protein intake - protein excretion) / protein intake] × 100

Fat digestibility (%) = [(fat intake - fat excretion) / fat intake] × 100

Carbohydrate digestibility (%) = [(carbohydrate intake - carbohydrate excretion) / carbohydrate intake] × 100

Nitrogen retention (g/d) = nitrogen intake - fecal nitrogen - urinary nitrogen

Net protein utilization (%) = (nitrogen retention / nitrogen intake) × 100

Protein biological value (%) = [nitrogen retention / (nitrogen intake - fecal nitrogen)] × 100

Statistical Analysis

Data were expressed as “mean ± standard deviation,” organized using Excel, and analyzed using the GLM procedure in SAS 8.0 software. Duncan’s multiple range test was used for post-hoc comparisons. Differences were considered significant at P<0.05 and highly significant at P<0.01.

Results

2.1 Effects of Dietary Linoleic Acid Level on Growth Performance of Growing Blue Foxes

As shown in Table 2, dietary linoleic acid level highly significantly affected daily gain during 90-110 days, 110-123 days, and 123-138 days of age ($P < 0.01$). When dietary linoleic acid ranged from 0.92% to 3.36%, daily gain at each age stage showed an increasing trend with elevated linoleic acid levels, with Group achieving the highest daily gain at all stages.

Table 3 demonstrates that dietary linoleic acid level highly significantly affected average daily gain and feed-to-gain ratio ($P < 0.01$), but had no significant effect on dry matter intake ($P > 0.05$). With dietary linoleic acid increasing from 0.92% to 3.36%, average daily gain showed an upward trend. Groups and were highly significantly lower than Groups and ($P < 0.01$), but did not differ significantly from Groups and ($P > 0.05$). The feed-to-gain ratio was highest in Group and lowest in Group. Groups and were highly significantly higher than Groups and ($P < 0.01$), but showed no significant differences compared to Groups and ($P > 0.05$). No significant differences were observed among Groups, , and ($P > 0.05$).

2.2 Effects of Dietary Linoleic Acid Level on Nutrient Digestibility of Growing Blue Foxes

Table 4 reveals that dietary linoleic acid level highly significantly affected fat, carbohydrate, and dry matter digestibility ($P < 0.01$), but had no significant effect on protein digestibility ($P > 0.05$). Fat digestibility increased with dietary linoleic acid level, peaking in Group and reaching its lowest value in Group. Group was highly significantly lower than Groups, , and ($P < 0.01$), but did not differ significantly from Groups and ($P > 0.05$). No significant differences were found among Groups, , and ($P > 0.05$). Carbohydrate digestibility was highest in Group and lowest in Group, with Groups, , and being highly significantly lower than Groups, , and ($P < 0.01$). Dry matter digestibility was highest in Group and lowest in Group, with Group being highly significantly lower than Groups, , and ($P < 0.01$), but showing no significant differences compared to Groups and ($P > 0.05$). No significant differences were observed among the other groups ($P > 0.05$).

2.3 Effects of Dietary Linoleic Acid Level on Nitrogen Metabolism of Growing Blue Foxes

Table 5 shows that dietary linoleic acid level significantly affected fecal nitrogen and protein biological value ($P < 0.05$) and highly significantly affected urinary nitrogen ($P < 0.01$), but had no significant effects on nitrogen intake, nitrogen retention, or net protein utilization ($P > 0.05$). Fecal nitrogen was highest in Group and lowest in Group, with Group being significantly lower than Groups and ($P < 0.05$), but showing no significant differences compared to

Groups , , and ($P>0.05$). Urinary nitrogen was highest in Group , highly significantly higher than Groups and ($P<0.05$), but not significantly different from Groups , , and ($P>0.05$). Protein biological value was highest in Group , significantly higher than Group ($P<0.05$), with no significant differences compared to other groups ($P>0.05$).

Discussion

3.1 Effects of Dietary Linoleic Acid Level on Growth Performance of Growing Blue Foxes

Corn oil, rich in linoleic acid (approximately 40% content), was used as the linoleic acid source in this study. Previous research has demonstrated that linoleic acid, as an essential fatty acid, promotes animal growth. Thiel-Cooper et al. reported that increasing dietary conjugated linoleic acid from 0.12% to 1.00% improved average daily gain and reduced feed-to-gain ratio in pigs. Jin Yinghai found that safflower oil supplementation (primarily linoleic acid) significantly increased average daily gain and decreased feed-to-gain ratio in finishing pigs. Our results align with these findings, showing that dietary linoleic acid level significantly affected average daily gain and feed-to-gain ratio in growing blue foxes. Foxes fed 3.36% linoleic acid achieved the highest average daily gain and lowest feed-to-gain ratio, though these did not differ significantly from those fed 2.14% linoleic acid. From an economic perspective, a dietary linoleic acid level of 2.14% appears sufficient to meet growth requirements. However, some studies reported no significant effects of linoleic acid supplementation on performance in laying hens, ducks, or 2-month-old meat rabbits, possibly due to differences in supplementation forms and physiological stages, warranting further investigation.

3.2 Effects of Dietary Linoleic Acid Level on Nutrient Digestibility of Growing Blue Foxes

Our results indicate that fat digestibility in growing blue foxes increased gradually with dietary linoleic acid level. Austreng et al. demonstrated in mink that fat digestibility depends on the ratio of saturated to unsaturated fatty acids, with unsaturated fatty acids being more digestible than saturated ones. Jin Yinghai similarly reported increasing fat digestibility with higher dietary linoleic acid levels. These collective findings suggest that linoleic acid enhances fat digestibility. Although dietary linoleic acid level did not significantly affect protein digestibility, carbohydrate digestibility showed an increasing trend, possibly because fat digestion and absorption indirectly influenced carbohydrate utilization in growing blue foxes. Appropriate dietary linoleic acid levels may promote carbohydrate absorption. Dry matter digestibility initially decreased then increased with dietary linoleic acid level, and high dietary linoleic acid levels (1.33%-3.36%) increased dry matter intake, suggesting that higher corn

oil proportions may improve diet palatability and consequently enhance feed intake in blue foxes.

3.3 Effects of Dietary Linoleic Acid Level on Nitrogen Metabolism of Growing Blue Foxes

Nitrogen metabolism reflects protein utilization in the body. After consuming diets, nitrogen-containing compounds are digested, with a portion used for protein synthesis and the remainder excreted via feces and urine to maintain nitrogen balance. Fecal nitrogen represents unabsorbed nitrogen largely influenced by dietary protein levels, while urinary nitrogen comprises nitrogen from absorbed amino acids metabolized by tissues, primarily affected by amino acid balance. Li Ronggang et al. found that linoleic acid supplementation did not significantly alter nitrogen apparent digestibility, nitrogen apparent metabolic rate, or protein biological value in weaned to 2-month-old meat rabbits. Wang Shengwei et al. reported that appropriate dietary linoleic acid levels improved nitrogen metabolism and increased nitrogen deposition in Italian honeybee larvae. In our study with blue foxes, dietary linoleic acid level significantly affected protein biological value but not nitrogen retention or net protein utilization. Nitrogen retention, net protein utilization, and biological value were all highest at 3.36% linoleic acid, possibly due to linoleic acid's involvement in nitrogen metabolism and promotion of nitrogen deposition. Considering both feed cost reduction and maintenance of growth performance, a dietary linoleic acid level of 2.14% is recommended for growing blue foxes.

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