Effects of Betaine and L-Carnitine on Growth Performance, Slaughter Performance, Organ Development, and Fat Deposition in Mongolian-Cold Hybrid Sheep: Postprint

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Abstract

This study aimed to investigate the effects of betaine and L-carnitine on growth performance, slaughter performance, organ development, and fat deposition in Mongolian-Han hybrid sheep. The experiment adopted a single-factor design. A total of 270 Mongolian-Han hybrid sheep with a body weight of (29.0±0.4) kg were randomly allocated into 3 groups, with 3 replicates per group and 30 sheep per replicate. The control group (CON) was fed a basal diet, while the betaine group (BN) and L-carnitine group (L-CN) received the basal diet supplemented with 1‰ betaine and 0.4‰ L-carnitine, respectively. The pre-trial period was 15 days, and the formal trial period was 40 days. The results showed: 1) The average daily feed intake of the L-CN group was significantly lower than that of the CON and BN groups ($P<0.05$), with no significant differences in other growth performance indices among groups ($P>0.05$). 2) No significant differences were observed among groups in pre-slaughter live weight, carcass weight, dressing percentage, eye muscle area, or carcass fat content ($P>0.05$). 3) No significant differences were found in visceral organ development indices among groups ($P>0.05$). 4) The large intestine weight and its proportion to pre-slaughter live weight in the L-CN group were significantly lower than those in the CON group ($P<0.05$), with no significant differences in other gastrointestinal development indices among groups ($P>0.05$). 5) The total fat weight and its proportion to pre-slaughter live weight in both BN and L-CN groups were significantly lower than those in the CON group ($P<0.05$), while the L-CN group had significantly higher total fat weight and its proportion to pre-slaughter live weight than the BN group ($P<0.05$). The tail fat weight and its proportion to pre-slaughter live weight in the CON group were significantly higher than those in the BN group ($P<0.05$). No significant differences were detected among groups in rumen fat, intestinal fat, pericardial fat, or perirenal fat weights and their proportions to pre-slaughter live weight (P>0.05). In conclusion, dietary supplementation with betaine and L-carnitine both reduced fat deposition in Mongolian-Han hybrid sheep, primarily decreasing tail fat weight, indicating that betaine and L-carnitine have certain effects on reducing body fat in Mongolian-Han hybrid sheep.

Full Text

Effects of Betaine and L-Carnitine on Growth Performance, Slaughter Performance, Organ Development and Fat Deposition of Mongolian Hybrid Sheep

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Abstract

This study investigated the effects of betaine and L-carnitine on growth performance, slaughter performance, organ development, and fat deposition in Mongolian hybrid sheep. A single-factor experimental design was employed with 270 Mongolian hybrid sheep (body weight: 29.0±0.4 kg) randomly allocated into three groups, each consisting of three replicates of 30 sheep. The control group (CON) received a basal diet, while the betaine group (BN) and L-carnitine group (L-CN) received the basal diet supplemented with 1‰ betaine and 0.4‰ L-carnitine, respectively. The pre-trial period lasted 15 days, followed by a 40-day experimental period.

The results showed: (1) The average daily feed intake (ADFI) of the L-CN group was significantly lower than that of the CON and BN groups (P<0.05), while other growth performance indicators showed no significant differences among groups (P>0.05). (2) No significant differences were observed among groups in pre-slaughter live weight, carcass weight, dressing percentage, eye muscle area, or GR value (P>0.05). (3) No significant differences were found in visceral organ development indices among groups (P>0.05). (4) The large intestine weight and its proportion of pre-slaughter live weight in the L-CN group were significantly lower than those in the CON group (P<0.05), while other gastrointestinal development indices showed no significant differences among groups (P>0.05). (5) The total fat weight and its proportion of pre-slaughter live weight in both the BN and L-CN groups were significantly lower than those in the CON group ($P<0.05$), with the L-CN group showing significantly higher values than the BN group ($P<0.05$). The tail fat weight and its proportion of pre-slaughter live weight in the CON group were significantly higher than those in the BN group ($P<0.05$). No significant differences were detected among groups in rumen fat, intestinal fat, pericardial fat, or perirenal fat weights and their proportions of pre-slaughter live weight ($P>0.05$).

These findings indicate that dietary supplementation with betaine and L-carnitine reduced fat deposition in Mongolian hybrid sheep, primarily by decreasing tail fat weight, suggesting that both additives may play a role in reducing body fat in this breed.

Keywords: Mongolian hybrid sheep; betaine; L-carnitine; growth performance; slaughter performance; fat deposition

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Introduction

With improving living standards, consumers have become increasingly concerned about meat quality. However, most sheep production systems in China rely on intensive feeding, where high-concentrate diets are commonly used to achieve rapid weight gain. Combined with reduced physical activity compared to grazing systems, this management approach often results in excessive fat deposition at slaughter, leading to decreased meat quality and increased health risks for consumers, including higher incidence of diabetes and cardiovascular diseases. Consequently, exploring nutritional strategies to reduce excessive fat deposition in intensively fed sheep has become a research priority.

Several green and safe functional additives have shown promise in livestock production, promoting growth while improving lean meat accretion and reducing fat deposition. Betaine, chemically known as trimethylglycine, is a quaternary ammonium compound with a trimethyl structure that serves as a multifunctional nutritional feed additive. In animal production, betaine has been reported to maintain vitamin efficacy, alleviate stress, promote growth, and improve meat quality. Previous research suggests that betaine’ s role as an effective methyl donor gives it advantages in regulating fat metabolism, potentially by increasing peripheral fat breakdown or reducing fat synthesis. Studies in finishing pigs have demonstrated that betaine and L-carnitine supplementation can reduce backfat thickness and increase eye muscle area.

L-carnitine, a vitamin-like nutrient that is non-toxic and harmless, exists in L- and D-forms, with the L-form being physiologically significant and widely distributed in animal and plant tissues. Its chemical structure resembles choline, and its physiological properties are similar to amino acids. As an essential carrier for long-chain fatty acid -oxidation, L-carnitine promotes fatty acid oxidation and plays an important role in amino acid metabolism, thereby enhancing protein deposition. Additionally, L-carnitine facilitates the absorption of fat-soluble vitamins, calcium, and phosphorus; promotes ATP transport to the mitochondrial inner membrane; stimulates digestive juice secretion; improves nutrient utilization; and enhances metabolism, growth, and disease resistance.

While previous studies have demonstrated the beneficial effects of these additives on growth performance and fat metabolism in other animal species, their effects on sheep, particularly regarding fat deposition, remain poorly documented. Therefore, this experiment was designed to evaluate the effects of betaine and L-carnitine supplementation on growth performance, slaughter performance, organ development, and fat deposition in Mongolian hybrid sheep, with the ultimate goal of reducing fat deposition and providing a basis for practical application in sheep production.

1.1 Experimental Time and Location

The experiment was conducted at the research base of the Inner Mongolia Hetao Institute of Agricultural and Animal Husbandry Technology from October to December 2015, lasting 55 days (15-day pre-trial period and 40-day experimental period).

1.2 Experimental Materials

Betaine: Anhydrous betaine with 98% purity was added at 1‰ (dry matter basis) throughout the entire experimental period.

L-carnitine: A commercial product named Kangliwang containing 50% active L-carnitine was added at 0.4‰ (dry matter basis) throughout the entire experimental period.

1.3 Experimental Design

A single-factor randomized design was employed using 270 Mongolian hybrid sheep (75 days old, body weight: 29.0±0.4 kg) divided into three groups with similar average body weights. Each group contained three replicates of 30 sheep. The control group (CON) received the basal diet, while the betaine group (BN) and L-carnitine group (L-CN) received the basal diet supplemented with 1‰ betaine and 0.4‰ L-carnitine, respectively. The basal diet was formulated to meet the nutritional requirements of meat-type hybrid sheep at the 30–40 kg body weight stage. The concentrate was pelleted, and roughage was mixed using a total mixed ration (TMR) mixer. Premix was provided by Beijing Precision Animal Nutrition Research Center. The composition and nutrient levels of the basal diet are presented in Table 1 .

1.4 Management Practices

Sheep were housed in separate pens by replicate, with pens disinfected every 15 days. During the 15-day pre-trial period, all sheep were tagged, vaccinated, and dewormed. Throughout the 40-day experimental period, feed was provided at 05:00 and 16:00 daily, with concentrate offered first followed by mixed roughage at a concentrate-to-roughage ratio of 52.5:47.5 (air-dry basis). Sheep had free access to water, and any health issues were promptly treated. Pens were cleaned daily to maintain hygiene. Body weight was recorded at the start and end of the experiment and every 15 days thereafter. Residual feed was collected every three days starting from day one of the experimental period, and feed allowance was adjusted to maintain approximately 10% refusals. On day 55, five sheep per group (15 total) with body weights close to the group average were selected for slaughter to determine slaughter performance, organ development, and fat deposition indices.

1.5 Measurement Indices

1.5.1 Growth Performance Initial body weight (IBW) was recorded on day 1 of the experimental period, and final body weight (FBW) was recorded on day 40. Body weight was measured every 15 days throughout the trial. Daily feed allowance was recorded, and residual feed was collected and weighed every three days. These data were used to calculate average daily gain (ADG), average daily feed intake (ADFI), and feed conversion ratio (FCR).

ADFI = (Total feed offered - Total residual feed) / Number of feeding days
ADG = (Final body weight - Initial body weight) / Number of feeding days
FCR = ADFI / ADG

1.5.2 Determination of Slaughter Performance, Organ Indices, and Fat Deposition On day 55, sheep from each group were mixed, and five healthy sheep with body weights close to the group average were selected from each group (15 total) for slaughter. Sheep were weighed at 16:00, then fasted for 16 hours with water restriction before being reweighed prior to slaughter at 07:00 the following day. After stunning, sheep were slaughtered by jugular venesection.

Pre-slaughter live weight was recorded for each sheep. After slaughter, the head, hooves, and viscera were removed, and the carcass weight, internal organ weights, and various fat depot weights were measured. The digestive tract was emptied and thoroughly rinsed before weighing the rumen, reticulum, omasum, abomasum, small intestine, and large intestine.

Key measurement methods and calculation formulas were as follows:

Carcass weight (kg): The weight of the carcass after blood removal, skinning, head removal (at the atlas-axis joint), removal of lower limb bones and distal portions, and evisceration (with kidneys and perirenal fat retained).

Eye muscle area: The cross-section of the longissimus dorsi muscle between the last and second-to-last ribs was traced on transparent paper, and the area was calculated using the formula: Eye muscle area (cm^2) = Height × Width × 0.7.

Carcass fat content value (GR value): Measured as the tissue thickness 11 cm from the dorsal midline between the 12th and 13th ribs using vernier calipers, serving as an indicator of carcass fat content.

Dressing percentage (%) = (Carcass weight / Pre-slaughter live weight) × 100

1.6 Data Processing and Analysis

Experimental data were organized using Excel 2007 and analyzed using the ANOVA procedure in SPSS 18.1 software for one-way analysis of variance. Duncan' s multiple comparison test was used when significant differences were detected. Differences were considered significant at $P<0.05$.

Results

2.1 Effects of Betaine and L-Carnitine on Growth Performance of Mongolian Hybrid Sheep

As shown in Table 2 , the ADFI of the L-CN group was significantly lower than that of the CON and BN groups ($P<0.05$), while no significant difference was observed between the BN and CON groups ($P>0.05$). Both the L-CN and BN groups exhibited slightly lower ADG compared to the CON group, but the differences were not significant ($P>0.05$). The CON group showed lower FCR than the L-CN and BN groups, though this difference was not statistically significant ($P>0.05$). These results indicate that dietary betaine and L-carnitine supplementation did not significantly affect weight gain performance or feed conversion efficiency, but L-carnitine significantly reduced ADFI.

2.2 Effects of Betaine and L-Carnitine on Slaughter Performance of Mongolian Hybrid Sheep

Table 3 shows that no significant differences were observed among groups in pre-slaughter live weight, carcass weight, dressing percentage, eye muscle area, or GR value ($P>0.05$). However, among the three groups, the CON group had the smallest eye muscle area and the largest GR value.

2.3 Effects of Betaine and L-Carnitine on Internal Organ Development of Mongolian Hybrid Sheep

As presented in Table 4 , the L-CN group had greater heart weight than the CON group, while other internal organ weights were lower in both the BN and L-CN groups compared to the CON group. However, no significant differences were detected in internal organ weights among the three groups ($P>0.05$). Similarly, no significant differences were found in the proportions of internal organ weights to pre-slaughter live weight among the CON, BN, and L-CN groups ($P>0.05$), indicating that dietary betaine and L-carnitine supplementation did not significantly affect internal organ development.

2.4 Effects of Betaine and L-Carnitine on Gastrointestinal Development of Mongolian Hybrid Sheep

Table 5 reveals that the large intestine weight and its proportion of pre-slaughter live weight in the CON group were significantly higher than those in the L-CN group ($P<0.05$), while the BN group showed no significant differences from either the CON or L-CN groups ($P>0.05$). Although the CON group had higher rumen, reticulum, omasum, abomasum, and total stomach weights compared to the BN and L-CN groups, no significant differences were observed in the weights of individual stomach compartments among groups ($P>0.05$). No significant differences were detected among groups in the proportions of rumen, reticulum, omasum, or abomasum weights to total stomach weight or to pre-slaughter live weight, nor in total stomach weight or its proportion of pre-slaughter live weight ($P>0.05$). Additionally, no significant differences were found in small intestine weight or its proportion of pre-slaughter live weight among groups ($P>0.05$). These results suggest that dietary betaine and L-carnitine supplementation did not significantly affect stomach and small intestine development in Mongolian hybrid sheep, though L-carnitine may have inhibited large intestine development.

2.5 Effects of Betaine and L-Carnitine on Fat Deposition in Mongolian Hybrid Sheep

Table 6 demonstrates that tail fat weight followed the order CON group > L-CN group > BN group, with the CON group being significantly higher than the BN group ($P<0.05$). The proportion of tail fat weight to pre-slaughter live weight was also significantly higher in the CON group compared to the BN group ($P<0.05$). Total fat weight in the CON group was significantly higher than in both the BN and L-CN groups ($P<0.05$), while the L-CN group was significantly higher than the BN group ($P<0.05$). The proportion of total fat weight to pre-slaughter live weight ranked as CON group > L-CN group > BN group, with the CON group significantly higher than both the BN and L-CN groups ($P<0.05$), and the L-CN group significantly higher than the BN group ($P<0.05$). No significant differences were observed among groups in intestinal fat, rumen fat, pericardial fat, or perirenal fat weights ($P>0.05$), nor in the proportions of these fat depots to total fat weight or pre-slaughter live weight ($P>0.05$). However, the CON group generally had higher weights for these four fat depots compared to the BN and L-CN groups, except for perirenal fat. These findings indicate that both betaine and L-carnitine reduced fat deposition in Mongolian hybrid sheep, with betaine showing superior fat-reducing effects.

Discussion

3.1 Effects of Betaine and L-Carnitine on Growth Performance of Mongolian Hybrid Sheep

Betaine and L-carnitine are alkaloids that participate in nutrient metabolism, enhance animal growth performance, and improve fat metabolism, classifying them as novel nutrient repartitioning agents. Betaine promotes animal growth primarily by participating in methyl metabolism and playing a crucial role in the methylation cycle that regenerates methionine from homocysteine. Although numerous studies have reported growth-promoting effects of betaine, Cui et al. found that betaine supplementation did not significantly improve growth performance in meat sheep, a result consistent with our findings. This discrepancy may be attributed to two factors: first, partial degradation of betaine to trimethylamine in the rumen, and second, the relatively high protein level in our experimental diet, which may have prevented betaine from exerting its methionine-sparing effect.

Reports on L-carnitine' s growth-promoting effects have been inconsistent. Rincker et al. found that supplementing piglet diets with 50–100 mg/kg L-carnitine improved average daily gain and feed-to-gain ratio, suggesting beneficial effects on weaned piglet performance. Conversely, Owen et al. reported that L-carnitine supplementation reduced average daily feed intake and average daily gain in growing-finishing pigs at various stages. Ding' s research on Tan sheep yielded similar results, with L-carnitine reducing ADG by 5.58% over a 60-day period and by 7.53% during the first 30 days. Our results align with these latter findings, as the L-CN group showed a 5.96% reduction in ADG and a 3.29% reduction in ADFI compared to the CON group. Two possible explanations exist: first, since ADFI and ADG are positively correlated, L-carnitine may have reduced ADG by decreasing feed intake, possibly due to adverse effects on diet palatability; second, under our dietary conditions, L-carnitine may have enhanced fatty acid -oxidation, thereby reducing fat deposition and consequently decreasing weight gain.

3.2 Effects of Betaine and L-Carnitine on Slaughter Performance of Mongolian Hybrid Sheep

Slaughter performance directly reflects animal growth performance and serves as a crucial economic indicator. In our study, no significant differences were observed among groups in pre-slaughter live weight, carcass weight, or dressing percentage, indicating relatively consistent meat production performance during this growth stage. Using our team' s nutrient requirements for 30–40 kg sheep, dressing percentages were 43.50%, 42.44%, and 43.5% for the CON, BN, and L-CN groups, respectively, which are comparable to values reported by Wan et al. for Dorper × thin-tailed Han crossbred sheep at similar weights. This suggests that Mongolian hybrid sheep achieve similar dressing percentages to Dorper × thin-tailed Han sheep under equivalent nutritional levels.

Eye muscle area and GR value are important indicators of lamb carcass quality and fat content, with larger eye muscle area indicating higher lean meat percentage and higher GR value indicating greater fat content. Our results showed GR values ranked as CON > L-CN > BN, with both the L-CN and BN groups showing reduced GR values compared to the CON group, while both groups had larger eye muscle areas than the CON group. Huang et al. reported that dietary betaine supplementation (1,250 mg/kg) increased lean meat percentage by 5.2% and eye muscle area by 17.6% in growing pigs. These findings suggest that betaine and L-carnitine supplementation may improve lean meat percentage and reduce fat content in sheep, with the BN group showing the lowest GR value and largest eye muscle area, indicating potentially the highest lean meat percentage.

3.3 Effects of Betaine and L-Carnitine on Internal Organ and Gastrointestinal Development of Mongolian Hybrid Sheep

Internal organ weights reflect overall animal condition to some extent. Our results showed no significant differences between the L-CN and BN groups and the CON group in liver, lung, or spleen weights or their proportions of pre-slaughter live weight, indicating that betaine and L-carnitine supplementation did not affect internal organ development when nutritional levels were consistent. This indirectly demonstrates that nutritional level is a key factor influencing organ development.

Gastrointestinal development, particularly rumen development, directly affects feed intake and digestion capacity, ultimately determining future growth performance. Our results showed no significant differences among groups in the weights or proportions of individual stomach compartments or total stomach weight relative to pre-slaughter live weight, indicating that stomach and small intestine development remained coordinated with overall body growth in Mongolian hybrid sheep. Notably, the L-CN group exhibited significantly reduced large intestine weight and its proportion of pre-slaughter live weight compared to the CON group. Ji et al. reported a positive correlation between dry matter intake and fecal dry matter output. Combined with our findings, this suggests that reduced feed intake in the L-CN group may have decreased fecal output, consequently reducing large intestine function and weight.

3.4 Effects of Betaine and L-Carnitine on Fat Deposition in Mongolian Hybrid Sheep

Most sheep breeds in China are fat-tailed, an evolutionary adaptation that enables energy storage during feed scarcity to ensure survival. Our experimental sheep belong to this category, characterized by substantial tail fat deposition. In this study, we evaluated fat deposition using weights of rumen fat, intestinal fat, pericardial fat, perirenal fat, tail fat, and total fat. Anatomically, tail fat represented the largest proportion of body fat, accounting for 41–57% of total fat (excluding subcutaneous fat), confirming that the tail is the primary fat storage site in Mongolian sheep. Similar findings were reported for Xinjiang Maigaiti sheep, another fat-tailed breed, where tail fat exceeded 50% of total fat weight.

Our results also showed that betaine and L-carnitine supplementation did not significantly affect visceral fat depots (rumen, intestinal, pericardial, and perirenal fat), likely because these fats serve protective and cushioning functions for internal organs and remain relatively stable within certain ranges. In contrast, tail fat serves as an energy reserve, and its deposition level does not critically affect survival, making it more responsive to dietary interventions.

The selection of fat-tailed Mongolian hybrid sheep was intended to maximize the detection of fat-reducing and lean-enhancing effects. Our results clearly demonstrate that both additives significantly affected fat deposition. Betaine was initially used in poultry and aquaculture feeds to replace methionine and choline, but its fat-reducing effects were later discovered in swine production. Our study provides the first evidence of betaine’ s fat-reducing effects in sheep, with the BN group showing a 989.6 g reduction in total fat weight (38.7% decrease) and a 2.59 percentage unit reduction in total fat proportion relative to pre-slaughter live weight (37.2% decrease) compared to the CON group. These results align with findings from other species. Wang et al. reported that supplementing finishing pigs with 1,000 mg/kg betaine reduced backfat thickness and leaf fat weight by 14.81% and 25.53%, respectively.

Betaine may influence lipid metabolism through several pathways: (1) enhancing betaine-homocysteine methyltransferase activity to promote methionine synthesis from homocysteine, providing methyl groups for carnitine synthesis and thereby increasing fatty acid -oxidation; (2) increasing hormone-sensitive lipase (HSL) activity while decreasing fatty acid synthase (FAS), malate dehydrogenase (MDH), and lipoprotein lipase (LPL) activities, thereby accelerating fat breakdown and inhibiting fat synthesis; and (3) modulating lipid metabolism hormonally by increasing leptin and insulin levels, which enhances fat catabolism.

L-carnitine also reduced fat deposition in our study, with the L-CN group showing a 578.4 g reduction in total fat weight and a 1.4 percentage unit decrease in total fat proportion compared to the CON group. Similar results were reported by Ding in Tan sheep, where 200 mg/kg L-carnitine supplementation significantly reduced tail fat weight by 31.4%. L-carnitine’ s fat-reducing effect stems from its role as the sole carrier for transporting fatty acids into the mitochondrial matrix for oxidation, making it a key regulator of fatty acid oxidation. Wall et al. suggested that L-carnitine supplementation increases free carnitine levels, which combine with acetyl-CoA to form acetylcarnitine, reducing acetyl-CoA accumulation and relieving inhibition of transferase enzymes, thereby facilitating fatty acid oxidation. The rate of -oxidation is determined by L-carnitine concentration, with higher concentrations promoting faster fat oxidation and utilization.

Both betaine and L-carnitine influence fat deposition through L-carnitinemediated fatty acid oxidation; however, our data suggest betaine' s fat-reducing effect was more pronounced, likely due to additional synergistic pathways involving enzyme activity modulation and hormonal regulation.

Conclusions

1. Dietary supplementation with betaine or L-carnitine did not significantly affect the growth performance of Mongolian hybrid sheep.

2. Both betaine and L-carnitine supplementation reduced fat deposition in Mongolian hybrid sheep, with betaine showing superior efficacy.

3. The most pronounced effect of betaine and L-carnitine supplementation was on reducing tail fat deposition.

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