Effects of Concentrate Supplementation on Mutton Quality of Tibetan Pengbo Semi-fine Wool Sheep (Postprint)

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Abstract

This experiment aimed to investigate the effects of concentrate supplementation on the meat quality of Tibetan Pengbo semi-fine wool sheep. Thirty adult Pengbo semi-fine wool ewes were selected and randomly divided into 3 groups according to the principle of homogeneity: grazing without supplementation (control, G1) group, concentrate supplementation 200 g/d (G2) group, and concentrate supplementation 400 g/d (G3) group, with 10 animals per group. Supplementation was conducted daily at 19:30, and the formal experimental period was 75 d. At the end of the formal experimental period, slaughter was conducted and the longissimus dorsi muscle was collected to determine nutrient content and histological characteristics. The results showed that: high-level concentrate supplementation increased the total dry matter intake of ewes, and the average daily gain of G3 group was significantly higher than that of G1 and G2 groups ($P<0.05$); concentrate supplementation had no significant effect on mutton pH1 h, pH24 h, drip loss, and moisture and crude ash content ($P>0.05$), but the cooking yield, crude protein content, intramuscular fat content, and muscle fiber density of G3 group were significantly higher than those of G1 group ($P<0.05$); the total amino acids, essential amino acids, and aspartic acid content of mutton in G2 and G3 groups were significantly higher than those in G1 group ($P<0.05$), the phenylalanine and lysine content of G3 group were significantly higher than those of G1 group ($P<0.05$), and the threonine and glutamic acid content of G3 group tended to be higher than those of G1 group ($0.05\leq P<0.10$); concentrate supplementation had no significant effect on saturated fatty acids, monounsaturated fatty acids, and polyunsaturated fatty acids content of mutton ($P>0.05$), and the C18:1 trans-9 content of G3 group tended to be lower than that of G1 group ($0.05\leq P<0.10$). The above results indicate that concentrate supplementation can significantly improve the nutritional value of Pengbo semi-fine wool sheep mutton and enhance meat tenderness, and supplementation at 400 g/d per head was superior to 200 g/d.

Full Text

Effects of Concentrate Supplementation on Meat Quality of Pengbo Semi-Fine Wool Sheep in Tibet

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Abstract

This study investigated the effects of concentrate supplementation on meat quality of Pengbo semi-fine wool sheep in Tibet. Thirty adult Pengbo semi-fine wool ewes were randomly allocated to three groups (n=10) according to homogeneous principle: grazing without supplementation (G1, control), grazing plus 200 g/d concentrate supplementation (G2), and grazing plus 400 g/d concentrate supplementation (G3). Supplementation was provided daily at 19:30 during a 75-day trial period. At the conclusion of the trial, all ewes were slaughtered and Longissimus dorsi muscle samples were collected to determine nutrient composition and histological characteristics. Results demonstrated that high-level concentrate supplementation increased total dry matter intake, with G3 exhibiting significantly higher average daily gain compared to G1 and G2 ($P<0.05$). Concentrate supplementation did not significantly affect pH at 1 hour (pH1 h) or 24 hours (pH24 h) post-mortem, drip loss, moisture content, or crude ash content ($P>0.05$). However, G3 showed significantly higher cooked meat percentage, crude protein content, intramuscular fat content, and muscle fiber density compared to G1 ($P<0.05$). G2 and G3 groups displayed significantly elevated total amino acid, essential amino acid, and aspartic acid contents relative to G1 ($P<0.05$), while phenylalanine and lysine contents were significantly higher in G3 than G1 ($P<0.05$). Threonine and glutamic acid contents in G3 tended to be higher than G1 ($0.05\leq P<0.10$). Concentrate supplementation did not significantly affect saturated fatty acid, monounsaturated fatty acid, or polyunsaturated fatty acid contents ($P>0.05$), though C18:1 trans-9 content in G3 tended to be lower than G1 ($0.05\leq P<0.10$). These findings indicate that concentrate supplementation significantly enhances nutritional value and improves meat tenderness in Pengbo semi-fine wool sheep, with 400 g/d supplementation proving superior to 200 g/d.

Keywords: Tibet Pengbo semi-fine wool sheep; grazing; supplementation; meat quality; amino acids; fatty acid composition

Introduction

Since the founding of the People' s Republic of China, sheep farming in Tibet has achieved substantial development. In 2014, total sheep and goat inventory in Tibet reached 11.9 million head, including 7.49 million sheep [1], ranking seventh nationally [2]. In Tibet' s unique natural environment, grazing sheep produce meat with excellent flavor and rich broth. However, traditional pastoral sheep farming in Tibet relies exclusively on grazing, resulting in long feeding periods and low slaughter rates. In 2014, the combined slaughter rate for sheep and goats was only 32.11% [3].

To increase market supply, farmers in better-developed agricultural regions such as Shannan City, Lhasa City, and surrounding counties have implemented concentrate supplementation (barley, wheat, or commercial concentrates) during grazing, achieving significant weight gain improvements. Grazing with concentrate supplementation improves nutritional balance [4,5], enhancing not only production performance but also meat quality. Early research demonstrated that lambs supplemented with high-energy feeds such as corn and wheat (except barley) produced better flavor than purely grazed animals [6], while concentrate-fed lambs primarily receiving corn showed lower polyunsaturated fatty acid (PUFA) content than grazed counterparts [7,8]. Supplementing grazing yaks in Qinghai' s Haibei Prefecture with mixed feeds containing corn, soybean meal, barley, alfalfa meal, and hay significantly improved meat tenderness [9]. Despite the unique flora and fauna resources of Tibet' s alpine grasslands, scientific research remains relatively limited, with few reports on the effects of supplementation on feed intake and meat quality of Tibetan sheep. This study selected the renowned Pengbo semi-fine wool sheep as experimental subjects, establishing three concentrate supplementation levels to analyze differences in ewe feed intake and nutrient deposition in meat, thereby elucidating the effects of concentrate supplementation on meat quality of grazing Tibetan sheep and providing scientific basis and technical approaches for improving sheep productivity in Tibet.

1.1 Experimental Time and Location

The experiment was conducted from August to November 2015 in Bailing Village, Kazi Township, Linzhou County, Tibet Autonomous Region. The grazing pasture was dominated by *Kobresia pygmaea* and *Carex moorcroftii* vegetation.

1.2 Experimental Animals and Grouping

Thirty Pengbo semi-fine wool ewes aged 3-4 years (average body weight 26.98 kg) were randomly divided into three groups (n=10) according to homogeneous principle: G1 (control, grazing without supplementation), G2 (grazing plus 200 g/d concentrate), and G3 (grazing plus 400 g/d concentrate). The concentrate was a complete formula feed purchased from Tibet Jiufeng Feed Co., Ltd. Nutritional levels of concentrate and pasture are presented in . The experimental period lasted 87 days, including a 12-day preliminary period and a 75-day formal trial period.

The concentrate consisted of 65% corn, 10% soybean meal, 8% cottonseed meal, 6% rapeseed meal, 10% wheat bran, and 1% premix. Dry matter (DM), crude protein (CP), and neutral detergent fiber (NDF) contents were determined according to Zhang et al. [10], while metabolizable energy (ME) was calculated using the method of Freer [11].

1.3 Feeding Management

All experimental ewes were released for grazing at 09:00 daily on approximately 500 mu of natural grassland, returned to rest at 13:00, released again at 15:00, and returned at 19:00. After returning, ewes were placed in separate pens for supplementation (except G1) at 19:30 daily. All animals had free access to water, and body weight was measured every 15 days before grazing after overnight fasting. At the conclusion of the feeding trial, six ewes with similar body weight were selected from each group for slaughter. Feed was withheld for 24 hours and water for 2 hours prior to slaughter.

1.4 Sample Collection and Preparation

Pasture samples were collected in mid-September. Fifty 1 m $\times$ 1 m quadrats were established within the grazing area, and all edible plant above-ground parts were clipped, oven-dried to constant weight, ground, and passed through a 40-mesh sieve for determination of saturated n-alkane content. During the same period, three randomly selected ewes from each group were dosed with one C32 capsule before grazing and after returning daily (preparation method described in Zhang [12]) as an external marker. Fecal samples were collected on day 7 of dosing for five consecutive days. Thirty to fifty percent of daily fecal collections from each ewe were composited as analytical samples, which were then pooled for each animal after collection and processed identically to forage samples for analysis.

Meat samples were collected 45 minutes post-slaughter. The right side Longissimus dorsi muscle was excised, sealed in plastic bags, and stored at -20°C. After transport to the laboratory, 50-60 g fresh samples were sliced thinly, placed in petri dishes, and freeze-dried for 96 hours using a CHRIST Alpha (2-4 LSC, Germany) freeze dryer operating at -87°C and 0.006 MPa. Freeze-dried samples were ground into powder using a coffee grinder, mixed thoroughly, sealed in self-sealing bags wrapped with aluminum foil, and stored for nutrient analysis. Additionally, 3-5 g of right side Longissimus dorsi muscle was collected at slaughter, fixed in 10% formalin solution for 48 hours, secondarily fixed, and processed for paraffin sectioning and hematoxylin-eosin (HE) staining according to conventional histological methods.

1.5 Measurement Indicators and Methods

Dry matter (DM), crude protein (CP), and neutral detergent fiber (NDF) contents in forage and concentrate were determined according to methods provided by Zhang et al. [10]. Herbage intake was measured using the saturated n-alkane method [13], with n-alkane concentrations in forage and concentrate determined using the same methodology. Meat pH was measured using a pH meter (PHS-3C). Drip loss and cooked meat percentage were determined using methods described by Zhang [12]. Meat CP content was measured using a FOSS Kjeltec-2300 nitrogen analyzer (Sweden). Intramuscular fat (IMF) content was determined using an ANKOM XT-15 fat analyzer (USA). Amino acid content was measured using hydrochloric acid hydrolysis with an automatic amino acid analyzer (L-8900, Hitachi, Japan). For fatty acid analysis, samples were methylated according to GB/T 17377-2008, and the supernatant was analyzed using gas chromatography (450-GC, Shimadzu, Japan) with the following conditions: capillary column 60 m × 250 m × 0.25 m, injection temperature 260°C, detector temperature 270°C, split ratio 20:1, and injection volume 1.0 L. Muscle tissue sections were observed under an optical microscope (400×), with 6-10 fields photographed per sample. Image analysis software was used to determine myocyte number, diameter, and density per field, with at least 60 myocytes measured per section.

1.6 Data Statistics and Calculation

Herbage intake was calculated using the formula of Mayes et al. [13] with C31/C32 as the alkane pair. Experimental data were analyzed using one-way ANOVA in SAS 8.2 software. Differences were considered tendencies when $0.05 \leq P < 0.10$ and significant when $P < 0.05$. Duncan' s multiple range test was used for post-hoc comparisons when significant differences were detected.

Results

2.1 Feed Intake of Ewes

presents DM, CP, ME, and NDF intake from concentrate and pasture for each group. Overall, G3 exhibited higher total dry matter intake (DMI), total crude protein intake (CPI), and total metabolizable energy intake (MEI) than G2 and G1, while total neutral detergent fiber intake (NDFI) was lower than the latter two groups.

2.2 Effects of Concentrate Supplementation on Average Daily Gain (ADG) and Carcass Weight

As shown in , despite no significant differences in initial body weight ($P > 0.05$), concentrate supplementation significantly affected ADG ($P < 0.05$). G3 was significantly higher than G2 and G1 ($P < 0.05$), while G2 did not differ significantly from G1 (P>0.05). Pre-slaughter live weight and carcass weight did not differ significantly among groups (P>0.05).

2.3 Effects of Concentrate Supplementation on Meat Physicochemical Properties

shows that concentrate supplementation did not significantly affect pH1 h, pH24 h, meat color, or drip loss (P>0.05). However, cooked meat percentage in G2 and G3 was significantly higher than in G1 (P<0.05).

2.4 Effects of Concentrate Supplementation on Conventional Nutrient Content and Muscle Fiber Histological Characteristics

As presented in , concentrate supplementation did not significantly affect moisture or crude ash content (P>0.05), but CP and IMF contents in G2 and G3 were significantly higher than in G1 (P<0.05), with no significant difference between G2 and G3 (P>0.05).

demonstrates that G3 tended to have lower muscle fiber diameter and area than G2 and G1 (0.05≤P<0.10). Conversely, muscle fiber density in G3 was significantly greater than in G2 and G1 (P<0.05), while G2 did not differ significantly from G1 (P>0.05).

2.5 Effects of Concentrate Supplementation on Amino Acid Content

reveals that aspartic acid content in G2 and G3 was higher than in G1 (P<0.05). Threonine and glutamic acid contents in G3 tended to be higher than in G1 (0.05≤P<0.10). Serine, glycine, alanine, cysteine, valine, methionine, isoleucine, leucine, tryptophan, histidine, arginine, and proline contents did not differ significantly among groups (P>0.05). Phenylalanine and lysine contents in G3 were higher than in G1 (P<0.05), while G2 did not differ significantly from G1 (P>0.05). Total amino acid and essential amino acid contents in G2 and G3 were significantly higher than in G1 (P<0.05).

2.6 Effects of Concentrate Supplementation on Fatty Acid Content

As shown in , concentrate supplementation did not significantly affect most fatty acid contents except C17:1 (P>0.05). C17:1 content in G2 was significantly lower than in G1 and G3 (P<0.05), with no significant difference between the latter two groups (P>0.05). C18:1 trans-9 content in G3 tended to be higher than in G1 (0.05≤P<0.10). Saturated fatty acid (SFA), monounsaturated fatty acid (MUFA), and polyunsaturated fatty acid (PUFA) contents did not differ significantly among groups (P>0.05).

Discussion

3.1 Feed Intake of Ewes

Concentrate supplementation improves production performance by increasing nutrient intake of grazing livestock. Gekara et al. [14] reported that supplementing lactating beef cows with concentrate while grazing 12 hours daily increased DMI from 8.1 to 8.6 kg/d. Zhang et al. [15] found that concentrate supplementation maintained normal DMI in time-restricted grazing lambs while improving ADG. Supplementing corn significantly increased DMI, CPI, and MEI in Pelibuey lambs, thereby accelerating weight gain [16]. Protein supplementation improved growth rate and wool production in young Merino lambs, with effects persisting after supplementation ceased [17]. In this study, although high-level concentrate supplementation produced limited increases in DMI, it markedly elevated CPI and MEI, significantly improving ADG and demonstrating the production and quality benefits of 400 g/d concentrate supplementation.

3.2 Effects of Concentrate Supplementation on Meat Physicochemical Properties

pH is closely related to meat quality traits including tenderness, water-holding capacity, and color. Fresh meat pH 45 minutes post-slaughter typically ranges from 5.9-6.5, subsequently decreasing to 5.4-5.7 as muscle glycogen undergoes glycolysis. All groups in this study exhibited pH24 h values within this normal range. Meat color reflects muscle physiological, biochemical, and microbiological changes, with color intensity primarily determined by myoglobin content. Grazing livestock typically produce darker meat, and concentrate supplementation can alter meat color [18,19]. The darker meat color observed across all groups may indicate higher myoglobin content in plateau sheep meat (myoglobin is a cytoplasmic heme protein that stores and transports oxygen in muscle tissue). Cooked meat percentage is an important indicator of water-holding capacity, with grazing sheep showing higher drip loss than housed sheep [20]. Concentrate supplementation improved water-holding capacity and increased cooked meat percentage by enhancing nutritional status and promoting IMF deposition. IMF loosens muscle microstructure, increasing water adsorption capacity. Simultaneously, muscle moisture is relatively reduced as water is displaced by fat, and fat-containing muscle becomes more compact after cooking and cooling, reducing losses.

3.3 Effects of Concentrate Supplementation on Conventional Nutrient Content and Muscle Fiber Histological Characteristics

Moisture content plays a crucial role in maintaining meat quality, with lean meat typically containing 72-75% water [21]. Moisture content is inversely related to fat content. In this study, moisture content across groups ranged from 70.35-71.17%, lower than typical values, likely due to higher IMF content. IMF is another critical factor affecting meat quality, closely related to water-holding capacity, tenderness, and flavor. Appropriate IMF enhancement improves flavor, juiciness, and reduces toughness [22]. Research has established IMF as an important precursor for meat flavor development [23], with optimal palatability occurring at 3.5-4.5% IMF content [24]. IMF content in this study (5.03-8.10%) exceeded this range, suggesting that Pengbo semi-fine wool sheep meat may have suboptimal texture, which is expected given that semi-fine wool sheep are primarily selected for wool production with secondary meat purposes. High-energy, high-nutrient diets can increase IMF content in sheep meat [25]. In this study, high-level supplementation increased MEI and CPI by 1.34 and 1.60 MJ/d and 20 and 21 g/d compared to low-level supplementation and non-supplemented grazing, respectively, resulting in significantly increased IMF content. Additionally, increased dietary CP content enhances muscle protein synthesis efficiency and promotes body protein deposition [26], explaining the significantly increased CP content in high-level supplemented meat. Yu [18] reported that concentrate supplementation significantly increased IMF and CP contents in Longissimus dorsi muscle of grazing Xinjiang Brown cattle, improving meat color and tenderness. Meat tenderness is closely related to muscle fiber diameter and density [27,28], with smaller diameter and higher density associated with more tender, higher quality meat. Tibetan sheep under extensive management with long grazing periods and high activity levels typically exhibit coarser muscle fibers and limited IMF distribution [29]. Nutritional level effects on meat quality are manifested at the cellular level through differences in muscle fiber size [30]. High-level concentrate supplementation in this study provided higher nutritional status, significantly increased IMF content, and consequently increased muscle fiber density, producing finer muscle fibers and improved tenderness. Similarly, Kong et al. [9] reported that concentrate supplementation reduced shear force and improved tenderness in grazing yaks. Crude ash content reflects overall mineral element composition, and Zhao et al. [31] demonstrated that concentrate supplementation did not affect mineral content in sheep meat. Collectively, both low- and high-level concentrate supplementation promoted carcass protein deposition without compromising mineral nutrition, while high-level supplementation additionally improved meat tenderness and texture.

3.4 Effects of Concentrate Supplementation on Amino Acid and Fatty Acid Contents

Raw meat contains approximately 20% protein, serving as an important protein source for humans and providing various essential amino acids including lysine, isoleucine, and methionine. Of the 22 amino acids required by humans, sheep meat contains 17. In this study, essential amino acids accounted for 56.22%, 56.39%, and 56.17% of total amino acids across the three groups, relatively higher than values reported for Qinghai semi-fine wool sheep (50.81%) and plateau Tibetan sheep (51.04%) [32], indicating high nutritional value of Pengbo semi-fine wool sheep meat. Threonine and lysine are essential amino acids that play important roles in promoting human growth and development. High-level concentrate supplementation in this study significantly increased essential and total amino acid contents, particularly threonine and lysine, thereby improving nutritional and health functions. Glutamic acid, aspartic acid, glycine, alanine, phenylalanine, and tyrosine are flavor-enhancing amino acids that impart special umami taste to meat, with meat palatability depending on their content. High-level supplementation significantly increased or tended to increase aspartic acid, phenylalanine, and glutamic acid contents, suggesting enhanced umami flavor and improved consumer appeal. Furthermore, flavor-enhancing amino acids possess diabetes prevention and antihypertensive properties, conferring higher nutritional and health value to grazing plus supplementation systems.

In addition to amino acids, fatty acid composition and content are important factors affecting meat quality. Dietary energy level influences tissue fatty acid profiles. Reduced dietary energy level significantly increased linolenic acid content in lamb body fat [33], while simultaneous reduction of dietary energy and protein levels significantly increased oleic acid, linolenic acid, MUFA, and PUFA contents in backfat and leaf fat of Bamei pigs while decreasing SFA content [34]. In this study, despite higher MEI and CPI in high-level supplemented sheep, SFA, MUFA, and PUFA contents did not differ significantly from other groups, while trans-oleic acid (C18:1 trans-9) content was significantly reduced. Oleic acid is the most important MUFA in sheep meat, typically considered beneficial due to its cholesterol- and LDL-lowering effects. However, as a trans fatty acid, C18:1 trans-9 can elevate blood cholesterol and increase cardiovascular disease risk [35]. High-level concentrate supplementation significantly reduced C18:1 trans-9 content, improving fatty acid nutrition. These results demonstrate that 400 g/d concentrate supplementation not only preserves unsaturated fatty acids but also reduces harmful fatty acid components, thereby enhancing health benefits.

Conclusion

Concentrate supplementation significantly improves nutritional value and tenderness of Pengbo semi-fine wool sheep meat, with supplementation at 400 g/d per head superior to 200 g/d.

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