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Mineral supplements improve the nutritional value of forage and the livestock carrying capacity of high-Andean pastures

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ABSTRACT

This study evaluated the effects of calcium-based soil amendments and pasture composition on forage productivity, nutritional quality, and livestock carrying capacity in high-Andean grassland systems. A field experiment was analyzed using linear mixed-effects models to assess the effects of soil amendments (limestone, dolomite, and gypsum), pasture types (*Lolium multiflorum*, *Trifolium pratense*, and their mixture), and their interaction on productivity, nutritional composition, derived nutritional indicators, and feed-related performance. Soil amendments significantly increased forage productivity and feed availability, improving dry matter production, crude protein yield, energy yield, and livestock carrying capacity. Limestone and gypsum showed the most consistent positive effects across productivity-related variables. Pasture composition mainly influenced forage nutritional quality. *Trifolium pratense* increased crude protein concentration, protein-derived energy, and protein: fiber ratio, while *Lolium multiflorum* showed higher fiber and carbohydrate concentrations. Mixed pastures exhibited intermediate values, reflecting functional complementarity between species. Although total energy concentration remained unaffected by treatments, calcium-based amendments increased the proportion of protein-derived energy, improving the nutritional efficiency of the forage. Multivariate analysis confirmed clear differentiation between amended and non-amended systems, with legumes associated with improved nutritional quality and productivity-related indicators. In conclusion, calcium-based soil amendments enhance forage production and feed availability, while legume inclusion improves forage protein value. The combined use of soil fertility management and pasture diversification represents an effective strategy to improve feed resources and livestock production efficiency in high-Andean systems.

Keywords: crude protein, dry matter availability, grazing management, pasture productivity, soil amendments, total energy.

INTRODUCTION

High-Andean pastoral systems constitute the primary feed resource for livestock production in mountain ecosystems, sustaining milk and meat production under extensive grazing conditions (Pérez-Suárez et al., 2018). However, forage productivity in these systems is frequently constrained by soil degradation, low nutrient availability, and the high acidity characteristic of Andean soils (Arias-Arredondo et al., 2026). Soil acidity reduces the availability of essential nutrients such as calcium, magnesium, and phosphorus, while increasing the solubility of potentially toxic elements, thereby negatively affecting forage establishment, growth, and persistence (Rahman and Upadhyaya, 2021; Huang et al., 2023).

Low forage productivity in high-Andean grasslands is commonly associated with poor nutritional quality, particularly due to limited protein availability for grazing animals (Arias-Arredondo et al., 2026). These nutritional constraints reduce animal performance and livestock carrying capacity, especially during periods of reduced pasture growth (Buxton et al., 1995). Therefore, improving both forage biomass production and nutritive value represents a major challenge for the sustainability and efficiency of mountain livestock systems (Cooke et al., 2025).

Mineral amendments such as limestone, dolomite, and agricultural gypsum have been widely used to improve soil chemical properties and mitigate the negative effects of soil acidity in agricultural and pastoral systems. Previous studies have shown that these amendments can increase nutrient availability, stimulate plant growth, and enhance forage biomass production in acidic soils (Minato et al., 2023; Makaza et al., 2026). In addition, the incorporation of legumes such as *Trifolium pratense* has been associated with higher crude protein concentrations and improved forage nutritional quality compared with pure grass stands such as *Lolium multiflorum* (Vallejos-Cacho et al., 2024).

Nevertheless, most available studies have focused primarily on forage yield or isolated nutritional components, whereas studies simultaneously integrating forage productivity, nutritional functionality, energy contribution, protein yield per unit area, and livestock carrying capacity remain scarce, particularly in high-Andean pastoral systems. This limitation restricts the understanding of the functional effects of mineral amendments on the sustainability and feeding value of mountain grazing systems (Vallejos-Cacho et al., 2024; Arias-Arredondo et al., 2026).

In this context, integrated evaluations linking forage productivity with functional nutritional indicators and livestock-support variables are needed. Such an approach may contribute to the development of ecological intensification strategies aimed at increasing forage production and nutritional value without compromising the sustainability of high-Andean ecosystems (Duchicela et al., 2024). The novelty of the present study lies in the simultaneous evaluation of forage productivity, proximal composition, derived nutritional indicators, protein yield, energy yield, and livestock carrying capacity in high-Andean pastures subjected to different mineral amendments. In addition, grass monocultures, legume monocultures, and mixed forage associations were compared under acidic mountain soil conditions.

The hypothesized that mineral amendments simultaneously improve forage productivity, nutritional quality, and carrying capacity in high-Andean pastoral systems, and that these responses vary according to pasture type. Therefore, the aim of this study was to evaluate the effects of limestone, dolomite, and agricultural gypsum on biomass production, nutritional composition, energy-related indicators, protein yield, and livestock carrying capacity in pastoral systems dominated by *Lolium multiflorum*, *Trifolium pratense*, and their associations.

MATERIAL AND METHODS

Study area and experimental site

The study was conducted under field conditions at the Pampa Grande Annex, Cajabamba Province, Cajamarca Region, Peru, on a research field of the National Institute of Agrarian Innovation (INIA). The experiment was established on 31 May 2024, and forage evaluations were performed on 30 October 2024. The site is located at UTM coordinates 823363.13 E and 9157608.45 N (WGS 84, zone 17S), at 2,636 m a.s.l. The area presents a temperate montane climate, with annual temperatures ranging from 8.7 to 21.3 °C and annual precipitation between 900 and 1,381 mm, concentrated mainly from November to April. The soils are derived from sedimentary materials of the Farrat and Chulec formations. Initial soil conditions showed slightly acidic pH (5.9–6.3), low electrical conductivity (2.8–4.5 mS m^{-1}), and moderate organic matter content (20–32 g kg^{-1}), with no detectable exchangeable aluminium toxicity. Available phosphorus and potassium ranged from 40–78 and 63–177 mg kg^{-1}, respectively. The soil was predominantly classified as clay loam. Surrounding land use is predominantly livestock-based, with forage production systems oriented to milk and meat production.

Soil preparation was carried out using a disc plough to a depth of approximately 30 cm, followed by cross-harrowing to ensure adequate soil homogenisation according to standard agronomic practices for high-Andean forage systems. The experimental area covered 648 m² and comprised 36 plots measuring 2 × 2 m (4 m² each), arranged in three blocks under a randomized complete block design with a 4 × 3 factorial arrangement.

Experimental design and soil amendments

The experiment was established under a randomized complete block design (RCBD) with a 4 × 3 factorial arrangement. The first factor consisted of four soil amendment treatments: control (without amendment), limestone, dolomite, and agricultural gypsum. The second factor corresponded to three pasture types: *Lolium multiflorum* monoculture, *Trifolium pratense* monoculture, and a mixed pasture of *L. multiflorum* + *T. pratense*. The experimental design included three blocks as random factors, resulting in a total of 36 experimental units (4 amendments × 3 pasture types × 3 blocks).

Soil amendments were applied before pasture establishment to improve soil chemical conditions associated with acidity constraints in high-Andean soils. A uniform dose of 1 t·ha^{-1} was incorporated into the soil during land preparation before sowing. This moderate application rate was selected based on agrotechnical recommendations for high-altitude grassland maintenance, balancing soil chemical correction with socio-economic and logistical feasibility for extensive livestock production systems (Puenayan Irua and Benavides Rosales, 2025). Furthermore, global meta-analytical and field evidence supports the use of lower, strategic liming rates over heavy corrective doses in upland ecosystems, as a dose of 1 t·ha^{-1} avoids abrupt pH or osmotic shifts in the rhizosphere, thereby preventing chemical and microbial shock to native soil communities while optimizing economic profitability for local producers (Enesi et al., 2023; Morton, 2020). In line with this, contemporary field trials have demonstrated that excessive liming application can trigger a decline in available plant nutrients like phosphorus and calcium, emphasizing the ecological importance of moderate, tailored application rates to sustain soil nutrient dynamics (Leumassi Mbotchak et al., 2024). Sowing densities were 35.0 kg·ha^{-1} for *L. multiflorum* monoculture, 30.0 kg·ha^{-1} for *T. pratense* monoculture, and 24.5 and 10.5 kg·ha^{-1} for *L. multiflorum* and *T. pratense*, respectively, in the mixed pasture. Pasture species were subsequently established according to the assigned treatment combinations.

Biomass production

Forage biomass was evaluated 152 days after pasture establishment. Aboveground biomass within each experimental plot was harvested at ground level from a 4 m² sampling area, collected, and weighed immediately in the field to determine fresh matter yield (FM), following the methodology described by Arias et al. (2021).

Subsamples were pre-dried and subsequently oven-dried at 80 °C for 48 h until constant weight to determine dry matter (DM). Fresh matter (FM) and dry matter (DM) yields were expressed as t ha^{-1}. Biomass values obtained from the 4 m² sampling area were extrapolated to a hectare basis.

The percentage of dry matter (%DM) was calculated as:

$$\%DM = \left(\frac{Dry\ weight}{Fresh\ weight}\right) \times 100 \quad (1)$$

where: *dry weight* corresponds to oven-dried biomass and fresh weight to the initial *fresh biomass* collected in the field.

Dry matter yield (DM yield) was estimated from the relationship between fresh and dry biomass using the following equation:

$$DM\ yield = FM\ yield \left(\frac{\%DM}{100}\right) \quad (2)$$

where: *FM* yield corresponds to fresh matter production expressed as t ha^{-1}.

Proximal composition analysis

After oven drying, plant samples were ground using a Willey mill and passed through a 1-mm mesh sieve to obtain homogeneous material. Samples were analyzed at the Food Analysis Laboratory (LAA), National Institute of Nutrition, Peru.

Nutritional parameters were determined according to official AOAC methods (Latimer, 2023): Ash content was determined using AOAC method 930.05, crude fat using AOAC method 930.09, and crude protein using AOAC method 2001.11 (Kjeldahl method; N × 6.25). Crude fiber was determined according to AOCS method Ba 6-84 (AOCS, 2017). Carbohydrate content and total energy (kcal 100 g^{-1} dry weight) were estimated according to the procedures described by Collazos (1993).

Derived nutritional indicators

Derived nutritional indicators were calculated to evaluate the functional nutritional quality of the forage under different soil amendments and pasture types. These indicators included the protein: fiber ratio, total energy concentration, and the proportion of energy derived from protein.

The protein: fiber ratio was estimated as an integrative indicator of forage quality, reflecting the balance between nutritive components (crude protein) and structural components (crude fiber) associated with reduced digestibility (Van Soest, 1994), according to the following equation:

$$Protein{:}\,fiber\ ratio = \frac{Crude\ protein\ (\%)}{Crude\ fiber\ (\%)} \tag{3}$$

where: *crude protein* and *crude fiber* were expressed on a dry matter basis.

Total energy concentration (kcal kg^{-1} dry matter) was estimated from the proximal composition of the forage following the procedures described by Collazos (1993). Energy values were calculated considering the caloric contribution of protein, fat, and carbohydrates.

The percentage of energy derived from protein was calculated to estimate the relative contribution of protein to the total forage energy content (FAO, 2003), using the following equation:

$$Protein - derived\ energy\ (\%) = \frac{Protein\ energy}{Total\ energy} \times 100 \tag{4}$$

where: *protein energy* was estimated assuming an energetic value of 4 kcal g^{-1} protein.

These indicators were used to characterize the nutritional functionality of the forage and to assess the effects of soil amendments and pasture composition on forage quality in high-Andean pastoral systems.

Integrated productivity and nutritional variables

Integrated variables combining forage productivity and nutritional quality were calculated to evaluate the functional performance of the pasture systems under different soil amendments and pasture types. These variables included dry matter availability, crude protein yield, energy yield, and carrying capacity.

Dry matter availability (DM availability; t ha^{-1}) was estimated from total dry matter production adjusted by a forage utilization efficiency factor of 0.60, corresponding to rotational grazing management (Holechek et al., 2000). The calculation was performed as follows:

$$DM\ availability = Dry\ matter\ yield \times 0.60 \tag{5}$$

where: *dry matter yield* was expressed as t ha^{-1}.

Crude protein yield (t ha^{-1}) was calculated as the product of dry matter yield and crude protein concentration:

$$Crude\ protein\ yield = \frac{Dry\ matter\ yield \times Crude\ protein\ (\%)}{100} \tag{6}$$

Energy yield (kcal ha^{-1}) was estimated by multiplying dry matter yield by the total energy concentration of the forage calculated from proximal composition:

$$Energy\ yield = Dry\ matter\ yield \times Total\ energy \tag{7}$$

where: *total energy* was expressed as kcal kg^{-1} *dry matter* and estimated according to the caloric contribution of protein, fat, and carbohydrates following Collazos (1993) and FAO conversion factors (FAO, 2003).

Carrying capacity (AU ha^{-1} $year^{-1}$) was estimated based on dry matter availability and the annual forage requirement of a 450-kg cow equivalent to one animal unit (AU). An annual forage requirement of 4.93 t DM AU^{-1} $year^{-1}$ was considered (Holechek et al., 2000; NRC, 2016). Carrying capacity was calculated as:

$$Carrying\ capacity = \frac{DM\ availability}{4.93} \tag{8}$$

where: *DM* availability was expressed as t ha^{-1} and 4.93 corresponds to the annual dry matter requirement of one animal unit.

These integrated indicators were used to assess the combined effects of soil amendments and pasture composition on forage productivity, nutritional functionality, and livestock support capacity in high-Andean pastoral systems.

Statistical analysis

Statistical analyses were performed using R software (R Core Team, 2020). The experiment followed a randomized complete block design with a factorial arrangement of four soil amendments and three pasture types. The effects of soil amendment, pasture type, and their interaction on forage production, nutritional composition, derived nutritional indicators, and integrated productivity variables were evaluated using linear mixed-effects models. Soil amendment, pasture type, and their interaction were considered fixed effects, while block was included as a random effect. Analysis of variance (ANOVA) was performed on the fitted models, and when significant differences were detected ($p < 0.05$), Tukey's HSD test was applied for mean comparisons. Estimated marginal means ± standard error were used for graphical representation. A principal component analysis (PCA) was performed using standardized variables to evaluate multivariate relationships among forage productivity and nutritional traits. Model assumptions were verified through residual analysis, and statistical significance was established at $p < 0.05$.

RESULTS

Forage production

Analysis of variance from the linear mixed-effects models showed that soil amendment significantly affected fresh and dry biomass production ($p < 0.001$; Table 1). Pasture type significantly influenced fresh biomass production ($p < 0.001$), whereas no significant effect was observed for dry biomass production ($p = 0.447$). The interaction between soil amendment and pasture type was not significant for either variable ($p > 0.05$).

The application of soil amendments increased fresh and dry biomass production compared with the control treatment (Figure 1A–B). Limestone produced the highest fresh biomass values, followed by agricultural gypsum, whereas the control showed the lowest production. For dry biomass, limestone, dolomite, and agricultural gypsum resulted in significantly higher values than the control, with no significant differences among amendments.

Pasture type also affected fresh biomass production (Figure 1C). *Trifolium pratense* monoculture showed the highest fresh biomass production, followed by the *Lolium–Trifolium* mixture, whereas *Lolium* monoculture presented the lowest values.

Nutritional quality: proximal composition

Analysis of variance showed that forage proximal composition was differentially affected by soil amendment, pasture type, and their interaction (Table 2).

Crude protein concentration was significantly affected by both soil amendment ($p < 0.01$) and pasture type ($p < 0.001$), whereas their interaction was not significant. Agricultural gypsum, limestone, and the control treatment showed higher protein concentrations (24–26%) than dolomite, which presented the lowest value (≈19%) (Figure 2A). Among pasture types, *Trifolium pratense* showed the highest protein concentration (≈29%), followed by the *Lolium–Trifolium* mixture (≈24%), whereas *Lolium multiflorum* monoculture presented the lowest values (≈18%) (Figure 2B).

Fiber concentration was not affected by soil amendment, but differed significantly among pasture types ($p < 0.001$). *The Lolium–Trifolium* mixture and *Lolium* monoculture showed the

Table 1. Analysis of variance (ANOVA) for fresh and dry biomass production under different soil amendments and pasture types

Source of variation	DM (t ha⁻¹) (F)	FM (t ha⁻¹) (F)
Soil amendment	19.73 ***	31.86 ***
Pasture type	0.83 ns	28.06 ***
Soil amendment × Pasture type	0.54 ns	1.4 ns

Note: Results from the linear mixed-effects models with soil amendment, pasture type, and their interaction as fixed effects, and block as a random effect. DM = dry matter; FM = fresh matter. F = ANOVA F statistic; p-value = significance level. $p < 0.05$ (*), $p < 0.01$ (**), $p < 0.001$ (***); ns = non-significant

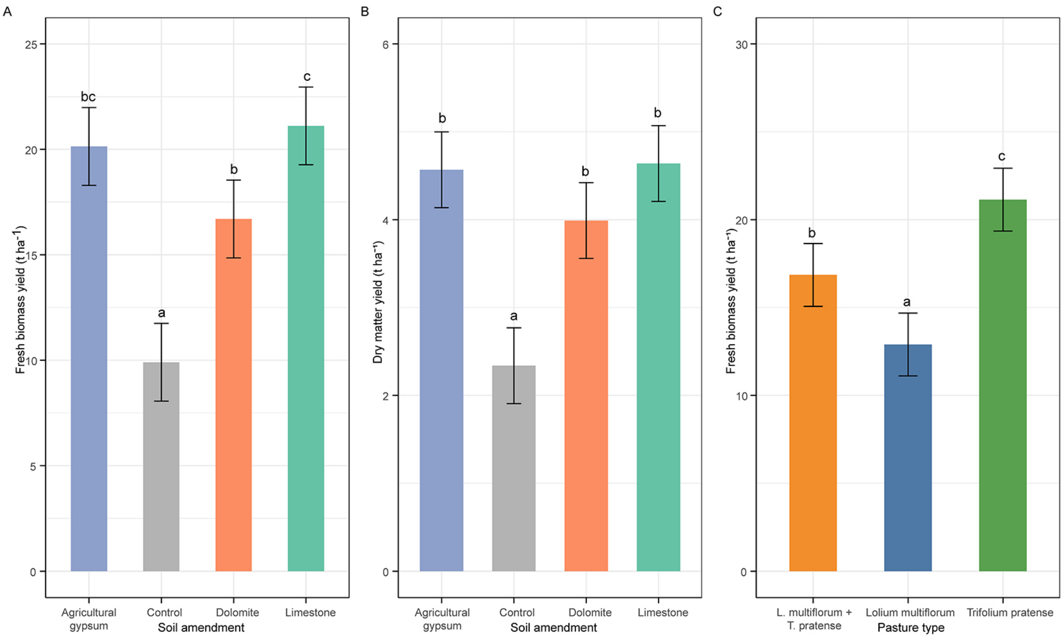

Figure 1. Effect of soil amendments on fresh biomass production (A) and dry biomass production (B), and effect of pasture type on fresh biomass production (C). Bars represent estimated marginal means ± standard error from the linear mixed-effects models. Different letters indicate significant differences among treatments according to Tukey's HSD test ($p < 0.05$)

highest fiber concentrations (≈25–26%), while *Trifolium* monoculture presented the lowest value (≈18%) (Figure 2C). No significant interaction between soil amendment and pasture type was detected for this variable.

Fat concentration was significantly influenced by soil amendment ($p = 0.006$), whereas pasture type alone had no significant effect. However, a significant interaction between soil amendment and pasture type was detected ($p = 0.045$), indicating that fat concentration varied according to treatment combination (Table 2). In general, agricultural gypsum showed the highest fat concentrations in the mixed pasture, whereas the control treatment presented the lowest values, particularly in *Lolium* monoculture (Figure 2D).

Ash concentration was significantly affected by both soil amendment and pasture type ($p < 0.001$), meanwhile their interaction was not significant (Table 2). Agricultural gypsum showed the highest ash concentration (≈10.5%), whereas the control treatment presented the lowest value (≈8.2%). Likewise, the *Lolium–Trifolium* mixture and *Trifolium* monoculture showed higher ash concentrations (≈9.5–10.5%) than *Lolium* monoculture (≈8.1%).

Carbohydrate concentration was not significantly affected by soil amendment, but differed among pasture types ($p = 0.002$), with no significant interaction between factors (Table 2). *Lolium* monoculture showed the highest carbohydrate concentration (≈39%), whereas the mixed pasture and *Trifolium* monoculture presented lower and similar values (≈29–32%).

Table 2. Analysis of variance (ANOVA) of proximal composition variables under different soil amendments and pasture types

Source of variation	Crude protein (%)	Fiber (%)	Fat (%)	Ash (%)	Carbohydrates (%)
Soil amendment	6.18 **	0.38 ns	5.38 **	7.8 ***	2.38 ns
Pasture type	27.06 ***	14.8 ***	0.34 ns	17.44 ***	8.77 **
Soil amendment × Pasture type	1.58 ns	1.38 ns	2.62 *	0.97 ns	0.78 ns

Note: Results from the linear mixed-effects models with soil amendment, pasture type, and their interaction as fixed effects, and block as a random effect. F = ANOVA F statistic; p-value = significance level. $p < 0.05$ (*), $p < 0.01$ (**), $p < 0.001$(***); ns = non-significant.

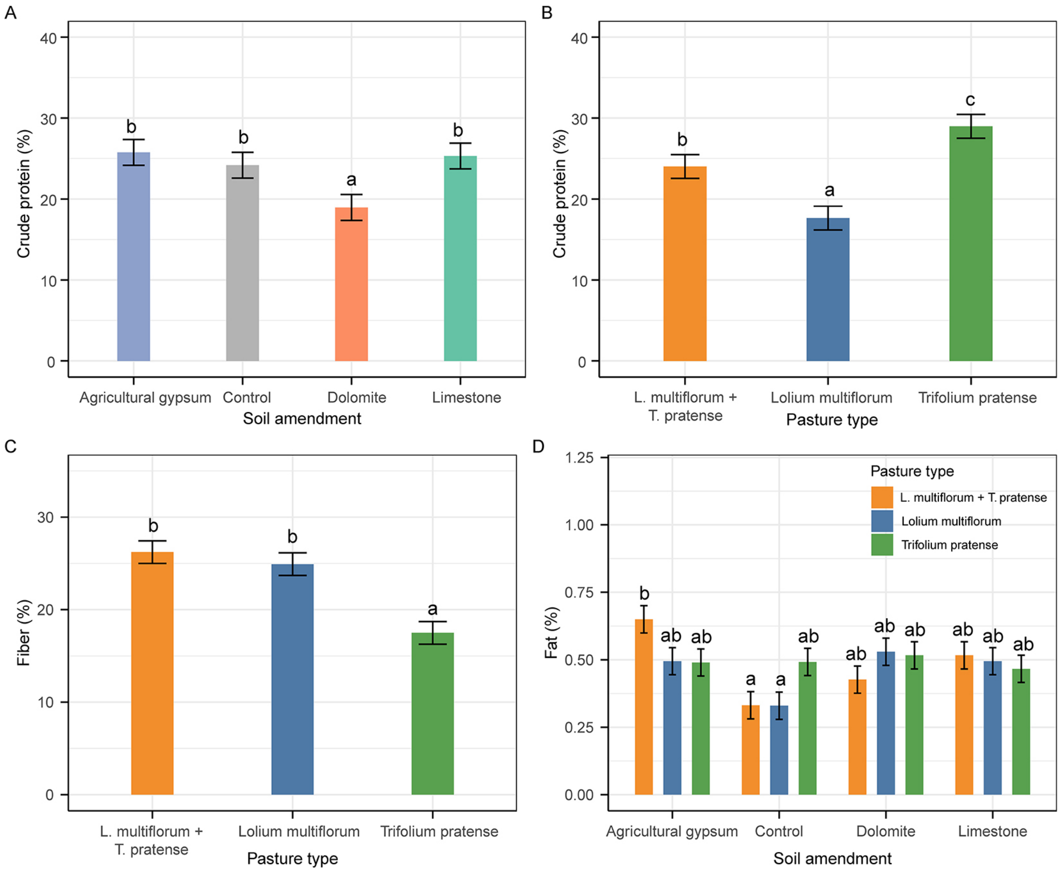

Figure 2. Effects of soil amendments and pasture type on forage nutritional composition. (A) Crude protein concentration according to soil amendment. (B) Crude protein concentration according to pasture type. (C) Crude fiber concentration according to pasture type. (D) Interactive effects of soil amendments and pasture type on crude fat concentration. Bars represent estimated marginal means ± standard error. Different letters indicate significant differences according to Tukey's test ($p < 0.05$)

Derived nutritional indicators

Analysis of variance showed that derived nutritional indicators responded differently to soil amendments and pasture type (Table 3). The protein ratio and the proportion of protein-derived energy were mainly influenced by pasture composition, whereas protein-derived energy was also significantly affected by soil amendments.

The protein: fiber ratio was not significantly affected by soil amendment, whereas pasture type showed a highly significant effect ($p < 0.001$). The interaction between factors was not significant. As shown in Figure 3A, *Trifolium pratense* presented the highest protein ratio (≈1.7), significantly exceeding *Lolium multiflorum* (≈0.7) and the mixed pasture *L. multiflorum* + *T. pratense* (≈1.0), which did not differ significantly from each other.

Table 3. Analysis of variance (ANOVA) for derived nutritional indicators of pastures under different soil amendments and pasture types

Source of variation	Protein:fiber ratio	Total energy (kcal kg^{-1})	Protein-derived energy (%)
Soil amendment	1.32 ns	2.01 ns	6.56**
Pasture type	31.32***	1.05 ns	31.19***
Soil amendment × Pasture type	1.48 ns	0.83 ns	1.57 ns

Note: Results from the linear mixed-effects models with soil amendment, pasture type, and their interaction as fixed effects, and block as a random effect. F = ANOVA F statistic; p-value = significance level. $p < 0.05$ (*), $p < 0.01$ (**), $p < 0.001$(***); ns = non-significant.

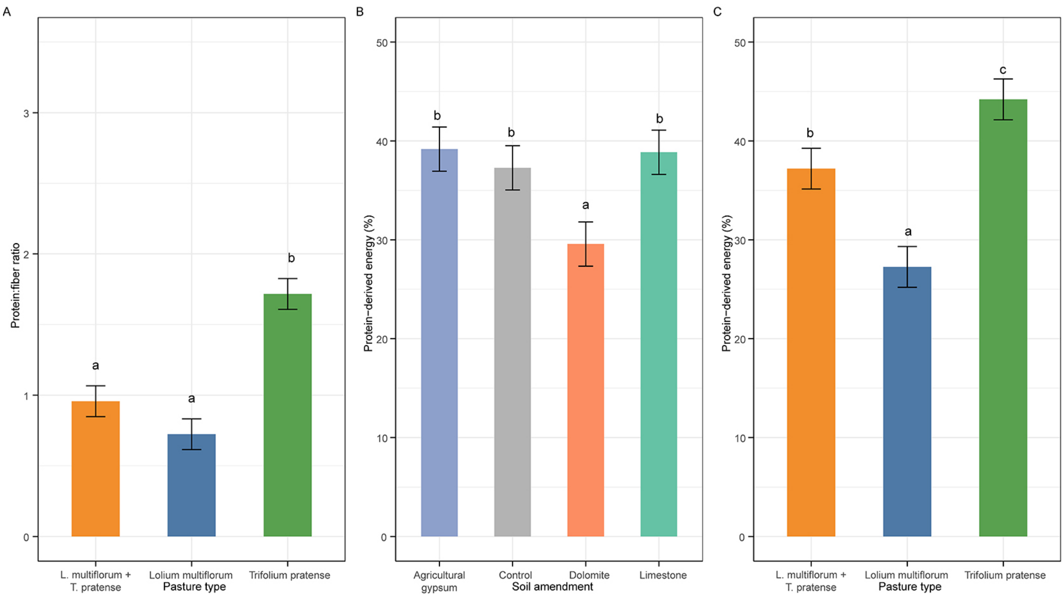

Figure 3. Effects of soil amendments and pasture types on the protein: fiber ratio and the proportion of protein-derived energy in high-Andean forages. (A) Protein: fiber ratio according to pasture type. (B) Percentage of protein-derived energy according to soil amendment. (C) Percentage of protein-derived energy according to pasture type. Bars represent means ± standard error. Different letters indicate significant differences among treatments according to Tukey's test (p < 0.05).

Total energy concentration (kcal kg^{-1} DM) was not significantly affected by soil amendment, pasture type, or their interaction (Table 3), indicating relatively stable forage energy content among treatments.

In contrast, the percentage of protein-derived energy was significantly influenced by both soil amendment ($p = 0.002$) and pasture type ($p < 0.001$), whereas the interaction effect was not significant ($p = 0.204$). According to Figure 3B, agricultural gypsum, limestone, and the control treatment showed the highest proportions of protein-derived energy (≈37–39%), whereas dolomite presented the lowest value (≈30%). Similarly, Figure 3C showed that *T. pratense* exhibited the highest proportion of protein-derived energy (≈44%), followed by the mixed pasture (≈37%), while *L. multiflorum* showed the lowest value (≈27%).

Integrated productivity and nutritional variables

Integrated productivity and nutritional variables were influenced mainly by soil amendment, whereas pasture type showed more specific effects on crude protein yield (Table 4). No significant amendment × pasture interactions were detected, indicating relatively consistent pasture responses across soil amendment treatments.

Dry matter availability (DMA) was significantly affected by soil amendment ($p < 0.001$), whereas pasture type ($p = 0.447$) and the interaction effect were not significant (Table 4). Gypsum and limestone showed the highest DMA values (≈2.7–2.8 t ha^{-1}), followed by dolomite (≈2.4 t ha^{-1}), while the control treatment presented the lowest forage availability (≈1.4 t ha^{-1}) (Figure

Table 4. Analysis of variance (ANOVA) for integrated productivity and nutritional variables under different soil amendments and pasture types

Source of variation	DMA	Crude protein yield	Energy yield	Carrying capacity
Soil amendment	19.73 ***	12.52 ***	20.48 ***	19.73 ***
Pasture type	0.83 ns	6.77**	0.6 ns	0.83 ns
Soil amendment × Pasture type	0.54 ns	0.86 ns	0.48 ns	0.54 ns

Note: DMA = dry matter availability. Results from the linear mixed-effects models with soil amendment, pasture type, and their interaction as fixed effects, and block as a random effect. F = ANOVA F statistic; p-value = significance level. p < 0.05 (*), p < 0.01 (**), p < 0.001(***); ns = non-significant.

4A). These results indicate that mineral amendments substantially improved forage availability under high-Andean grazing conditions.

Crude protein yield was significantly influenced by both soil amendment and pasture type, without significant interaction effects (Table 4). Gypsum and limestone promoted the highest protein yields (≈1.2 t ha^{-1}), whereas the control and dolomite treatments showed lower values (Figure 4B). Among pasture types, *Trifolium pratense* showed the highest crude protein yield (≈1.1 t ha^{-1}), followed by the mixed pasture, while *Lolium multiflorum* monoculture presented the lowest values (Figure 4C). This pattern confirms the greater protein contribution associated with legume-based pasture systems.

Energy yield was significantly affected by soil amendment ($p < 0.001$), whereas pasture type and the interaction effect were not significant (Table 4). These results indicate that improvements in forage energetic productivity depended primarily on soil amendment management rather than pasture composition.

Livestock carrying capacity showed a response pattern similar to DMA, with significant effects of soil amendment ($p < 0.001$), but not pasture type or the interaction effect (Table 4). Gypsum and limestone treatments achieved the highest carrying capacity values (≈0.55–0.57 AU ha^{-1}), followed by dolomite (≈0.49 AU ha^{-1}), while the control treatment showed the lowest value (≈0.28 AU ha^{-1}) (Figure 4D). Overall, mineral amendments improved not only forage productivity, but also the potential livestock support capacity of high-Andean pasture systems.

Post-harvest soil chemical properties

Post-harvest soil chemical properties varied among soil amendment treatments (Table 5). Limestone application produced the highest soil pH values and exchangeable calcium concentrations, particularly under *Lolium multiflorum* monoculture. In contrast, dolomite resulted in soil pH and exchangeable calcium values that remained close to those observed in the control treatment. Agricultural gypsum-maintained soil pH values similar to the control while exhibiting exchangeable calcium concentrations comparable to or slightly higher than those recorded in the unamended plots. Exchangeable magnesium, available phosphorus, and available potassium showed moderate variation among treatments. Overall, limestone was associated with the greatest changes in soil chemical properties, whereas gypsum and dolomite produced more limited modifications during the evaluation period.

Multivariate integration of forage nutritional quality and productivity

Principal component analysis (PCA) identified the main associations among forage nutritional composition, nutritional indicators, and productivity-related variables across soil amendments and pasture types (Figure 5). The first two principal components explained 57.5% of the total variability, with PC1 and PC2 accounting for 33.3% and 24.2%, respectively.

PC1 was positively associated with crude protein, ash content, energy-related variables, and dry matter yield, whereas carbohydrates showed a negative association along this axis. This pattern suggests that treatments located on the positive side of PC1 were characterized by greater relative protein contribution and improved nutritional composition, while treatments on the negative side were more associated with carbohydrate accumulation.

PC2 was positively related to crude fiber, fat content, and dry matter availability, reflecting a gradient associated with forage productivity and structural composition. The close orientation between protein and energy-related variables indicated a strong positive association, whereas the opposite distribution between protein and carbohydrates reflected an inverse relationship between protein concentration and carbohydrate accumulation.

Regarding treatment distribution, gypsum and limestone tended to cluster on the positive side of PC1, associated with greater protein concentration, ash content, and forage productivity. In contrast, the control treatment was located closer to the negative region of PC1 and PC2, reflecting lower nutritional quality and productive performance. Dolomite showed a more intermediate and dispersed distribution pattern.

Among pasture types, *Trifolium pratense* was mainly associated with greater protein concentration, protein-derived energy, and ash content, confirming its superior nutritional contribution. *Lolium multiflorum* was more closely associated with fiber and carbohydrates, reflecting a greater structural contribution and lower relative protein content. The mixed pasture showed an intermediate distribution, suggesting a balance between forage productivity and nutritional quality.

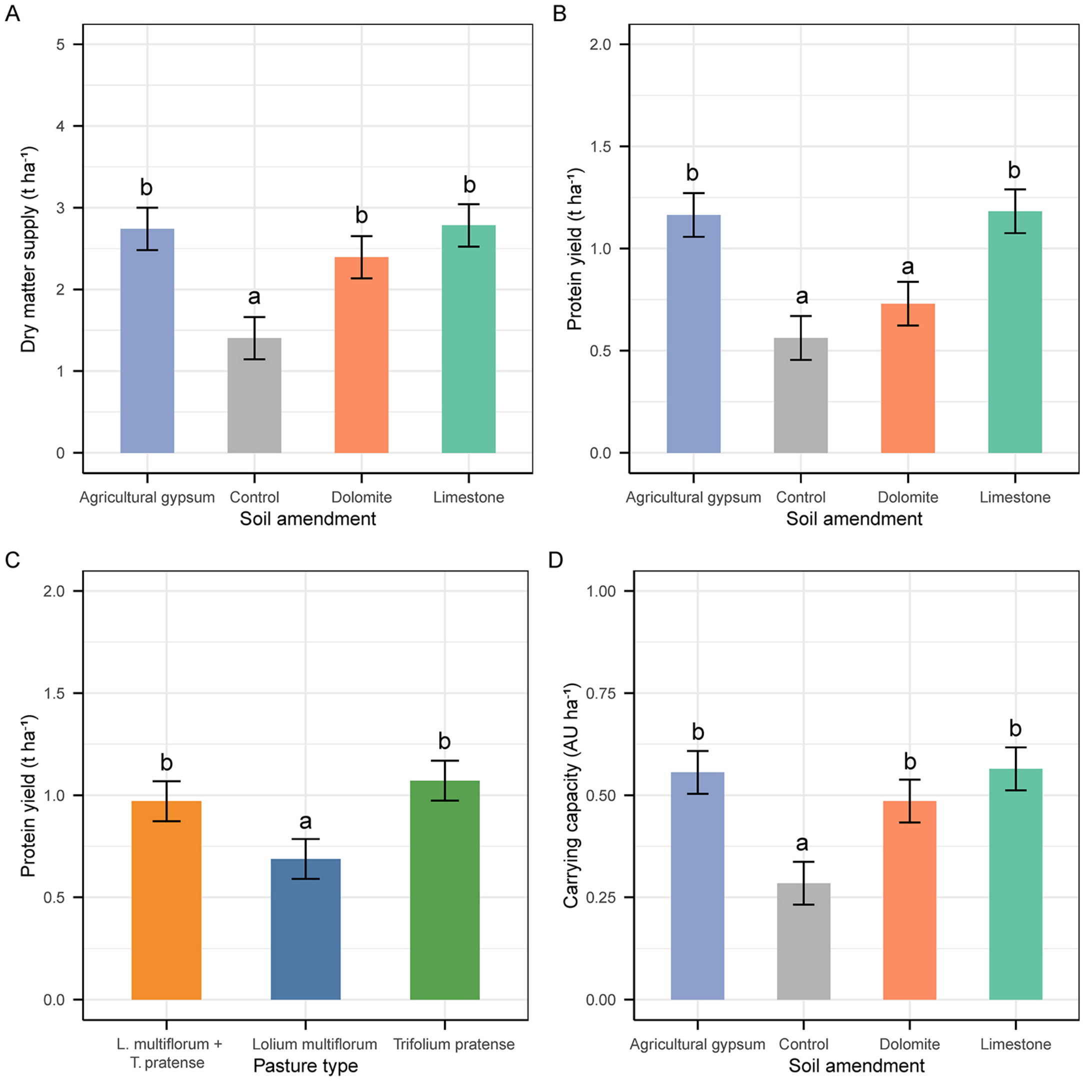

Figure 4. Effects of soil amendments and pasture types on dry matter availability, crude protein yield, nd livestock carrying capacity in high-Andean pasture systems. (A) Dry matter availability (DMA). (B) Crude protein yield according to soil amendment. (C) Crude protein yield according to pasture type. (D) Livestock carrying capacity. Bars represent means ± standard error. Different letters indicate significant differences among treatments according to Tukey's test ($p < 0.05$). AU = animal unit

Overall, the PCA showed that gypsum and limestone were associated with greater forage productivity and improved nutritional composition, whereas the inclusion of *Trifolium pratense* was associated with higher crude protein concentration, protein-derived energy, and ash content. These multivariate patterns indicate that both soil amendment and pasture composition contributed to the nutritional and productive performance of the pasture systems.

DISCUSSION

The present study demonstrated that calcium-based mineral amendments substantially improved forage productivity, crude protein yield, gross energy yield, and livestock carrying capacity in high-Andean pasture systems. These results indicate that improving soil chemical conditions in moderately acidic soils can directly enhance the quantity and nutritional value of available forage, with clear implications for animal feeding systems. The lack of significant amendment × pasture type interactions suggests that soil amendment effects were consistent across pasture compositions, reinforcing their broad applicability in mixed forage-based feeding systems.

The increases in fresh and dry biomass following limestone, dolomite, and gypsum application are consistent with previous studies reporting

Table 5. Post-harvest soil chemical properties under different soil amendments and pasture types

Soil amendment	Pasture type	pH	Exchangeable Ca (cmol(+) kg⁻¹)	Exchangeable Mg (cmol(+) kg⁻¹)	Available P (mg kg⁻¹)	Available K (mg kg⁻¹)
Control	*Lolium multiflorum*	6.37 ± 0.07	8.97 ± 0.25	1.21 ± 0.15	97.70 ± 43.10	125.52 ± 21.12
	Lolium multiflorum + Trifolium pratense	6.30 ± 0.06	8.68 ± 0.59	1.05 ± 0.04	78.37 ± 10.44	86.52 ± 5.89
	Trifolium pratense	6.40 ± 0.06	8.39 ± 0.33	1.10 ± 0.08	81.50 ± 22.70	96.02 ± 6.58
Limestone	*Lolium multiflorum*	6.97 ± 0.03	12.36 ± 0.42	1.25 ± 0.06	71.02 ± 3.66	113.63 ± 14.78
	Lolium multiflorum + Trifolium pratense	6.80 ± 0.06	10.00 ± 0.55	1.19 ± 0.07	67.40 ± 4.64	100.90 ± 28.79
	Trifolium pratense	6.38 ± 0.04	9.32 ± 0.10	1.09 ± 0.04	58.10 ± 3.12	73.72 ± 8.22
Dolomite	*Lolium multiflorum*	6.33 ± 0.06	9.10 ± 0.20	1.35 ± 0.07	76.50 ± 5.41	90.18 ± 6.56
	Lolium multiflorum + Trifolium pratense	6.40 ± 0.10	8.88 ± 0.02	1.19 ± 0.05	65.80 ± 2.80	70.45 ± 5.17
	Trifolium pratense	6.17 ± 0.07	8.84 ± 0.42	1.25 ± 0.18	96.32 ± 30.71	112.06 ± 35.87
Agricultural gypsum	*Lolium multiflorum*	6.50 ± 0.06	9.32 ± 0.16	1.07 ± 0.03	70.84 ± 11.32	144.70 ± 17.90
	Lolium multiflorum + Trifolium pratense	6.40 ± 0.06	9.09 ± 0.20	0.93 ± 0.02	65.01 ± 14.09	92.50 ± 9.62
	Trifolium pratense	6.27 ± 0.13	9.15 ± 0.36	0.70 ± 0.09	63.07 ± 4.40	89.22 ± 16.19

Note: Values are expressed as mean ± standard error (n = 3). Soil properties were determined after completion of the field experiment. Ca = exchangeable calcium; Mg = exchangeable magnesium; Available P = available phosphorus; Available K = available potassium.

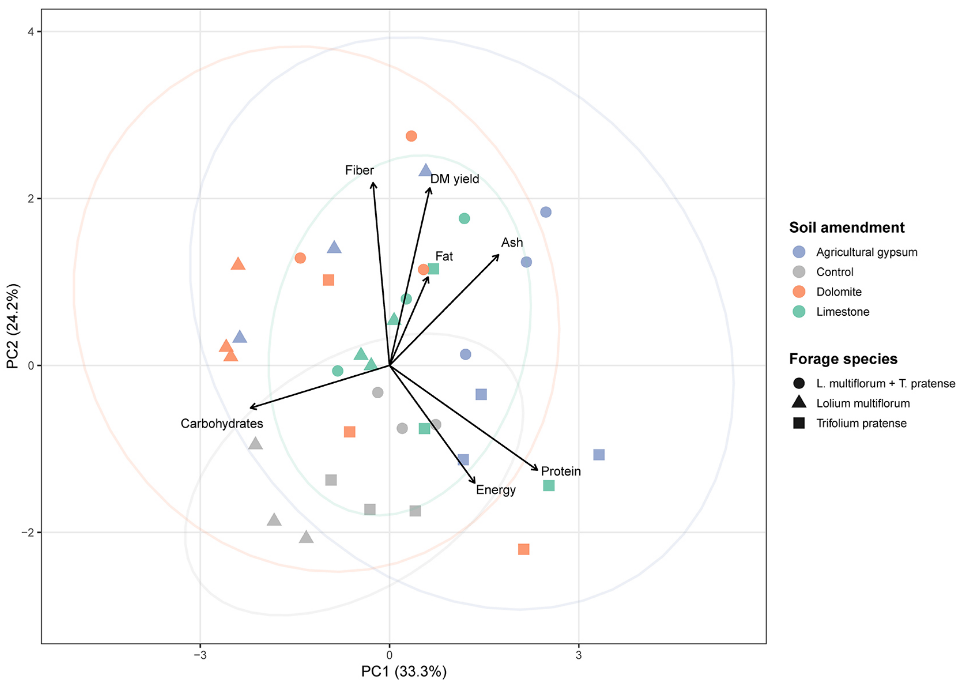

Figure 5. Principal component analysis (PCA) of forage nutritional composition and productivity-related traits under different soil amendments and pasture types

improved forage yield following calcium supplementation and soil acidity management in acidic environments (Minato et al., 2023; Makaza et al., 2026). From an animal nutrition perspective, higher forage biomass directly increases feed availability per unit area, which is a primary determinant of stocking rate and system productivity. Calcium plays a central role in root development, membrane function, and nutrient uptake efficiency, which translates into improved plant growth and forage supply under limiting soil conditions (Thor, 2019; Jing et al., 2024). The stronger responses observed under limestone and gypsum suggest that both pH correction and improved calcium availability contributed to higher forage production, thereby increasing the potential feed base for grazing animals. Gypsum may also enhance subsoil conditions and root exploration, improving access to water and mobile nutrients, as previously reported in forage systems (Moura et al., 2016; Souza et al., 2025).

Conversely, the relatively weaker effect of dolomite compared to limestone and gypsum may be associated with differences in its chemical structure, solubility, and dissolution dynamics under high-altitude environments. Dolomite ($CaMg(CO_3)_2$) possesses a dense crystalline structure that generally dissolves more slowly than calcite ($CaCO_3$) and agricultural gypsum ($CaSO_4 \cdot 2H_2O$), potentially delaying the release of calcium and the modification of soil chemical properties (Dugalić et al., 2024; Enesi et al., 2023). This interpretation is supported by the post-harvest soil chemical properties observed after the experiment (Table 5). Limestone produced the highest soil pH values and exchangeable calcium concentrations, whereas dolomite generated only minor changes relative to the control treatment. These results suggest that the slower dissolution of dolomite may have limited its capacity to modify soil chemical conditions during the evaluation period.

Under cold upland conditions, where mineral weathering processes may proceed more slowly, amendments with greater solubility could provide a more rapid supply of calcium during the establishment phase, potentially explaining the stronger vegetative responses observed in the present study (Morton, 2020; Puenayan Irua and Benavides Rosales, 2025). This interpretation is further supported by the post-harvest soil analysis (Table 5), which showed that soil pH and exchangeable calcium under the dolomite treatment remained close to those observed in the unamended control. Consequently, the observed treatment responses are consistent with meta-analytical and field evidence indicating that highly soluble calcium sources can promote faster improvements in nutrient availability and vegetation productivity than less reactive carbonate materials (Enesi et al., 2023; Leumassi Mbotchak et al., 2024).

Trifolium pratense showed the highest crude protein concentration, protein ratio, and protein-derived energy, confirming its value as a high-quality feed legume in ruminant nutrition. In contrast, *Lolium multiflorum* exhibited higher fiber and carbohydrate concentrations, indicating a forage profile more oriented toward structural carbohydrates and lower nitrogen content. These differences are consistent with established forage composition patterns, while legumes tend to provide more protein and grasses provide more structural fiber (Arias-Arredondo et al., 2025). The mixed pasture showed intermediate values, suggesting a practical balance between forage yield and protein supply, which is relevant for formulating more balanced grazing or cut-and-carry diets (Mi et al., 2025; Lüscher et al., 2014).

Although the total crude energy concentration remained relatively stable across treatments, the application of gypsum and limestone increased the proportion of energy derived from protein and the crude protein yield. These results suggest that soil amendments primarily influenced the nutritional composition of the forage rather than its total energy density. From an animal nutrition perspective, this change is particularly relevant, as greater protein availability promotes ruminal microbial activity, improves forage digestibility, and increases nitrogen utilization efficiency (Lima et al., 2023). Improvements in nutrient uptake and calcium-mediated physiological processes in plants could explain the increased protein accumulation observed in the amended soils (Jing et al., 2024; Zhang et al., 2025).

A key outcome of this study is the improvement in integrated feed-related indicators, particularly dry matter availability, crude protein yield, and livestock carrying capacity. These variables directly reflect the feed production potential of pasture systems (Macdonald et al., 2017). Unlike studies that focus primarily on forage biomass or individual nutritional traits, the present study demonstrated that mineral amendments simultaneously increased forage availability, protein production per unit area, and livestock support capacity. The higher carrying capacity observed under limestone and gypsum treatments indicates that soil fertility management can significantly increase the amount of livestock that can be supported per unit area without compromising forage supply (Espinoza et al., 2026). This is particularly relevant in high-Andean systems, where feed availability is often the main constraint for ruminant production systems. Similar relationships between pasture fertilization, forage yield, and stocking rate have been reported in grazing-based livestock systems (Cooke et al., 2025).

The post-harvest soil chemical data further supported the observed forage responses.

Limestone increased soil pH and exchangeable calcium concentrations more effectively than dolomite, whereas gypsum-maintained soil pH values similar to the control while sustaining exchangeable calcium concentrations during the evaluation period. These contrasting soil responses are consistent with the productivity patterns observed across treatments and provide a mechanistic explanation for the superior performance of limestone and gypsum.

The PCA confirmed the integrated nature of forage productivity and nutritional quality responses. Gypsum and limestone were associated with higher crude protein concentration, ash content, dry matter yield, and protein-derived energy, indicating a dual improvement in feed quantity and quality (Khan et al., 2025). In contrast, the control treatment was associated with higher carbohydrate accumulation and lower overall feed value. The mixed pasture showed an intermediate position, reflecting a practical compromise between forage yield and nutritive quality. More importantly, the ordination pattern suggests that forage productivity and nutritional quality responded in a coordinated rather than independent manner. This pattern is consistent with the simultaneous improvement of biomass production and feed value observed under calcium-based amendment treatments, generating an overall enhancement in pasture performance. The dispersion observed for dolomite may be related to differences in calcium–magnesium balance, which can influence nutrient uptake efficiency and forage composition in grassland systems (Gransee and Führs, 2013).

Although root development, nutrient uptake kinetics, and microbial activity were not directly measured, the observed improvements in forage productivity and nutritional quality are consistent with the known physiological functions of calcium in plant growth and nutrient acquisition. Therefore, the proposed mechanisms should be interpreted as biologically plausible explanations supported by the soil and forage responses observed in the present study rather than as directly verified processes.

From an applied animal production perspective, these findings highlight the value of combining soil fertility management with forage species diversification to improve feed availability and quality. In extensive grazing systems, particularly in mountain environments, increasing both forage supply and protein content is essential to improve animal performance and system efficiency (Duchicela et al., 2024). The integration of legumes such as *Trifolium pratense* with calcium-based soil amendments represents a practical strategy to enhance the nutritional value of pasture-based diets. Moreover, the comparable performance of gypsum and limestone suggests that improving calcium availability may be as important as soil acidity correction for increasing forage productivity in moderately acidic Andean grasslands. Together, these results demonstrate that integrated soil and pasture management can substantially enhance feed resources and livestock carrying capacity in high-Andean production systems.

CONCLUSIONS

Calcium-based soil amendments, particularly limestone and agricultural gypsum, significantly improved forage productivity and feed-related performance in high-Andean pasture systems. These amendments increased dry matter availability, crude protein yield, and livestock carrying capacity, demonstrating their effectiveness in enhancing both forage supply and feeding potential under moderately acidic soil conditions.

Pasture composition played a key role in determining forage nutritional quality. *Trifolium pratense* consistently improved crude protein concentration and protein-related nutritional indicators, while *Lolium multiflorum* contributed higher fiber and carbohydrate content. Mixed pastures provided an intermediate response, suggesting functional complementarity between grasses and legumes for balancing forage yield and nutritive value.

Overall, limestone and gypsum improved the quantity of feed available for livestock, while the inclusion of *Trifolium pratense* enhanced its nutritional quality, particularly protein supply. Therefore, the findings demonstrate that the combined use of soil fertility management and legume-based pasture systems as a practical strategy to improve feed availability and nutritional value in high-Andean livestock production systems.

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