

Aerial phytomass production and growth performances of five common tall species in Andean grasslands at harvesting and application of natural fertilizers

ABSTRACT

The objective of the research was to evaluate: aerial cover, canopy height and aerial phytomass production in five of the most common tall species in the area, in order to articulate it to a plant fiber production project. For the evaluation, 5 plots of 900 m² were fenced, including 50 subplots of 32 m², which were fertilized with cattle manure and phosphate rock. Monthly measurements were made from October 2020 to June 2021. At the end of the control, significant differences were observed for $P = .01$. Between plots they ranged from 1533 ± 508 to 2909 ± 192 cm² in canopy cover (CC), from 36.9 ± 0.89 to 38.3 ± 2.31 cm in canopy height (CH) and 44 ± 9.11 to 129 ± 9.11 g DM/plant in Aerial Phytomass Production (AFP). Between species, they ranged from 2067 ± 876 to 2975 ± 200 cm² in CC, from 33.2 ± 3.86 to 40.3 ± 1.24 cm in CH, and from 49.4 ± 11.52 to 162.4 ± 8.49 g DM/plant in AFP. By fertilization effect the AFP varied as follows: those fertilized with phosphate rock reached 109.5 ± 6.72 g DM/plant (14.7%), with cattle manure 88.8 ± 6.72 g DM/plant (-5.3%), and finally the control plants with 93.5 ± 15.03 g DM/plant, which means. A high canonical correlation was also observed between biological variables (canopy cover, canopy height, and dry matter production) and climatic variables (maximum temperature, minimum T. in °C, and rainfall in liters/m²). It was shown that tussock species respond quickly to cutting and natural fertilization has beneficial effects on biological indicators, despite the altitudinal difference between the plots and the particular species.

Keywords: biological indicators, fertilization, lignin content, environmental functions

1. INTRODUCTION

“Andean grasslands are vegetal formations covered by tall grassland species, mainly of the genera *Calamagrostis*, *Festuca* and *Jarava*” [1], which are consumed very little or almost nothing by animals due to their high cellulose and lignin content [2]; however, they fulfil very important environmental functions such as: carbon capture, regulation of the hydrological system, maintenance and protection of Andean biodiversity, soil cover, etc. [3]. Unfortunately, due to the fact that these ecosystems are underutilized by Andean cattle ranchers, they are threatened by burning, with the idea of provoking a tender regrowth that can be used for grazing [4]

“The anthropogenic action not only causes the loss of vegetation cover and accumulated organic carbon but also compromises the integral health of the ecosystem, affected by the degradation of the soil due to the heat generated by the fire, the loss and extinction of small grassland species, that were protected by the tall species, providing seed capable of being propagated in the overgrazed areas” [5], thus reducing the capacity for self-regeneration of the grassland ecosystem. In this understanding, “the alternative of converting Andean grasslands into a source of plant fiber as a building material for housing” [6], which it is being studied in another project, is being pursued. However, “the reversal of grassland burning, through the custody and maintenance of the ecosystem, to give it a beneficial use for both the grassland and the herders, requires the prior generation of knowledge on the

speed of regrowth growth, cover and net primary productivity that will serve as a basis for organizing strategies for the sustainable use of ecosystems with grassland formation” [4].

“The recovery capacity of manually cut grassland depends on several factors, however, for the present study, priority was given to explaining the phenomenon of growth in relation to climate, expressed in temperature and rainfall, to soil amendment through the application of cattle manure and rock phosphate” [7]. According to Jiang et al [8], “precipitation and temperature are the climatic factors involved in plant growth, and plants respond to the stimulation of soil water available to the roots”, however, “the increase of precipitation out of normal as well as the shortage of precipitation, on the other hand, plants are adapted to a comfortable temperature range that optimizes the development and life cycle of plants, limiting or nullifying this possibility in extreme conditions or outside the comfort range” [9-11].

“Human intervention through soil fertilization using natural inputs such as phosphate rock and livestock manure helps plant nutrition, depending on their composition and degree of mineralization” [12]. In this context, phosphate rock supplies highly significant microelements in plant nutrition in addition to inorganic phosphorus [13], and, due to its poorly soluble nature and slow mineralization, constitutes an effective fertilising option in the case of natural pastures [12]; meanwhile, livestock manure provides organic matter that enlivens the soil and the nitrogen necessary for plants [14]. This function varies according to the animal species and the diet they consume [2].

Experience similar to the case study in Peru is very scarce, but some studies related to the phenology of some species have been carried out, while other studies in Uruguay worked on N and P fertilization [13]. The net aerial primary production of these species in natural conditions was reported by Yaranga et al. [15] for *Calamagrostis intermedia* 383±18.6 grams of dry matter per plant (g DM/plant), for *C. antoniana* 313±17.6 g DM/plant, *Festuca rigidifolia* 216±23.1 g DM/plant, *Festuca sp* 182±24.3 g DM/plant, and *C. tarmensis* 104±21.6 g DM/plant.

In this context, the objective was to evaluate the aerial cover, canopy height, and aerial phytomass production of five common tall species in the area: *Calamagrostis intermedia* (paja chamik), *C. antoniana* (hatún pork'e), *C. tarmensis* (pork'e), *Festuca rigidifolia* (chilligua cachi), *F. sp* (chilligua), on plots fertilized with cattle manure and rock phosphate, during a 9-month control period.

2. MATERIALS AND METHODS

2.1 Study area

The study was carried out in the community of Acopalca in the central mountain range of Peru, located between UTM coordinates L18 S: 481880, E 8672695 and 4941157, E 8683594 between 3498 and 5510 meters above sea level (Figure 1), east of the Mantaro Valley. This Peasant Community is populated by rural families with cattle, sheep and alpacas. The pastures are distributed above 3800 m above sea level, but the study plots were located between 4012 and 4333 m altitude. In this environment the average seasonal temperature varies from -8°C at dawn to 16.2°C during the day in the dry period (May to September) and from 4°C to 12°C in the rainy period (October to April) and the average annual rainfall is 1170 mm per year.

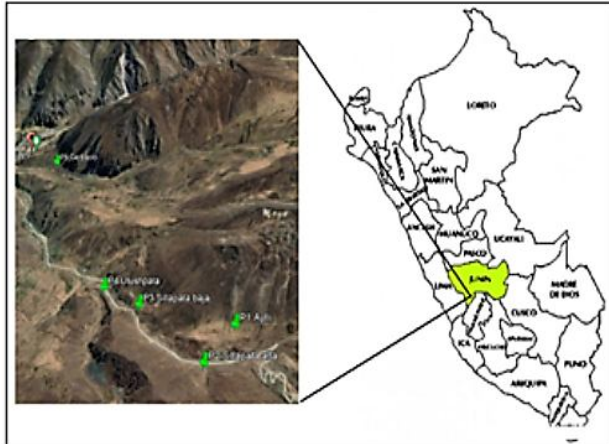


Figure 1. Location of the study area in the central Andes of Peru.

1.1 Data collection

The experimental plots were selected by convenience, taking into account the dominance of a species of interest in the area, where a 900 m² plot was fenced considering the method suggested by Otzen & Manterola [16], with wooden posts and barbed wire. Within the plot, 5 subplots of 64 m² were marked, dividing it into two halves, to be fertilized with a broadcast application of cattle manure (4000 kg per hectare) and phosphate rock (1500 kg/ha) with P₂O₅ from 18 to 22%, CaO from 28 to 30%, MgO from 0.5 to 0.6% and SO₄ from 3 to 5%. This action was replicated in five plots spaced 0.8 to 3 km apart.

Measurement of canopy cover and shoot canopy height

In each subplot and type of fertilization, 10 plants were randomly selected, i.e. 20 plants per subplot, 100 plants per plot, and 500 plants for the experiment. In addition, 10 additional plants were marked in each plot located outside the subplots that served as control plants or without fertilization effect. All selected plants, after identification of the species at the Universidad Nacional del Centro del Perú, were cut manually using a metal sickle, at an average height of 3 to 5 cm in September 2020. Data collection began 30 days after cutting. Firstly, the height of regrowth of the plant was measured in centimeters, with 3 measurements in different parts of the plant with the help of a pleximeter graduated in centimeters, recording the resulting average as data. Secondly, the average canopy cover of the leaves was measured, for which the vertical projection of the leaf flag was measured in centimeters, making two measurements in the form of a cross. With these data, the canopy cover was calculated by adopting the ellipsoidal shape of the plant, for which the following formula was used:

$$\text{Canopy cover (CC)} = r * r * \pi \quad [17]$$

Where r = radius, π = constant.

These measurements were performed monthly from October 2020 to June 2021

Measurement of phytomass production

The previous measurement in the month of June 2021 was carried out including the material cut from the monitored plant, in the same way as the initial cut, i.e. weighing the phytomass obtained in grams with the help of a digital hand scale with milligram precision, then the stems were separated from the leaves and weighed to obtain the yield of stems per plant. Finally, samples of the cut material per plant were extracted, packed and, after identification, sent to the Microbiology Laboratory of the Universidad Nacional del Centro del Perú for drying in an oven at 105 °C for 24 hours, for final weighing and to obtain the

percentage of dry material, with which the total dry weight of the plants in grams (DM) was calculated.

1.2 Data analysis

The data collected were arranged in double-entry matrices, the factors and variables: aerial cover, leaf height in rows and monthly data in columns; likewise the climatic factors: minimum and maximum landscape temperature and rainfall recorded in each plot in rows and respective data in columns, all in an Excel spreadsheet. The study hypotheses were analyzed by means of the "Generalised linear mixed model" method recommended by Dicoyskiy & Pedroza [16], using the free software Rstudio vs 4.1.2, using the following equation:

$$Y_{ijkl} = \mu + \Omega_i + \beta_j + \lambda_k + \varepsilon_{ijkl}$$

where:

Y_{ijkl} : Plant characteristic evaluated.

Ω_i : The effect of the plot on the plant characteristic evaluated.

β_j : The effect of the species

λ_k : The random effect of the plant characteristic evaluated.

ε_{ijkl} : Random effect of variation.

A canonical correlation analysis was also carried out between the biological variables: aerial cover, leaf height and phytomass production, with environmental variables: low temperature, maximum temperature, rainfall and the monthly amount of rain accumulated in each plot, using the PAS vs 3 software. 14, under the multiple linear correlation model: $X=(X_1, X_2, \dots, X_p)$ and $Y=(Y_1, Y_2, \dots, Y_q)$ recommended by Trendafilov & Gallo [18].

3. RESULTS AND DISCUSSION

3.1 Canopy cover

The aerial canopy cover of all plants had a rapid response, so in the first 60 days after cutting, the plants of the subplots fertilized with cattle manure (CM) presented an average canopy cover of 1397.11 cm² at the beginning of the control (a second month after cutting), which progressively increased to 1853.39 cm² at the eighth month of control, then decreased to 596.03 cm² at the ninth month. 39 cm² at the eighth month of control, then decrease to 596.03 cm² at the ninth month; in this same logic, the fertilized with rock phosphate (RP) varied from 1248.64 cm² at the beginning, 2565.29 cm² at the eighth month and 561.10 cm² at the ninth month; while, in the control plants (CP) without fertilization varied from 1056.36 cm² to 1495.69 cm² and 529.95 cm² (Figure 2a). The trend of canopy cover expansion showed a rapid recovery in the first two months after cutting due to the reserve of nutrients that the plants stored naturally, then it slowed down in the following months, until March 2021 in which the acceleration of the expansion of the cover is again observed due to the effect of the rainy period (April - May), to suffer a sharp drop in values in the month of June 2021, due to the effect of the dry period and low temperature. On statistical analysis, the highest aerial canopy cover was achieved by those subplots fertilized with phosphate rock with 2783±162 cm² for $P = .01$, then those fertilized with cattle manure with 2199±166 cm², and finally the control subplots with 1866±428 cm² (Figure 3. b). This indicates that the application of natural manures had positive effects on shoot development, which was evidenced by the statistical difference obtained.

Regarding the aerial canopy cover observed between plots, it has been noted that no similar trend was observed for the effect of the fertilizers applied, as each plot showed a different result (Table 1). The response up to the time of the first control was notoriously low in Otush Palla (P4) with only 370.28 cm² of canopy cover, while in Sillapata Alta (P2) and Sillapa Baja (P3) started with more than 1024 cm², and the plots in Aylli (P1) and Gerbacio (P5) showed an initial expansion bordering 1860 cm². Likewise, the time at which they reached the greatest expansion of canopy cover was also heterogeneous, as in P1 it reached the seventh month of control, in P2 and P3 the eighth month, and in P4 and P5 the ninth month. At the statistical analysis for the end of the measurement period, plot P1 showed the greatest expansion for $P = .05$ with 2909 ± 192 cm², followed by plots P3 and P2 with averages of 2480 ± 207 cm² and 2109 ± 204 cm², and finally plots P4 and P5 with averages of 1818 ± 802 cm² and 1533 ± 508 cm² (Figure 3. a). The difference in canopy cover was due to the heterogeneity of the genetic and morphological characteristics of the dominant species present in each plot.

In terms of species, the aerial canopy cover was as follows: *Calamagrostis intermedia* showed height of 2897.06 cm at the first control after cutting, reaching 4081.46 cm² at the ninth month of recovery, in the same sense, in *C. antoniana* it changed from 2162.87 to 4464.10 cm², in *C. tarmensis* from 1050.76 to 4934.05 cm², in *Festuca rigidifolia* from 1519.83 to 2567.31 cm² and in *F. sp* from 835.30 to 2951.21 cm². At the statistical analysis of the final measurement in June 2021, the species *C. intermedia* reached the highest extent of aerial canopy cover, with statistical significance for $P = .05$, was with the average of 2975 ± 200 cm², followed by *C. antoniana* with 2642 ± 277 cm², finally followed by *F. sp*, *F. rigidifolia* and *C. tarmensis* with 2089 ± 272 , 2082 ± 193 and 2067 ± 876 cm² respectively, which behaved as similar (Figure 3. e). The higher significant difference of the first two named species is due to the morphological characteristic of plants with higher number of tillers, wider leaves and taller stems.

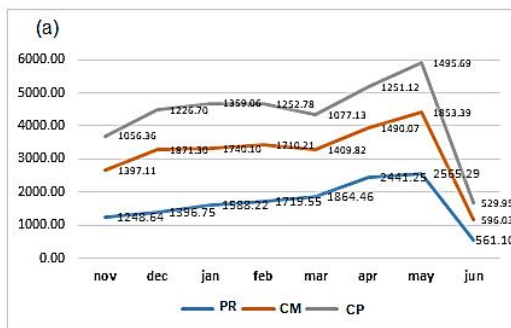


Table 1. Monthly canopy cover of plant according to plots

	P1	P2	P3	P4	P5
nov	1055.53	1860.07	1859.71	370.28	1024.60
dec	1214.23	2192.58	2273.26	602.96	1208.23
jan	1515.65	2315.67	2194.41	548.95	1237.61
feb	1678.06	2201.23	2052.65	570.40	1301.90
mar	1604.26	1718.63	1971.94	573.60	1383.92
apr	2689.21	1958.75	2031.16	635.43	1322.86
may	2268.10	2385.26	2920.70	799.50	1483.71
jun	2338.98	1976.32	2515.95	902.04	1484.98

Figure 2a. Evolution of aerial canopy cover according to the type of fertilization and, table 1 Evolution of aerial canopy cover according to plots.

“The morphological indicators of some improved forage grasses have often been measured to evaluate the forage quality and phenological evolution of certain ecotypes adapted to specific environmental conditions using various morphological descriptors, mainly related to the characteristics of leaves and stems, as well as their dry matter production capacity and nutritional quality for livestock” [19, 20]; However, these measurements have not yet been extensively developed on native species such as high Andean grasslands, as the scarce information available is oriented towards the forage value of those species that are highly desirable for livestock, such as the study carried out in Uruguay [21] [22], with the

exception of net aerial primary productivity, which was studied by Yaranga et al [15]. This situation does not allow us to make objective comparisons with the results of this research.

In this sense, we can affirm that the application of rock phosphate to the soil benefited the development of the shoots by increasing the growth of the leaves and, therefore, the gain of greater canopy weight [23]. The greater leaf size and weight facilitated leaf folding whereby plants expanded their aerial cover, which was visually observed when plants with longer leaves tended to fold at a lower height than those with smaller leaves. “The increased plant development in the subplots fertilized with rock phosphate must have been enhanced by the macronutrients provided by phosphate, which are indispensable for key metabolic processes in plants, such as cell division, energy generation, macromolecule biosynthesis, membrane integrity, signal conductivity, photosynthesis and plant respiration” [24]. According to some literature, the action of phosphate is favored when it starts its mineralization process, which must have happened due to the action of organic acids, enzymes, and ion chelators that were secreted by soil microorganisms [23] [25] [24], favored by the soil moisture that was maintained in September and October due to the effect of the beginning of the rainy season in the control plots and the pulverized condition of the phosphate rock; However, some authors have reported that the mineralization process is very slow and takes longer than 3 years [26] [13], which suggests that the benefit of fertilization with this material will be long lasting and sustainable; this does not agree with the report of Zong et al [27] who reported that the application of rock phosphate in alpine steppes did not help the formation of leaves.

The application of cattle manure in the subplots also benefited shoot development, although to a lesser extent than rock phosphate. In this case, the beneficiary elements would have been soluble elements such as carbon (C), nitrogen (N), soluble phosphorus (P), and sulfur (S) [28] [29] which were washed away by the rainwater that had started in September and October 2021. The primary element involved in the development mainly of the leaves was undoubtedly nitrogen [30], the intensity of which depended on the asynchrony of the species [27]. However, the process of decomposition of non-lignified cellulose and hemicellulose, and lignin, as well as mineralization of all insoluble plant organic matter, will take longer depending on the course of local temperature and humidity in the following months and years [29] [31].

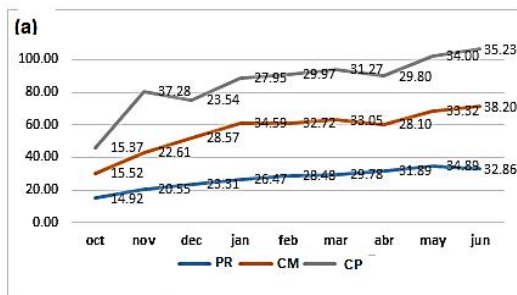
With respect to the behavior of the species, *C. intermedia* and *C. antoniana* showed greater aerial canopy cover, due to the length of the leaves, which ranged from 30 to 45 cm, and the weight of the leaves, which were wider than in the other species. This structural difference of the plants affected the folding of the leaves at a lower height gain, although to a lesser extent than due to the effect of the rock phosphate. The *Festuca* showed smaller leaves that were more erectile, so their canopy cover expansion was lower [32], and finally the species *C. tarmensis* showed thinner and semi-rigid leaf blades [33], which also did not allow a greater expansion of the canopy cover. In the field, it has been observed that the latter species grows at a higher plant density, which does not allow it to develop a greater canopy projection [33].

3.2 Canopy height

The canopy height gained by the shoots was not homogeneous, so in the subplots without fertilizer application, they ranged from 15.37 cm in the first control to 35.23 cm at the

ninth month of control (Figure 2. b); whereas, plants from subplots fertilized with cattle manure progressed from 15.52 to 38.20 cm; plants from subplots fertilized with rock phosphate from 14.92 to 34.89 cm. At the statistical analysis of the measurement in the ninth month, the highest height for $P = .05$ was achieved by the plants fertilized with cattle manure with an average of 40.2 ± 0.72 cm, followed by those plants without fertilization with an average of 38.2 ± 1.82 cm and finally those fertilized with rock phosphate with 33.8 ± 1.82 cm height (Figure 3b). Meanwhile, the behavior of canopy height at plot level, no significant difference was found for $P = .05$ whose averages were for P1 = 36.9 ± 0.89 cm, P2 = 36.4 ± 0.95 cm, P3 = 37.8 ± 0.96 cm, P4 = 32.8 ± 3.62 cm, and P5 = 38.3 ± 2.31 cm (Figure 3. d). The trend of variation in each plot is it shown in Figure 2b (Table 2).

Regarding the behavior between species, the greatest canopy height was shown by the species *C. antoniana*, which was superior for $P = .01$ with an average of 40.3 ± 1.24 cm, followed by *C. intermedia* with 38.5 ± 0.91 cm, then *F. sp* with 35.2 ± 1.22 cm, followed by *F. rigidifolia* with 35.1 ± 0.88 cm and finally *C. tarmensis* with 33.2 ± 3.86 cm for $P = .05$ (Figure 3. f). These differences are based on the morphological genetics of each species, which differentiates them in the robustness of the leaves and stems and the optimal growth size.



(b) Table 2. Average monthly plant height according to plots (cm)

	P1	P2	P3	P4	P5
oct	11.80	18.89	14.91	12.39	18.36
nov	40.20	23.45	23.26	14.57	21.59
dec	23.47	26.25	26.64	20.78	28.56
jan	26.37	32.72	35.45	22.38	31.44
feb	29.24	29.83	34.74	24.82	33.31
mar	29.69	31.44	34.58	25.39	35.75
apr	28.27	29.93	31.12	24.25	36.07
may	30.36	33.33	37.51	30.56	38.60
jun	36.31	35.74	38.74	29.59	36.77

Figure 2b. a) Evolution of canopy height according to the type of fertilizer applied (b) Evolution of canopy height according to the location of plots

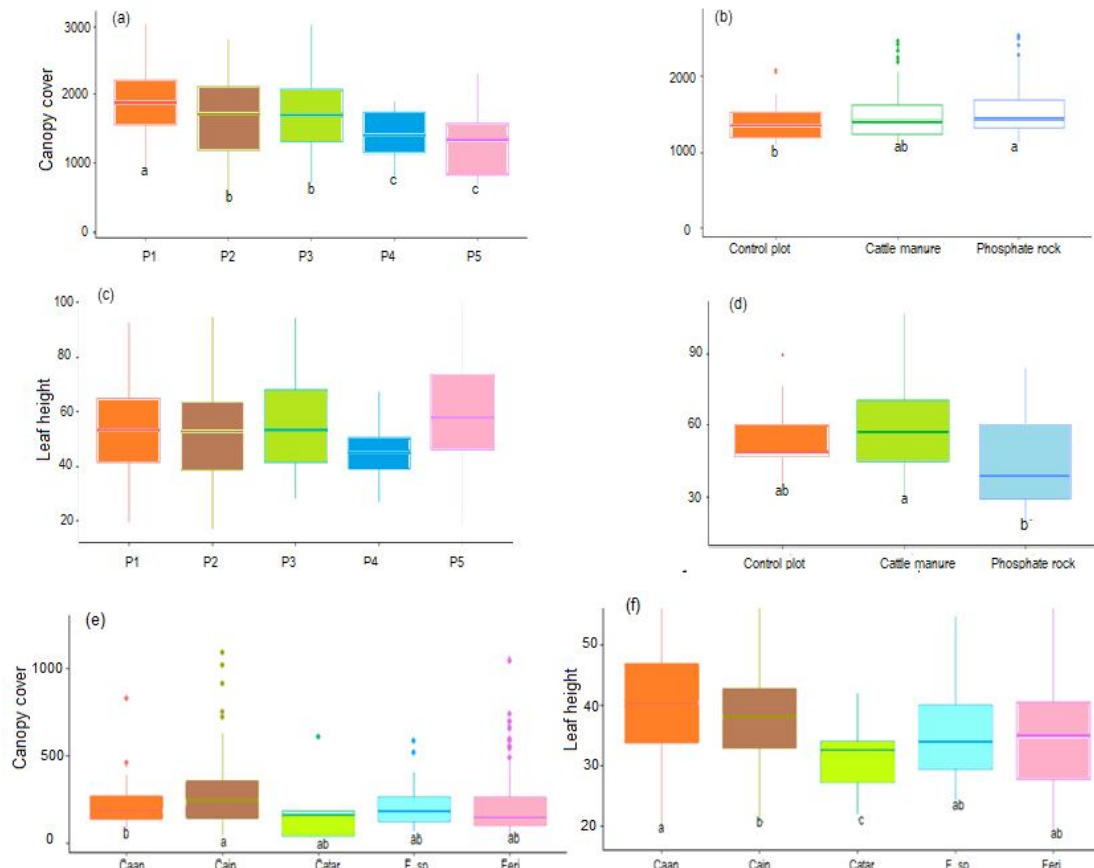


Figure 3. Least Significant Difference (LSD) of: a) aerial coverage by plots, b) canopy cover by fertilizer, d) leaf height by plots, c) leaf height by fertilizer.

The canopy height was higher in those subplots where cattle manure was applied, which provided soluble N and P when washed by rainwater [34] and also favored soil bacterial growth, through enzymatic activities and provision of microelements [2], which would have positively favored vertical growth with more solid leaves. In this way, they gained greater canopy height, while fertilizers with rock phosphate allowed the growth of less solid leaves that rapidly curved, losing vertical stability [14]. However, the comparison between plot locations showed no significant difference, because the differences found in canopy cover and canopy height were apparently homogeneous in the five plots despite the difference in altitudinal gradient. The combination of data from subplots with and without fertilization masked the specific difference observed due to the effect of the natural fertilizers applied. This statistical behavior suggests that generalized comparisons based on accumulated data from different interventions within a plot are not consistent for an interpretation of the effect of the altitudinal gradient.

“The difference in canopy height between species is based on the morphological nature of the species, as *C. antoniana* and *C. intermedia* are very tough and leathery species, with more solid canes” [33], while the others have thin leaves and thinner canes [32] [15].

3.3 Net aerial primary productivity

The net aerial primary productivity of the shoot of the different species cut reached the following average values: For the effect of the applied fertilizer, it has been observed that

there was no significant difference between subplots with and without natural fertilization for $P = .05$, however, in the subplots fertilized with phosphate rock reached the average of 109.5 ± 6.72 g DM/plant, in those fertilized with cattle manure it reached 88.8 ± 6.72 g DM/plant and finally the plants without fertilization reached 93.5 ± 15.03 g DM/plant (Figure 4. a)

Among the location of the plots, it was observed that P1, P2, and P3 were superior for $P = .01$ with 129 ± 9.11 , 125.2 ± 9.11 , and 124.2 ± 9.11 g DM/plant respectively, followed by P5 with 70.7 ± 9.11 g DM/plant, finally P4 with 44 ± 9.11 g DM/plant (Figure 4. b). Regarding the species of the plants under study, it has been observed that, *C. intermedia* (Cain) species was superior for $P = .05$ with 162.4 ± 8.49 g DM/plant, followed by *C. antoniana* (Caan) with 109.6 ± 7.60 g DM/plant, then by *F. rigidifolia* (Feri) with 81.3 ± 8.48 g DM/plant, finally the species *F. sp* (F. sp) with 59.9 ± 9.14 g DM/plant, and *C. tarmensis* (Cata) with 49.4 ± 11.52 g DM/plant with statistical similarity (Figure 4. c).

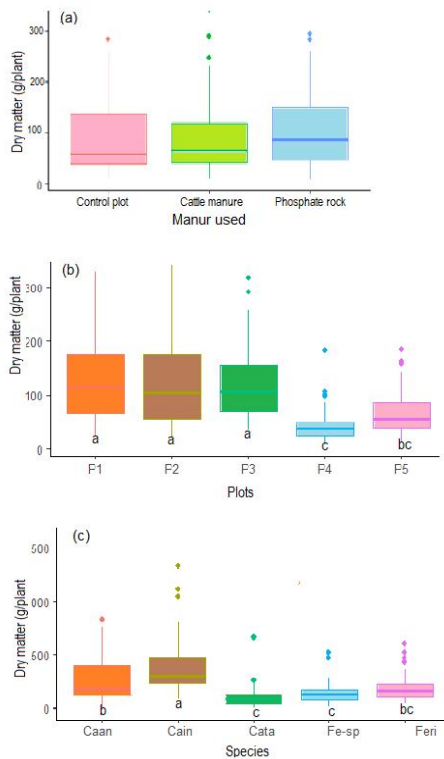


Figure 4. LSD of net aerial primary production: a) effect of applied fertilizer, b) effect of plot location, and c) effect of plant species.

However, the difference in aerial net primary production observed between plots was due to the particular characteristics of each species that were dominant in each particular case such as: the length and width of the leaves, height, and thickness of the stems, the number of bushes, in addition to the soil characteristics [15], in this understanding the first three plots in the production of dry matter, is based on: a) the species *C. intermedia* and *C. antoniana* are plants with greater tillering with more solid and thicker leaves and stems that were present in P1, P2, and P3, as opposed to the species *C. tarmensis*, which is characterized by a morphology contrary to that mentioned by Tovar [33] and was present in plot P4, b) plots P4 and P5 are located on shallower soils with a light brown color, which is indicative of soils with a lower concentration of carbon, organic matter, and

nitrogen, due to their higher concentration of silt and clay particles containing Fe oxides [37]. This corroborates that soil characteristics determine not only plant cover but also plant productive capacity [28] [29]. Finally, it was found that the harvested dry matter production of leaves and shoot stems were not similar to the original production observed by Yaranga et al. [15] who evaluated seven species including the species treated in this research under original conditions and found the following: *C. intermedia* with a production of 383 ± 18.6 g DM/plant (equivalent to 57.6% more than our result), *C. antoniana* 313 ± 17.6 g DM/plant (+64.09%), *F. rigidifolia* 216 ± 23.1 g DM/plant (+62.5%) *F. sp* 182 ± 24.3 (+68.01%), and *C. tarmensis* 104 ± 21.6 g DM/plant (+52.5%). This level of difference between the primary production obtained in original conditions that reached between 52.5% to 64.09% more than that obtained in the present investigation, would reaffirm the position of Wang et al. [11] who reported that the recovery of plants harvested by cutting or other anthropogenic actions reduces the productive capacity compared to that of origin; but as we do not have data until the moment of the stabilization of the growth of the species studied, we cannot yet affirm the percentage dimension of this reduction. However, it is necessary to mention that in the field observation after the end of the controls it has been noted that the shoots continued to grow, which allows us to affirm that the period of measurement carried out until the ninth month of control was premature and was not enough to know the necessary time in which the stabilization of growth occurs; this because the control plots were invaded by cattle grazing in the surrounding areas, which was the reason for the financial cut. The comment suggests that further measurements should be scheduled beyond 12 months.

3.4 Canonical correlation between biological and environmental variables

In the understanding that, rainfall is one of the most important variables in plant development, it has been observed that the effect of the altitudinal gradient in the level of rainfall was very marked among the control plots; On the other hand, the rainfall distribution pattern during the year showed an abnormal distribution, since the rainfall in the month of November 2020 was zero mm, likewise the rainfall in the month of February 2021 was scarce, which did not correspond to the referential pattern of rainfall observed in the last 30 years, in which the maximum level of rainfall occurs in the month of February. Figure 5. a, shows that in the month of February only 41.58 l/m² was recorded compared to the month of January when 164.57 l/m² was registered, while the months of March and April reached between 77.99 and 72.61 l/m², and the drop to 6.71 and 1.59 l/m² in the months of April and May are normal characteristics of the dry seasonal period [15].

Under these conditions, the canonical correlation was evaluated between the climatic variables: minimum temperature, and maximum temperature both in °C registered at landscape level, at the Acopalca Meteorological Station of the National Service of Meteorology and Hydrology of Peru, and with the monthly rainfall registered in each plot in liters/square meter (l/m²); with the biological variables: canopy cover in cm² and canopy height in cm. As they remain relatively close to the quadrant origin during the rainy season, the biological variables have not clearly exhibited a link with the rainfall variable; however, at the factor level, the rainy months and maximum temperature show some propensity to be associated in the fourth quadrant (Figure 5. b), it was also observed that the month of June showed a high positive correlation with low temperature, which means that, the more scarce the rainfall the lower the temperatures and this fact directly affects the

production of aerial biomass. The canonical correlation was explained with 92.58% of the data in axis 1.

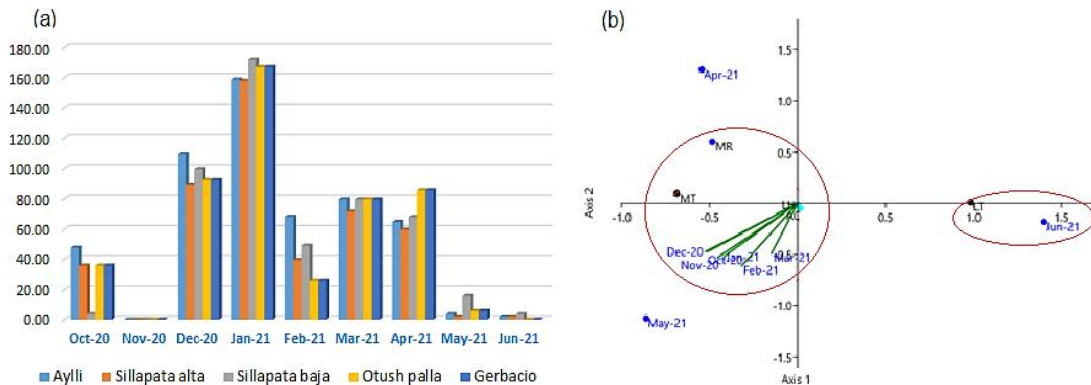


Figure 5. a) Distribution of monthly rainfall in each plot (P1= Aylli, P2= Sillapata alta, P3= Sillapata baja, P4 Otushpalla and P5= Gerbacio), b) canonical correlation between climatic variable: MT = maximum temperature, LT = low temperature, and MR= mounthly rainfall; and biological variables: CC= canopy cover, LH = leaf height.

Many studies on “global climate change affecting precipitation and temperature reveal that the normal distribution pattern observed over the last 20-30 years is becoming increasingly irregular” [38] [39], which is most pronounced at the local scale, as has been observed in conditions in Acopalca, where the level of precipitation was abnormal in November and February. This irregularity directly affects crop and grassland production yields in general [23] [38]; however, “the effect of this abnormality on shoot growth was not very evident, because soil moisture remains beyond the rainy days, as long as the shortage is not too prolonged” [40].

“The rainy months (October to March) and the maximum temperature, were the closest parameters that favored the development of canopy cover and canopy height” [33] [41], due to the higher concentration of chlorophyll in the plants in these periods, which allows for greater growth and accumulation of dry matter [42], meanwhile, “the less rainy months showed, on the contrary, the disarticulation of these parameters with the biological variables, which would explain that plants continue to grow or remain alive due to the morphological transformation of grassland species, expressed in the increased vertical growth of roots in search of water” [43] [5].

4. CONCLUSIONS

The application of rock phosphate and cattle manure on tussock Andean grassland species benefited the increased shoot development of *Calamagrostis antoniana*, *C. intermedia*, *Festuca rigidifolia*, *F. sp* and *C. tarmensis*, significantly for $P = .01$ in canopy expansion and canopy height, compared to control subplots, assessed up to nine months after cutting; also, the difference between plot locations was influenced by the variation of dominant species in each particular plot and the significant difference between them.

The net aerial primary productivity evaluated in g DM/plant did not show significant differences for $P = .05$ between those that were or were not fertilized with rock phosphate, and cattle manure, nor between the location of the plots; however, significant differences

were found for $P = .05$ between species, due to the morphological differences that characterize each species in particular, such as *C. intermedia* and *C. antoniana* are larger species, with longer and more leathery leaves and taller stems, compared to the others. On the other hand, it is necessary to mention that the response to the application of natural fertilizers is still incipient, due to the control time that was insufficient for the optimal mineralization process so that a proper evaluation of grassland species with the application of the fertilizers used should be carried out in a longer period.

The development of grassland species has also been observed to be somewhat correlated with rainfall and high temperatures, albeit not significantly, as the plants have continued to grow during periods of low rainfall and low temperatures as a result of their ecological adaptation to shifting altitude, soil, and temperature conditions.

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References

1. Yaranga R, Custodio M, Chanamé F, Pantoja, R. Diversidad florística de pastizales según formación vegetal en la subcuenca del río Shullcas, Junín, Perú, *Scientia Agropecuaria*, 2018; 9(4): 511–517. <https://doi.org/10.17268/sci.agropecu.2018.04.06>.
2. Wang J, Wang D, Li C, Seastedt TR, Liang C, Wang L, Sun W, Liang M, Li Y. Feces nitrogen release induced by different large herbivores in a dry grassland. *Ecol Appl*, 2018; 28: 201-211. <https://doi.org/10.1002/eap.1640>
3. Bremer LL, Farley K, DeMaagd, Suárez E. Biodiversity outcomes of payment for ecosystem services: lessons from páramo grasslands, *Biodiversity and Conservation*. Springer Netherlands, 2019; 28(4): 885–908. <https://doi.org/10.1007/s10531-019-01700-3>.
4. Yaranga R. *Ecosistemas de pastizal altoandino*. Edited by Centro de Investigación en Alta Montaña (CIAM) UNCP. Huancayo, Perú: 2019. ISBN: 9786124769740.
5. Jung EY, Gaviria J, Sun S, Engelbrecht B M. Comparative drought resistance of temperate grassland species: testing performance trade-offs and the relation to distribution. *Oecologia*, 2020; 192(4): 1023–1036. <https://doi.org/10.1007/s00442-020-04625-9>.
6. Velásquez S, Pelaes G, Gerañdo D. Uso de materiales vegetales en materiales compuestos de matriz polimérica: una revisión con miras a su aplicación en el diseño de nuevos productos. *Informador técnico* (Colombia), 2016; 80(1): 77-86.
7. Dita A, Muhammad J, Mtiaz M, Mehmood S, Qian Z, Tu S. Application of rock phosphate enriched composts increases nodulation, growth and yield of Chickpea. *International Journal of Recycling of Organic Waste in Agriculture*. 2018; 7:33–40. <https://doi.org/10.1007/s40093-017-0187-1>
8. Jiang C, Zhang H, Tang T, Labzovskii L. Evaluating the coupling effects of climate variability and vegetation restoration on ecosystems of the Loess Plateau, China, *Land Use Policy*. 2017; 69: 134-148
9. Liu W, Lü X, Xu W, Shi H, Hou L, Li L, Yuan W. Effects of water and nitrogen addition on ecosystem respiration across three types of steppe: The role of plant and microbial biomass. *Science of The Total Environment*, 2018; (619-620): 103–111. <https://doi.org/10.1016/j.scitotenv.2017.11.1>.
10. An S, Chen X, Zhang X, Lang W, Ren S, Xu L. Precipitation and minimum temperature are primary climatic controls of alpine grassland autumn phenology on the Qinghai-Tibet plateau. *Remote Sensing*, 2020; 12(3). <https://doi.org/10.3390/rs12030431>.

11. Wang H, Liu H, Cao G, Ma Z, Li Y, Zhang F, Zhao X, Zhao X, Jiang L, Nathan J, Sanders S, Classen A, He J. Alpine grassland plants grow earlier and faster but biomass remains unchanged over 35 years of climate change. *Ecology Letters*. 2020; 701-701. <https://doi.org/10.1111/ele.13474>.
12. Jouany C, Morel C, Ziadi N, Bélanger G, Sinaj S, Stroia C, Cruz P, Theau JP, Duru M. Plant and soil tests to optimize phosphorus fertilization management of grasslands. *European Journal of Agronomy*, 2021; 125. <https://doi.org/10.1016/j.eja.2021.126249>
13. Zapata F, Roy R. *Utilización de las rocas fosfóricas para una agricultura sostenible. Organización de las Naciones Unidas para la agricultura y la alimentación*. FAO, Roma. 2007; 94. ISBN:978-92-5-305030-7.
14. Elouear Z, Bouhamed F, Boujelben N, Bouzid J. Application of sheep manure and potassium fertilizer to contaminated soil and its effect on Zinc, Cadmium and lead accumulation by alfalfa plants, *Sustainable Environment Research*. 2016; 26: 161-135. <http://dx.doi.org/10.1016/j.serj.2016.04.004>.
15. Yaranga R, Art VanVure, Fuentes A, Fuentes R, Maraví K, Román M, Cáceres D, Fuentes CA. Andean Grassland Species: Net Aerial Primary Productivity, Density, Ecomorphological Indices, and Soil Characteristics, *Journal of Ecological Engineering*. 2021; 22(10): 163–175. <https://doi.org/10.12911/22998993/138816>.
16. Otzen T, Monterola C. Técnicas de Muestreo sobre una Población a Estudio. *Int. J. Morphol*. 2017; 35(1): 227-232.
17. Botana F, Recio T. A propósito de la envoltura de una familia de elipses. *Bol. Soc. Puig Adam*. 2013; 95: 15-30
18. Trendafilov N, Gallo M. *Canonical correlation analysis (CCA). In: Multivariate Data Analysis on Matrix Manifolds*. Springer Series in the Data Sciences. Springer, Cham, 2021; 450. https://doi.org/10.1007/978-3-030-76974-1_8.
19. Morales CR, Avendaño A, Melgoza G, Vega K, Quero A, Martínez M. Caracterización morfológica y molecular de poblaciones de pasto banderita (*Bouteloua curtipendula*) en Chihuahua, México. *Rev. Mex. Cienc. Agr*. 2016; (7):455-469.
20. Morillo AC, Tovar YP, Morillo E. Caracterización morfológica de *Selenicereus megalanthus* (K. Schum. ex Vaupel) Moran en la provincia de 25 Lengupá. *Ciencia en Desarrollo*. 2016; 7:23-33.
21. Del Pino A, Lezama F, Pezzani F, Parodi G. Persistencia de efectos a largo plazo de la fertilización fosfatada y la introducción de leguminosas en pastizales del Uruguay. *Agriscientia* 2021; 38 (1): 99-109. <http://dx.doi.org/10.31047/1668.298x.v38.n1.26856>.
22. Panella PG, Cardozo G, Cuadro R, Reyno R, Lezama F. La fertilización fosforada disminuye la riqueza y aumenta el número de especies exóticas de plantas en pastizales intersebrados con leguminosas. *Ecología Austral*. 2020; 30 (3): 331-496. <https://doi.org/10.25260/EA.20.30.3.0.1063>
23. Carlyle CN, Fraser LH, Turkington R. Response of grassland biomass production to simulated climate change and clipping along an elevation gradient. *Oecologia*. 2014; 174(3): 1065–1073. <https://doi.org/10.1007/s00442-013-2833-2>.
24. Rawat P, Das S, Shankhdhar D, Shankhdhar SC. Phosphate-solubilizing microorganisms: Mechanism and their role in phosphate solubilization and uptake. *Journal of Soil Science and Plant Nutrition*, 2021; 21: 49-68. <https://doi.org/10.1007/s42729-020-00342-7>.
25. Rolando JL, Turin C, Ramírez DA, Mares V, Moneris J, Quiroz R. Key ecosystem services and ecological intensification of agriculture in the tropical high-Andean Puna as affected by land-use and climate changes. *Agriculture, Ecosystems and Environment*. 2017; 236(1), 221–233. <https://doi.org/10.1016/j.agee.2016.12.010>.
26. Rajan SS, Fox RL, Saunders WM. Influence of pH, time and rate of application on phosphate rock dissolution and availability to pastures - II. Soil chemical studies. *Fertilizer Research*. 199; 28(1): 95–101. <https://doi.org/10.1007/BF01048860>.
27. Zong N, Hou G, Shi P, Zhou T, Yu J, Tian J. Different responses of community temporal stability to nitrogen and phosphorus addition in a non-degraded alpine grassland, *Ecological Indicators*. 2022; 143. <https://doi.org/10.1016/j.ecolind.2022.109310>.
28. Mulyani O, MacHfud Y, Setiawan A, Joy B. Potential of local organic matters in Jatinangor West Java Indonesia as raw materials for organic fertilizer. *IOP Conference Series: Earth and Environmental Science*. 2019; 393(1): 0 – 9. <https://doi.org/10.1088/1755-1315/393/1/012048>.
29. Marzi M, Shahbazi K, Kharazi N, Rezaei M. The influence of organic amendment source on carbon and nitrogen mineralization in different soils. *Journal of Soil Science and Plant Nutrition*. 2020; 20(1): 177–191. <https://doi.org/10.1007/s42729-019-00116-w>.
30. Altamirano A, Ircañaupa W. Fenología de cinco especies forrajeras en los bofedales de la cuenca alta del río Ichu y Pampas. *Ecología, Revista Peruana de Innovación Agraria*. 2020; 1(1): 38-50
31. Ma Q, Tang S, Pan W, Zhou J, Chadwick DR, Hill PW, Wu L, Jones DL. Effects of farmyard manure on soil S cycling: Substrate level exploration of high- and low-molecular weight organic S

- decomposition. *Soil Biology and Biochemistry*. 2021; 160. <https://doi.org/10.1016/j.soilbio.2021.108359>.
32. Catorci A, Tardella FM, Velasquez JL, Cesaretti S, Malatesta L, Zeballos H. How environment and grazing influence floristic composition of dry Puna in the southern Peruvian Andes. *Phytocoenologia*, 2014; 44(1–2): 103–119. <https://doi.org/10.1127/0340-269X/2014/0044-0577>.
 33. Tovar O. *Las gramíneas (Poaceae) del Perú*. Monografías del Real Jardín Botánico, RUIZIA Tomo 13, Madrid. 1993; 481.
 34. Das A, Patel DP, Lal R, Kumar MG, Layek, J, Shivakumar BG. Impact of fodder grasses and organic amendments on productivity and soil and crop quality in a subtropical region of eastern Himalayas, India. *Agriculture, Ecosystems and Environment*. 2016; 216: 274–282. <https://doi.org/10.1016/j.agee.2015.10.011>.
 35. Garay J, Bautista Y, Bernal A, Mendoza S, Martínez J, Sosa E, Joaquin S. Forage yield and quality of buffel ‘H-17’ and Urochloa hybrids at different regrowth ages under semi-arid conditions. *Grassland Science*. 2020; 66(4): 277-284. <https://doi.org/10.1111/grs.12278>.
 36. Mastalerczuk G, Borawska-Jarmułowicz B. Physiological and Morphometric Response of Forage Grass Species and Their Biomass Distribution Depending on the Term and Frequency of Water Deficiency. *Agronomy*. 2021; 11(12): 2471. <https://doi.org/10.3390/agronomy11122471>.
 37. Moritsuka N, Matsuoka K, Katsura K, Yanai J. Farm-scale variations in soil color as influenced by organic matter and iron oxides in Japanese paddy fields, *Soil Science and Plant Nutrition*. 2019; 65(2): 166-175. <https://doi.org/doi/10.1080/00380768.2019.1583542>.
 38. Adu-Boahen I K, Yaw I, Akugre M. Climatic Variability and Food Crop Production in the Bawku West District of the Upper East Region of Ghana. *Ghana Journal of Geography*. 2019; 11(1): 103 – 123. <https://dx.doi.org/10.4314/gjg.v11i1.7>.
 39. Méndez M, Calvo-Valverde LA, Maathuis B, Alvarado-Gamboa LF. Generation of Monthly Precipitation Climatologies for Costa Rica Using Irregular Rain-Gauge Observational Networks, *Water*. 2019; 11(1): 70. <https://doi.org/10.3390/w11010070>
 40. Tenelanda-Patiño D, Crespo-Sánchez P, Mosquera-Rojas G. Umbrales en la respuesta de humedad del suelo a condiciones meteorológicas en una ladera Altoandina. *Maskana*. 2018; 9(2): 53–65. <https://doi.org/10.18537/mskn.09.02.07>
 41. Muñoz J. Regeneración Natural : Una revisión de los aspectos ecológicos en el bosque tropical de montaña del sur del Ecuador. *Bosques Latitud Cero*. 2017; 7(2): 130–143.
 42. Chang J, Ciais P, Viovy N, Soussana JF, Klumpp K, Sultan B. Future productivity and phenology changes in European grasslands for different warming levels: Implications for grassland management and carbon balance. *Carbon Balance and Management*. 2017; 12(1). <https://doi.org/10.1186/s13021-017-0079-8>.
 43. Padilla FM, Mommer L, De Caluwe H, Smit-Tiekstra AE, Visser EJ, De Kroon H. Effects of extreme rainfall events are independent of plant species richness in an experimental grassland community. *Oecologia*. 2019; 191(1): 177–190. <https://doi.org/10.1007/s00442-019-04476-z>.