Nutritional value of cactus pear grown under different levels of nitrogen and phosphorus and two harvest frequencies

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1. Introduction

Cactus pear is a plant tolerant to water deficit and, therefore, very exploited in arid and semi-arid regions worldwide. The different species of cactus pear have high contents of water (866 g kg⁻¹ dry matter [DM]), minerals (168 g kg⁻¹ DM), and carbohydrates (751 g kg⁻¹ DM), with greater participation of non-fiber carbohydrates (NFC; approximately 479 g kg⁻¹ DM) and low crude protein (CP) content (62 g kg⁻¹ DM) and neutral detergent fiber (NDF; 278 g kg⁻¹ DM) (Batista et al., 2003). The low CP and NDF contents limit the inclusion of cactus pear in ruminant feed, and the inclusion of up to 50% DM of the diet avoids disturbances such as diarrhea (Gebremariam et al., 2006). However, the nutritional value of cactus pear varies according to the management conditions adopted (Dubeux Jr. et al., 2006).
Fertilization, for example, has a great influence on the quality of cactus pear. Nitrogen fertilization usually increases its CP content, and an increase of 100% or greater can be observed (Dubeux Jr. et al., 2006). Phosphate (P₂O₅) fertilization acts synergistically with N fertilization, potentiating N uptake by roots and protein synthesis in the aerial part of the plant (Prado, 2008). The effect of N×P interaction on the quality of cactus pear is still poorly explored. Similarly, harvest management may also influence cactus pear quality, although this effect has also been little studied. Higher cutoff frequency may result in the harvesting of a more tender material, with a higher crude and mineral protein content and lower DM, fiber, and organic matter contents (Pinos-Rodríguez et al., 2010).

The understanding of how cactus pear quality is affected by the interaction between N/P₂O₅ fertilization and harvest frequency can help in the establishment of management that contributes to a greater participation of cactus pear in ruminant diet or that allows greater use in the diet of high-production animals; our hypothesis is that the interaction between N and P fertilization and harvest management affects the nutritional quality of cactus pear. In this context, this study evaluated the effects of N/P fertilization and harvest frequency on the chemical, mineral composition, and in vitro degradation kinetics of cactus pear cv. Gigante.

2. Material and Methods

Two experiments were conducted from January 2011 to March 2013 in Quixadá and Tejuçuoca, Ceará, Brazil. Quixadá is located at 190 m altitude, at the geographical coordinates: 4°59’ S latitude and 39°01’ W longitude, with a BSw’h’ hot semi-arid climate, according to Koppen classification (1948). Tejuçuoca is located at 140 m altitude, at the geographical coordinates: 3°59’ S latitude and 39°34’ W longitude, with Aw climate, tropical with dry season, according to Koppen classification (1948).

The average rainfall, temperature and relative humidity verified in 2011 and 2012 were 1,042 and 602 mm, 27 and 26.9 °C, and 61.3 and 56.3% in Quixadá and 1,038 and 561 mm, 26 and 26.8 °C, and 67.2 and 63.8% in Tejuçuoca, respectively. Data were obtained at the Agroclimatological Station of the Universidade Federal do Ceará (Quixadá) and at the Agroclimatological Station of FUNCEME (Tejuçuoca).

The chemical characteristics of the soils at the 0.0-20.0 cm layer were: pH, 6.1 and 6.2; P, 5 and 6 mg dm⁻³; K, 260 and 243 mg dm⁻³; Ca + Mg, 6.80 and 7.20 cmol c dm⁻³; Ca, 3.40 and 4.00 cmol c dm⁻³; Mg, 3.40 and 3.20 cmol c dm⁻³; Al, absent; Na, 20 and 7 mg dm⁻³; organic matter (OM), 5.28 and 8.17 g kg⁻¹ for Quixadá and Tejuçuoca, respectively. The soil was classified as sandy and sandy-loam in Quixadá and Tejuçuoca, respectively.

Planting was performed before the beginning of the rainy season, and cladodes were buried 30 cm deep in the furrow in vertical position, with the cut part facing the ground surface, covering ⅔ and with no irrigation. The spacing was 2.0×0.10 m, totaling 50,000 plants ha⁻¹. The experimental design was performed in a completely randomized block design with four replications. The treatments consisted of nine combinations of N (urea, 45% N)/P (single superphosphate, 18% P₂O₅) assigned to the plots, and two harvest frequencies (annual and biannual) assigned to the subplots, totaling 72 experimental units in each site/municipality. The combinations of N/P₂O₅ were established according to the matrix plan puebla II, from the reference level of 100 kg ha⁻¹ year⁻¹ for both N and P₂O₅. By means of the matrix, N/P₂O₅ combinations were established, with reductions and increases of 90 and 30% from the reference levels, that is, 10, 70, 100, 130, and 190 kg ha⁻¹ year⁻¹. Each experimental unit had three rows of cactus pear, 4.0 m long and 6.0 m wide, totaling 24 m² and 120 plants. Fertilization was performed in the rainy season, with superphosphate available in only one application and urea in three applications, with 20 day-intervals. Urea was diluted in water using plastic bottles with the capacity to compose 1 L of solution, evenly distributed in the 4.0 m row, totaling 3.0 L per plot. The source of micronutrients was FTE-BR 12 (0.1% Mo, 0.8% Cu, 1.8% B, 2.0% Mn, 3.0% Fe, and 9.0% Zn) applied at 50 kg ha⁻¹ year⁻¹ and Ca and S balance was performed using agricultural gypsum and calcitic limestone, based on the
maximum level of single superphosphate. All nutrients except N were available in only one application, along with single superphosphate.

Samples of cactus pear were harvested preserving the primary cladodes, then minced, placed in identified bags, and frozen. Afterwards, samples were thawed and dried in a forced-ventilation oven at 55 °C to constant weight. Dry matter (method ID 930.15) and total nitrogen (TN, method ID 984.13) contents were determined according to the procedures described in AOAC (1990). The CP content was obtained by multiplying the TN content by 6.25. The OM content was calculated by subtracting the ash content (method ID 942.05) from 100. The NDF content was determined using the method described by Van Soest and Robertson (1985). Analyses of NDF were not treated with amylase and neither corrected for ash and protein. The total carbohydrates content was estimated as proposed by Sniffen et al. (1992). The P content was determined by spectrophotometry; S by tubidimetry; K by flame photometry; contents of Ca, Mg, Fe, Mn, and Cu, by atomic absorption spectrophotometry (AOAC, 1990).

The in vitro DM degradation kinetic parameters of cactus pear were estimated by the cumulative gas production technique, as described by Pell and Schofield (1993), with adaptations described below. Incubations were performed in glass flasks with a capacity of 150 mL. Each flask was added with 500 mg substrate and 40 mL McDougall buffer solution (McDougall, 1948) previously reduced with CO₂ flow, to adjust the pH to the 6.9-7.0 range. Subsequently, incubation flasks were added with 10 mL inoculum from a dairy cow with approximately 600 kg body weight, fistulated in the rumen, and fed a diet based on grass hay (60% DM) and concentrate (40% DM; ground corn and soybean meal). The diet provided was divided into two equal meals. The inoculum was collected by hand before the first meal, from the ruminal dorsal sac, through the fistula, filtered through a double layer of gauze and, subsequently, poured in a thermost bottle previously heated with water at 39 °C and immediately transported to the laboratory. The addition of buffer solution and inoculum to the glass flasks was carried out under CO₂ spray to maintain anaerobic conditions. Immediately afterwards, bottles were sealed with a rubber stopper and sealed with an aluminum seal and kept in a temperature-controlled room at 39 °C in a water bath. Samples were incubated in duplicate. From this moment on, the volume of the gases produced by fermentation of the substrate and accumulated in the vials was measured using a graduated syringe at times of 1, 2, 3, 6, 12, 15, 18, 22, 26, 30, 34, 40, 48, 60, 72, 96, and 120 h after incubation. After each reading, the flask was manually shaken to homogenize the mixture. Flasks containing only ruminal fluid and buffer solution were included as blanks to measure the total gas production from the inoculum. This value was subtracted from the total volume to obtain the net gas production (production of gas from the substrate).

The in vitro degradation kinetic parameters were estimated from cumulative gas production using the NLIN procedure of SAS software (Statistical Analysis System, version 9.0), according to the model described in France et al. (2000):

\[ G = A[1 - \exp(-c(t-L))], \]

in which \( G \) (mL) is the cumulative volume of gases at time \( t \), \( A \) (mL) is the maximum gas production, \( c \) (% h⁻¹) is the gas accumulation rate, and \( L \) (h) is the time of microbial colonization.

The effect of fertilization was determined by simple or multiple linear regression, adopting as model selection criterion the significance of the regression coefficient by the F-test, at the level of 0.1, 1, 5, and 10% probability. Levels of N and P were considered as independent variables. Regression equations were associated with two harvest frequencies (annual and biannual) and to two sites (Quixadá and Tejuçuoca) and used to estimate the total DM (TDM), CP, NDF, OM, TC, P, S, K, Ca, Mg, Fe, Mn, and Cu, as well as the fermentation kinetic parameters: \( A \), \( c \), and \( L \). For these analyses, the System for Statistical and Genetic Analysis (SAEG 9.1, 2007) was used. In the case of the effect of harvest frequencies on these variables, the means were compared by Tukey’s test, at a 5% probability level, using the procedure GLM of SAS.
3. Results

Most of the nutrients were influenced by fertilization, except for OM, in Quixadá under biannual harvest and in Tejuçuoca under annual harvest, and CP, in Tejuçuoca under annual harvest, where there was no fit to N and P levels (Table 1).

The TDM content was linearly reduced with P levels in Tejuçuoca under biannual harvest. In Quixadá, under annual harvest, there was a linear increase in the CP content with P levels. The maximum CP content was observed under biannual harvest in both sites with the combined N/P\textsubscript{2}O\textsubscript{5} doses of 190/190 for Quixadá and 130.03/190 kg ha\textsuperscript{-1} year\textsuperscript{-1} for Tejuçuoca. Fertilization did not result in a decrease in NDF content under biannual harvest in Quixadá and in both harvests in Tejuçuoca (Table 1).

Macrominerals of cactus pear cv. Gigante were influenced by fertilization, except for Mg and S in Tejuçuoca under annual harvest (Table 2). The P levels resulted in a quadratic effect for Ca content in Quixadá under biannual harvest and in Tejuçuoca under annual harvest, and K content in Tejuçuoca under annual harvest. The Ca content under annual harvest in Quixadá and in both harvests in Tejuçuoca was reduced in response to fertilization. The K content in Quixadá under biannual harvest was linearly reduced with the P levels. The S content increased with the N/P\textsubscript{2}O\textsubscript{5} levels. The lowest S content was obtained with the N/P\textsubscript{2}O\textsubscript{5} combination of 10/10 kg ha\textsuperscript{-1} year\textsuperscript{-1} in both harvest frequencies in Quixadá.

The Cu and Mn contents in Tejuçuoca under annual harvest increased linearly with the N levels, while Fe content in Quixadá under biannual harvest increased linearly in response to P levels (Table 3).

**Table 1 - Chemical composition in response to combinations of N and P levels in cactus pear cv. Gigante in Quixadá and Tejuçuoca**

| HF          | Nutrient | Regression equation                                                                 | R\textsuperscript{2} |
|-------------|----------|-------------------------------------------------------------------------------------|---------------------|
|             | TDM      | Y = 203.505 − 0.8113\*P + 0.00335\*P\textsuperscript{2}                              | 0.49                |
| Annual      | OM       | Y = 878.272 + 0.2263\*N + 0.001820\*N\textsuperscript{2} + 0.07256\*P + 0.001926\*P\textsuperscript{2} − 0.004816\*NP | 0.92                |
|             | CP       | Y = 59.7605 + 0.07724\*P                                                       | 0.29                |
|             | TC       | Y = 837.610 − 0.2345\*P                                                       | 0.84                |
|             | NDF      | Y = 343.2530 − 1.1946\*N + 0.004596\*N\textsuperscript{2}                       | 0.44                |
| Biannual    | TDM      | Y = 143.1690 + 0.4898N + 0.01157\*N\textsuperscript{2} + 0.4611P + 0.01135\*P + 0.02923\*NP | 0.98                |
|             | OM       | Y = 921.29                                                                     | -                   |
|             | CP       | Y = 44.0211 + 0.005999N − 0.001548\*N\textsuperscript{2} − 0.006810P − 0.001397\*P\textsuperscript{2} + 0.003464\*NP | 0.83                |
|             | TC       | Y = 859.707 − 0.1111\*P                                                        | 0.37                |
|             | NDF      | Y = 320.715 + 0.3890\*N − 1.2295\*P + 0.005777\*P\textsuperscript{2}            | 0.66                |
|             |          |                                                                                   |                     |
|             | TDM      | Y = 105.780 − 0.2602\*P + 0.0009695\*P\textsuperscript{2}                       | 0.89                |
| Annual      | OM       | Y = 886.37                                                                     | -                   |
|             | CP       | Y = 93.80                                                                      | -                   |
|             | TC       | Y = 788.735 − 0.2018\*P                                                       | 0.33                |
|             | NDF      | Y = 111.988 + 1.5019\*N + 1.4269\*P − 0.0117\*NP                              | 0.89                |
| Biannual    | TDM      | Y = 91.6905 − 0.06726\*P                                                      | 0.57                |
|             | OM       | Y = 887.350 + 0.1196\*N − 0.1232\*P                                          | 0.33                |
|             | CP       | Y = 59.1321 + 0.4005\*N − 0.001540\*N\textsuperscript{2} + 0.1063\*P         | 0.59                |
|             | TC       | Y = 795.809 − 0.2166\*P                                                        | 0.35                |
|             | NDF      | Y = 331.365 + 0.6441\*N − 0.004778\*N\textsuperscript{2} + 0.6882P − 0.004410\*P\textsuperscript{2} | 0.43                |

HF - harvest frequency; TDM - total dry matter (g kg\textsuperscript{-1} natural matter); OM - organic matter (g kg\textsuperscript{-1} DM); CP - crude protein (g kg\textsuperscript{-1} DM); TC - total carbohydrates (g kg\textsuperscript{-1} DM); NDF - neutral detergent fiber (g kg\textsuperscript{-1} DM); R\textsuperscript{2} - coefficient of determination.

***, ***, **, *, and  are significant effect at 0.1, 1, 5, and 10% probability by F test, respectively.
### Table 2 - Macrominerals in response to combinations of N and P levels in cactus pear cv. Gigante in Quixadá and Tejuçuoca

| HF       | Macromineral (g kg⁻¹ DM) | Regression equation                                                                 | R²  |
|----------|---------------------------|--------------------------------------------------------------------------------------|-----|
|         |                           | Quixadá                                                                              |     |
| Annual   | P                         | Y = 1.3278 - 0.002485*N - 0.007162*P + 0.00002819*P²                                 | 0.60|
|          | K                         | Y = 51.5797 - 0.2276*N - 0.0008021*N² - 0.1152*P - 0.001262**P² + 0.003428**NP    | 0.97|
|          | Ca                        | Y = 25.3493 - 0.01701*N                                                           | 0.46|
|          | Mg                        | Y = 9.4276 - 0.007529*P                                                            | 0.32|
|          | S                         | Y = 1.1565 + 0.07069***N - 0.0002366**N² + 0.04335*P - 0.0002234*P²              | 0.83|
| Biannual | P                         | Y = 0.6711 - 0.00087*N - 0.00002908*N² - 0.001956*P - 0.00002082*P² + 0.00006577**NP | 0.44|
|          | K                         | Y = 20.2429 + 0.02501*N - 0.0001276*N²                                            | 0.24|
|          | Ca                        | Y = 21.4868 + 0.03802*P - 0.0002449*P²                                         | 0.24|
|          | Mg                        | Y = 6.0266 + 0.01706*N - 0.0001056**N² + 0.01369*P - 0.00005654*P²              | 0.88|
|          | S                         | Y = 2.8499 + 0.07490**N - 0.0001917*N² + 0.09599***P - 0.0004798***P²           | 0.69|
|         |                           | Tejuçuoca                                                                            |     |
| Annual   | P                         | Y = 0.4000 + 0.004593*N - 0.00001763*N² + 0.005024**P - 0.0003228***P²            | 0.72|
|          | K                         | Y = 23.1727 + 0.04526P - 0.0003402*P²                                             | 0.74|
|          | Ca                        | Y = 35.0091 - 0.1261***P + 0.0003625*P²                                           | 0.87|
|          | Mg                        | Y = 8.68                                                                           | -   |
|          | S                         | Y = 6.86                                                                           | -   |
| Biannual | P                         | Y = 1.3383 - 0.01069***N + 0.00006287***N² - 0.008930***P + 0.00004773***P²      | 0.76|
|          | K                         | Y = 18.7499 + 0.01447*N + 0.0006780*N² + 0.04707*P - 0.0007327**P² - 0.001712*NP | 0.54|
|          | Ca                        | Y = 22.9494 - 0.04529***N + 0.04351*P + 0.0003539***P²                           | 0.92|
|          | Mg                        | Y = 12.1238 - 0.04158*N + 0.0001518*N² + 0.02180**P                             | 0.60|
|          | S                         | Y = 15.1768 - 0.04410*N + 0.000493**N² + 0.008297*P + 0.0004220*P² - 0.0007496*NP | 0.63|

HF - harvest frequency; R² - coefficient of determination.

***, **, *, and 込むindicate significant effect at 0.1, 1, 5, and 10% probability by F test, respectively.

### Table 3 - Microminerals in response to combinations of N and P levels in cactus pear cv. Gigante in Quixadá and Tejuçuoca

| HF       | Micromineral (mg kg⁻¹ DM) | Regression equation                                                                 | R²  |
|----------|---------------------------|--------------------------------------------------------------------------------------|-----|
|         |                           | Quixadá                                                                              |     |
| Annual   | Cu                        | Y = 0.04982 - 0.0002794**N + 0.000001171*N²                                       | 0.67|
|          | Fe                        | Y = 64.4255 - 0.1580*N + 0.3649*P - 0.001551*P²                                   | 0.22|
|          | Mn                        | Y = 140.095 - 0.4714N + 0.01088**N + 2.4986**P - 0.02129**NP                     | 0.67|
| Biannual | Cu                        | Y = 0.03189 - 0.0002426*P + 0.0000009935*P²                                       | 0.53|
|          | Fe                        | Y = 62.7841 + 0.07884*P                                                          | 0.13|
|          | Mn                        | Y = 118.124 + 1.2556**N - 0.005051**N² + 0.4225P - 0.003057**P²                   | 0.57|
|         |                           | Tejuçuoca                                                                            |     |
| Annual   | Cu                        | Y = 0.02507 + 0.0001074**N                                                      | 0.34|
|          | Fe                        | Y = 165.287 - 0.5845**P + 0.004042**P²                                          | 0.35|
|          | Mn                        | Y = 307.643 + 0.3825*N                                                          | 0.13|
| Biannual | Cu                        | Y = 0.014 + 4.76·10⁻⁴*N + 2.02·10⁻⁴*N² + 2.01·10⁻⁴*P + 3.56·10⁻⁴*P² - 8.43·10⁻⁴*NP | 0.80|
|          | Fe                        | Y = 43.9319 + 0.09494N + 0.060634**N - 0.5232*P + 0.006638**P² - 0.01696**NP       | 0.59|
|          | Mn                        | Y = 453.320 - 3.2112**N + 0.01869**N² + 0.7288*P                                | 0.65|

HF - harvest frequency; R² - coefficient of determination.

***, **, *, and 込むindicate significant effect at 0.1, 1, 5, and 10% probability by F test, respectively.
There was a quadratic effect of P levels on Cu contents in Quixadá under biannual harvest and Fe in Tejuçuoca under annual harvest. The quadratic effect of N levels was found for Cu content in Quixadá under annual harvest. The other microminerals, in the different sites and harvest frequencies, were fitted to the multiple regression model.

The *in vitro* degradation kinetic parameters of cactus pear cv. Gigante were influenced by fertilization, except the latency period in Quixadá under biannual harvest (Table 4). Most *in vitro* degradation kinetic parameters were fitted to the multiple regression model. The maximum total gas production was verified in the combined N/P levels of 10/190 and 190/92.09 kg ha\(^{-1}\) year\(^{-1}\) for Quixadá and Tejuçuoca, both under biannual harvest, respectively (Table 4). The maximum digestion rate was obtained in the combined N/P levels of 190/190, 14.19/10, and 35.89/10 kg ha\(^{-1}\) year\(^{-1}\) for Quixadá under annual and biannual harvest and Tejuçuoca under biannual harvest, respectively (Table 4).

The biannual harvest resulted in higher TDM, OM, NDF, and TC content and lower CP content in Quixadá (Table 5). In Tejuçuoca, under biannual harvest, there was lower TDM content and higher NDF content (Table 5).

The P and K content decreased under biannual harvest in Quixadá (Table 6). In Tejuçuoca, there was similarity between annual and biannual harvests for the P and K contents and the lowest Ca contents under biannual harvest. The Mg content increased in Tejuçuoca and decreased in Quixadá under biannual harvest. The S content of cactus pear was higher under biannual harvest in both sites (Table 6).

The harvest frequency did not influence the Fe content in Quixadá; on the other hand, under biannual harvest, the Cu and Mn contents decreased. In Tejuçuoca, the Cu and Mn contents were reduced under annual harvest, while the Fe content increased (Table 7).

The biannual harvest reduced the total gas production and degradation rate in Quixadá (Table 8); however, there was no effect of harvest frequency on the latency period. In Tejuçuoca, the digestion rate and latency period were higher under biannual harvest, and total gas production did not vary according to the harvest frequency (Table 8).

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### Table 4 - Kinetic parameters in response to combination of N and P levels in cactus pear cv. Gigante in Quixadá and Tejuçuoca

| HF     | Parameter | Regression equation | \(R^2\) |
|--------|-----------|---------------------|--------|
| **Quixadá** |           |                     |        |
| Annual | A         | \(Y = 217.6190 \pm 0.2991\*N\) | 0.90   |
|        | c         | \(Y = 0.05052 + 0.0001412\*N + 0.00006060\*P\) | 0.89   |
|        | L         | \(Y = 2.3884 + 0.01270\*N + 0.008298\*P + 0.0002373\*N^2 - 0.0004220\*NP\) | 0.88   |
| Biannual | A        | \(Y = 98.3020 - 0.02795\*N + 0.002425\*N^2 + 0.4312\*P - 0.003421\*NP\) | 0.73   |
|         | c        | \(Y = 0.07490 + 3.420·10^{-5}\*N - 4.184·10^{-6}\*N^2 - 3.318·10^{-10}\*P - 2.417·10^{-10}\*P^2 + 8.594·10^{-14}\*NP\) | 0.85   |
|         | L        | \(Y = 3.48\) | -      |
| **Tejuçuoca** |           |                     |        |
| Annual | A         | \(Y = 112.2980 + 0.0750\*N\) | 0.32   |
|        | c         | \(Y = 0.1174 + 0.0001306\*N\) | 0.53   |
|        | L         | \(Y = 2.9120 + 0.005058\*N + 0.003344\*P\) | 0.85   |
| Biannual | A       | \(Y = 100.7570 + 0.06532\*N + 0.3433\*P - 0.001864\*P^2\) | 0.72   |
|         | c        | \(Y = 0.1612 + 0.0001720\*N - 0.000003107\*N^2 - 0.00005732\*P + 0.000005104\*NP\) | 0.42   |
|         | L        | \(Y = 3.5777 + 0.01398\*N - 0.00004758\*N^2\) | 0.86   |

HF - harvest frequency; A - total gas production (mL g\(^{-1}\) DM); c - digestion rate (% h\(^{-1}\)); L - latency period (h); \(R^2\) - coefficient of determination. ***,**, *, and \(\ddagger\) indicate significant effect at 0.1, 1, 5, and 10% probability by F test, respectively.
### Table 5 - Chemical composition of cactus pear cv. Gigante according to harvest frequency (annual and biannual)

| Nutrient (g kg⁻¹ DM) | Harvest | SEM | P-value |
|----------------------|---------|-----|---------|
|                      | Annual  | Biannual |       |
| TDM¹                 | 163.70b | 193.87a | 0.60   | 0.0259 |
| OM                   | 900.80b | 921.29a | 0.28   | <0.0001|
| CP                   | 67.48a  | 46.17b  | 0.29   | <0.0001|
| TC                   | 810.55b | 849.10a | 0.51   | <0.0001|
| NDF                  | 279.87b | 307.04a | 0.77   | 0.0464 |

**Tejuçuoca**

| TDM¹                 | 92.17a  | 84.97b  | 0.14   | 0.0013 |
| OM                   | 886.37  | 886.99  | 0.22   | 0.0378 |
| CP                   | 93.80   | 90.72   | 0.25   | 0.5548 |
| TC                   | 769.12  | 774.13  | 0.41   | 0.5314 |
| NDF                  | 275.08b | 350.66a | 1.26   | <0.0001|

**DM** - total dry matter; **OM** - organic matter; **CP** - crude protein; **TC** - total carbohydrates; **NDF** - neutral detergent fiber; **SEM** - standard error of the mean.

¹ g kg⁻¹ natural matter.

Means followed by different lowercase letters in the same row are significantly different (P<0.05) by Tukey’s test.

### Table 6 - Macronutrients of cactus pear cv. Gigante according to harvest frequency (annual and biannual)

| Macronutrient (g kg⁻¹ DM) | Harvest | SEM | P-value |
|---------------------------|---------|-----|---------|
|                           | Annual  | Biannual |       |
| Tejuçuoca                 |         |         |        |
| P                         | 0.71a   | 0.51b   | 0.05   | 0.0009 |
| K                         | 30.09a  | 21.16b  | 1.29   | <0.0001|
| Ca                        | 23.65   | 22.24   | 0.42   | 0.0542 |
| Mg                        | 8.68a   | 7.10b   | 0.23   | <0.0001|
| S                         | 6.86b   | 11.61a  | 0.79   | <0.0001|

**Quixadá**

| P                         | 0.74    | 0.74    | 0.05   | 0.9899 |
| K                         | 23.46   | 22.66   | 0.47   | 0.3232 |
| Ca                        | 26.89a  | 18.38b  | 1.30   | <0.0001|
| Mg                        | 9.86b   | 11.83a  | 0.36   | <0.0001|
| S                         | 12.96b  | 14.48a  | 0.44   | 0.0319 |

**DM** - dry matter; **SEM** - standard error of the mean.

Means followed by different lowercase letters in the same row are significantly different (P<0.05) by Tukey’s test.

### Table 7 - Micronutrients of cactus pear cv. Gigante according to harvest frequency (annual and biannual)

| Micronutrient (mg kg⁻¹ DM) | Harvest | SEM | P-value |
|----------------------------|---------|-----|---------|
|                            | Annual  | Biannual |       |
| Tejuçuoca                  |         |         |        |
| Cu                         | 0.036a  | 0.020b  | 0.002  | <0.0001|
| Fe                         | 65.80   | 70.83   | 3.55   | 0.1110 |
| Mn                         | 238.68a | 183.11b | 11.05  | 0.0002 |

**Quixadá**

| Cu                         | 0.036b  | 0.056a  | 0.003  | <0.0001|
| Fe                         | 129.96a | 77.58b  | 8.23   | <0.0001|
| Mn                         | 347.17b | 436.49a | 21.17  | 0.0015 |

**DM** - dry matter; **SEM** - standard error of the mean.

Means followed by different lowercase letters in the same row are significantly different (P<0.05) by Tukey’s test.
4. Discussion

In general, the increase in the N/P\textsubscript{2}O\textsubscript{5} doses resulted in a reduced TDM content from cactus pear. The use of fertilizers in the cultivation of cactus pear favors the emergence of young cladodes (Silva et al., 2013), richer in water than the mature ones. On the other hand, the CP content of cactus pear increased with the N/P\textsubscript{2}O\textsubscript{5} doses, probably due to the synergistic effect between N and P\textsubscript{2}O\textsubscript{5}, which favors the absorption of both, via symport, in which two ions of opposite charges (NH\textsubscript{4}\textsuperscript{+} and H\textsubscript{2}PO\textsubscript{4}\textsuperscript{−3}) are absorbed together (Prado, 2008; Silva et al., 2013). Although there was a positive effect of N/P\textsubscript{2}O\textsubscript{5} levels, the CP content in Quixadá under biannual harvest was below 70 g kg\textsuperscript{−1} DM, minimum content required for satisfactory growth of ruminal microorganisms (Van Soest, 1994). The sandy soil, with lower moisture retention, may have contributed to reduced root-nutrient contact and reduced N availability in the soil solution in Quixadá, resulting in low CP content.

The effect of the N/P\textsubscript{2}O\textsubscript{5} doses on the NDF, TC, and OM contents of cactus pear reflects the alteration in growth dynamics in response to fertilization. The best growth condition of cactus pear with fertilization can result in the maturation of basal cladodes (close to the soil) and the emergence of young cladodes at the top of the plant (Dubeux Jr. et al., 2006; Silva et al., 2013). Pinos-Rodríguez et al. (2010) observed a reduction in CP content and digestibility and an increase in NDF and ADF according to cladode maturity (30, 37, 45, 60, 75, and 90 days of age). The TC content was reduced with fertilization, possibly due to the increase in CP content in this condition, in which soluble carbohydrates are used as carbon skeleton for protein synthesis (Peyraud and Astigarraga, 1998).

The presence of N-NH\textsubscript{4}\textsuperscript{+} in the soil can inhibit Ca uptake, since Ca is absorbed as Ca\textsuperscript{2+} (Silva et al., 2012), which justifies the reduction in the Ca content in cactus pear in response to N doses. The reduction in Ca content resulted in a decrease in the Ca:P ratio to 9.72:1.00 with the N/P\textsubscript{2}O\textsubscript{5} combination at 190/10 kg ha\textsuperscript{−1} year\textsuperscript{−1} in the biannual harvest in Tejuçuoca, which may increase P absorption in the gastrointestinal tract of ruminants (Wu et al., 2000). On the other hand, the increase in Ca content in cactus pear in response to the P\textsubscript{2}O\textsubscript{5} doses may have been due to the effect of this nutrient (P\textsubscript{2}O\textsubscript{5}) on the increase in root biomass (Bonfim-Silva et al., 2014), which improves nutrient uptake and accumulation, or the concentration effect related to plant growth (Teles et al., 2004), depending on the location and frequency of harvest in the present study. The increase in P\textsubscript{2}O\textsubscript{5} doses favors the conversion of the nutrient into an assimilable form, resulting in greater uptake and, consequently, greater P accumulation in the aerial part of cactus pear. The magnitude of this effect is associated with soil type, plant age, soil moisture content, harvesting time, etc (Teles et al., 2004; Silva et al., 2012).

The K content of cactus pear may vary according to fertilization due to competitive inhibition by the same absorption site between NH\textsubscript{4}\textsuperscript{+} and K\textsuperscript{+} (Silva et al., 2012). The Mg content of cactus pear may increase in response to N/P\textsubscript{2}O\textsubscript{5} doses. In the plant, Mg is a component of the RUBISCO enzyme (ribulose 1,5 bisphosphate carboxylase-oxygenase) (Galizzi et al., 2004), the main source of protein in plants.

### Table 8 - Kinetic parameters of cactus pear cv. Gigante according to harvest frequency (annual and biannual)

| Parameter | Harvest | SEM | P-value |
|-----------|---------|-----|---------|
|           | Annual  | Biannual |       |
| Quixadá   |         |         |       |
| A         | 186.48a | 129.38b | 7.55   | <0.0001 |
| c         | 0.072a  | 0.059b  | 0.003  | <0.0001 |
| L         | 3.60    | 3.48    | 0.16   | 0.555   |
|           |         |         |       |
| Tejuçuoca |         |         |       |
| A         | 119.22  | 119.18  | 1.69   | 0.980   |
| c         | 0.120b  | 0.132a  | 0.003  | 0.005   |
| L         | 3.32b   | 4.06a   | 0.15   | <0.0001 |

DM - dry matter; A - total gas production (mL g\textsuperscript{−1} DM); c - digestion rate (% h\textsuperscript{−1}); L - latency period (h); SEM - standard error of the mean.

Means followed by different lowercase letters in the same row are significantly different (P<0.05) by Tukey's test.
and favors the transport of P, especially in phosphorylated reactions (Oliveira et al., 2001). However, this effect on Mg content of cactus pear was not observed in the present study under annual harvest in both sites, probably due to the lesser development of the plant in response to the greater stress caused by the higher harvest frequency. By applying single superphosphate as a source of P, S was indirectly added, increasing its levels in the soil and, consequently, in the tissues of cladodes, which explains the increase in the S content with the N/P$_2$O$_5$ levels. The lowest S content obtained under annual and biannual harvesting in Quixadá may reduce the use of non-protein nitrogen for microbial protein synthesis, if there is no supplementation with this mineral (Van Soest, 1994).

The reduction in Cu content with fertilization in Quixadá in both harvests may have been due to the competitive inhibition between Cu$^{+2}$ and the absorbable forms of N (NH$_4^+$) and P (H$_2$PO$_4^-$) (Prado, 2008). The Cu content increased with fertilization in Tejuçuoca, which may be related to better growth conditions, which increases the proportion of photosynthetically active tissues, since Cu is present in chloroplasts (Prado, 2008). The increase in Fe content with P levels may be related to the transport via symport between P and Fe (Prado, 2008). Increasing Fe content may increase the requirement for Cu by ruminants (Phillippo et al., 1987). The Mn content varied with N and P levels, probably in response to variation in N content in cladode tissues (Galizzi et al., 2004).

The different management conditions in the present study resulted in different growth conditions and, therefore, appearance of young cladodes (Silva et al., 2013) and maturation of basal cladodes, with higher and lower digestibility, respectively. In Tejuçuoca, there was a higher degradation rate when compared with Quixadá. The higher CP content in Tejuçuoca may have improved the digestion of structural carbohydrates due to higher N supply to ruminal microorganisms (Van Soest, 1994). The increase of NDF content with the N/P$_2$O$_5$ doses may have resulted in longer colonization period (Mertens, 1997). In this case, low lignin content and high NFC content of cactus pear (Batista et al., 2003) may have compensated for the increased colonization period and have not compromised the ruminal degradation with N/P$_2$O$_5$ levels.

The maturation process (Van Soest, 1994; Titgemeyer et al., 1996; Wilson and Kennedy, 1996) of cladodes may increase OM, NDF, and TC contents and lower CP content in cactus pear (Pinos-Rodríguez et al., 2010). On the other hand, the better growth conditions and, consequently, budding of young cladodes (Silva et al., 2013), may reduce TDM content, as observed in Tejuçuoca. This could have also caused the dilution effect, justifying the lack of harvest frequency effect on OM, CP, and TC contents, but this was not enough to avoid the greater accumulation of NDF under biannual harvest in Tejuçuoca.

Phosphorus and potassium have high mobility in the plant, which reduces the concentration in the aerial part at a more advanced age (Teles et al., 2004), as observed in Quixadá under biannual harvest. In Tejuçuoca, there was a dilution effect, due to the better growth conditions, explaining the lack of effect of the harvest frequency on P and K contents in cactus pear.

Calcium and magnesium have lower mobility in the plant, which may result in higher concentration at more advanced stages (Silva et al., 2012); however, reduction may also occur due to the dilution effect, because of the emergence of new cladodes or of the delay in the maturation process of cladodes. It is worth mentioning that a large part of the Ca present in cactus pear is in the form of calcium oxalate, unavailable to production animals, making it necessary to supplement with Ca when supplying cactus pear, due to the high concentration of Ca in the aerial part of cactus pear (Dubeux Jr et al., 2021). In turn, the presence of S in the single superphosphate and the longer absorption time contributed to increase the S content under biannual harvest in both sites.

The Mn content of cactus pear was directly correlated with N content in the aerial part of the plant (Galizzi et al., 2004). The higher Mn content indicates higher absorption, which may have reduced the availability of Cu and Fe to the plants (Prado, 2008).

As observed in the evaluation of N/P$_2$O$_5$ doses in both sites, the effect of harvest frequency on ruminal kinetics parameters of cactus pear is also a reflection of growth dynamics, which alters the chemical composition of the plant. More mature cladodes, for example, produce a smaller gas volume in vitro, due
to the higher fraction of indigestible carbohydrate and lower fraction of rapid digestion carbohydrate; the opposite is observed in young cladodes (Dubeux Jr. et al., 2021). In the present study, the increase in digestion rate in Tejuçuoca under biannual harvest was associated with reduction of TDM content and increase in NDF content, demonstrating that this effect was possibly a result from the alteration in the composition of NFC. The latency period in Tejuçuoca was longer under biannual harvest, possibly due to the higher NDF content (Mertens, 1997). Nevertheless, the increase in NDF content was not sufficient to reduce the digestion rate of cactus pear. In this context, the low lignin content and the high NFC content may have compensated for the increase in NDF content (Batista et al., 2003). The reduction in CP content, 34% lower than the minimum required for normal rumen function (Van Soest, 1994), under biannual harvest in Quixadá, may have limited microbial growth and, consequently, reduced ruminal degradation. In Tejuçuoca, the lack of effect of harvest frequency on total gas production was related to the similarity of CP content between the annual and biannual harvest frequencies.

5. Conclusions

Nitrogen and phosphorus levels and harvest frequency influence the nutritional value of cactus pear cv. Gigante. The best nutritional value is achieved with the combined nitrogen/phosphorus levels of 190/190 and 130.03/190 kg ha$^{-1}$ year$^{-1}$ under biannual harvest in Quixadá and Tejuçuoca, respectively. The best Ca:P ratio is obtained at the N/P.O combination of 190/10 kg ha$^{-1}$ year$^{-1}$ under biannual harvest in Tejuçuoca. The annual harvest improves the nutritional value of cactus pear in Quixadá; however, in Tejuçuoca, the harvest frequency has weaker influence on the nutritional value of cactus pear.

Conflict of Interest

The authors declare no conflict of interest.

Author Contributions

Data curation: G.M.F. Gomes and M.N. Lopes. Formal analysis: G.M.F. Gomes, M.J.D. Cândido, D.B. Galvani and I. Soares. Investigation: G.M.F. Gomes, M.J.D. Cândido and M.N. Lopes. Methodology: G.M.F. Gomes, M.J.D. Cândido, M.N. Lopes, D.B. Galvani and I. Soares. Project administration: G.M.F. Gomes and M.J.D. Cândido. Supervision: M.J.D. Cândido. Visualization: G.M.F. Gomes and M.J.D. Cândido. Writing-original draft: G.M.F. Gomes and M.J.D. Cândido. Writing-review & editing: G.M.F. Gomes and M.J.D. Cândido.

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