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Productive potential and quality of the chipile *Crotalaria longirostrata* Hook. & Arn. as forage for small ruminants

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This study aimed to evaluate the biomass yield and crude protein content of *C. longirostrata* under different cutting frequencies and intensities, to assess its potential as forage for small ruminants on the coast of Oaxaca, Mexico. Twelve treatments were carried out with three replications, which included three cutting frequencies (35, 70, and 105 days) and four cutting intensities (5, 10, 15, and 20 cm), distributed in randomized complete blocks during the rainy and dry seasons. Variables such as dry matter (DM) yield of total biomass (TB), leaves, stems, and crude protein (CP) in leaves and stems were measured. The highest TB productions were recorded at the frequencies of 70 and 105 days in both periods, and the lowest at the frequency of 35 days; the highest leaf yields occurred at 70 days in rain and at 35 days in dry conditions; for stems, the highest value was observed at 105 days in rain and at 70 days in dry season. The CP was higher in the dry season (37%) in leaves compared to the rainy season and (14%) in stems. It is concluded that we results indicate that *C. longirostrata* can contribute to diversified and protein-rich forage resources in smallholder systems under tropical conditions.

KEYWORDS

chipile, frequency of cutting, biomass yield, crude protein, Oaxaca

1 Introduction

The global and national human population has increased substantially, increasing the demand for food for human consumption; however, food production grows at a different rate than the former, and, therefore, food self-sufficiency is not achieved. This situation is of such concern that it is included in the 2010–2050 agenda as a central objective (Van Dijk et al., 2021). The inhabitants of rural communities classified as extremely poor use native floristic resources as food sources, which for them are of great importance because of their low acquisition cost and even only collect them on their properties or in their backyards; among these resources is the chipile or chipilin *Crotalaria longirostrata* Hook & Arn; this species is native to southeastern Mexico where its leaves and tender stems are consumed as vegetables (Camarillo-Castillo and Mangan, 2020; Maldonado-Peralta et al., 2023) in a wide variety of stews and in rations for small ruminants (Fernández-Suárez et al., 2013; Palacios-Pola et al., 2016; Solórzano-Juarez, 2020; Salinas-Morales et al., 2022; Calonico and De la Rosa-Millan, 2023).

This species has important nutritional characteristics, among them are its high crude protein content, which can vary from 23.1 to 38.3% (Juárez-Fuentes et al., 2014), its leaves have high values of calcium, iron, vitamins such as thiamine, riboflavin, niacin, and ascorbic acid, and the essential amino acid lysine (Jiménez-Aguilar & Grusak, 2015; Mateos-Maces et al., 2020; Mendez-Lopez et al., 2023) and metabolites secondary (López et al., 2022). Therefore, the leaves of these plants can supplement the diet of the most economically vulnerable people and reduce malnutrition problems. These attributes make this plant well-valued as food for both humans and animals. In addition, as they belong to the *Fabaceae* family, they establish symbiosis with nitrogen-fixing bacteria; this aspect is important because it can reduce costs for the application of industrial nitrogen and damage to the environment; therefore, biological nitrogen fixation, being a sustainable process, can help improve the cultivation of chipile (CHP) and plants associated with CHP, coexisting in the same space in places with low levels of nitrogen in the soil (Camarillo-Castillo and Mangan, 2020).

However, despite the attributes that these plants possess, there is a lack of studies on agronomic management and the forage value of this species in Mexico (Mariaca-Méndez, 2012; Ventura-Ríos et al., 2022; Calonico and De la Rosa-Millan, 2023; Maldonado-Peralta et al., 2023). This study aimed to evaluate the biomass yield and crude protein content of *C. longirostrata* under different cutting frequencies and intensities, to assess its potential as forage for small ruminants on the coast of Oaxaca, Mexico.

2 Methods

The experiment was conducted from September 12, 2023, to April 12, 2024 in the rainy and dry season, at the Tecnológico Nacional de México/Instituto Tecnológico de Pinotepa, in the academic unit located in the municipality of San José Estancia Grande, Oaxaca, Mexico, located at coordinates 16° 22' N and 98° 13' W at 73 m above sea level and a warm sub-humid climate (García, 2004). The seeds used were harvested from a CHP plantation at the Instituto Tecnológico de

Pinotepa. When the fruits were dehydrated, the seeds were manually extracted, and the impurities were eliminated. Subsequently, they were introduced in a cloth and immersed in boiling water for 50 seconds to break dormancy (Rojas-García et al., 2021; Bertsouklis et al., 2023) and were manually sown in plastic germination trays of 200 cells, previously filled with soil from the place where the study was conducted, and two previously treated seeds were placed in each cell.

2.1 Soil preparation

An agricultural tractor was used to prepare the land where the study was carried out; three harrow passes were made until the soil was well loosened. Subsequently, three blocks of 36 x 7.8 m were plotted, with 12 experimental units of 7.8 x 3.0 m in each block, totaling 36 units. The sampling unit, measuring one square meter, contained 15 chipile plants. Two factors were evaluated: cutting frequency with three levels 35, 70, and 105 days and four cutting heights 5, 10, 15, and 20 cm above ground level; the combination of these was 12 treatments; in each block, these were randomly assigned. Four days after the plants were four days old, they were transplanted in furrows 50 cm apart and 25 cm from plant to plant; weed control was manual, and no fertilization was applied.

The variables evaluated were total biomass yield, dry weight of leaves, and dry weight of stems. To do this, biomass samples were collected with scissors, with 15 plants per square meter for each treatment. The samples were then weighed on a scale. Approximately 25% of the cut green material was separated into leaf and stem components, and their weight was recorded. They were placed in identified perforated paper bags and placed in a forced-air electric oven for 48 hours until they reached a constant weight; they were then reweighed, and their dry weight was recorded. The percentage of moisture and dry matter was determined, and the dry matter yield per hectare for total biomass, leaves, and stems was estimated using the formula $\text{kg DM ha}^{-1} = (\text{kg of green forage/ha}) \times (\text{percentage of dry matter}/100)$. Another variable was the crude protein content in leaves and stems, which was determined using the dry leaf and stem samples used for dry matter yield, and determined using the technique described by AOAC (2005).

2.2 Data analysis

The treatments were distributed in a randomized complete block design. The data obtained were subjected to analysis of variance using the SAS, Institute (2009), and the means of the treatments were compared using Tukey's $\alpha=0.05$ test.

3 Results and discussion

3.1 Biomass yield

Figure 1 presents the averages of total dry matter yield by rainy and dry season. A difference ($p<0.05$) was found between rainy and

dry seasons, in the rainy season, the highest yields occurred in the 70-days with 5892 kg of dry matter (DM) and, lower value 836 kg DM ha⁻¹ in 35-day. In rainfall, the average DM yield was reduced from 5630 to 2863 when intensity decreased from 5.0 cm to 20 cm due to a more significant amount of residual biomass left as a remnant. At both times of the year, the highest biomass yields occurred at the frequencies of 70 and 105 days, 5644 and 5147 kg DM ha⁻¹ in rain and dry, respectively ($p < 0.05$). The lowest values occurred at the frequency of 35 days for the seasons above with values of 836 and 2433 kg DM ha⁻¹ in rain and dry, same order; the higher value in the dry may be because when cutting the CHP plants in the rain, this stimulated a more significant emission of shoots by the plants and, with it a higher dry matter yield.

In the rainy season, the intensity of 5.0 cm presented the highest DM yield in the three frequencies of 35, 70, and 105 days, compared to the intensities of 10, 15, and 20 cm, which presented lower yields; this may be because, when cutting the plants at a height of 5 cm concerning the soil level, the amount of biomass harvested is more significant than the others. However, in the dry season, the same situation did not occur since at intensity of 5.0 cm, the lowest yield of 2203 kg DM ha⁻¹ was obtained compared to that observed at the intensities of 10, 15 and 20 cm; this may be due to a more significant amount of dead plants due to the effect of the cutting intensity applied.

The results found in this study are higher in the rainy season than those reported by Maldonado-Peralta et al. (2023) for CHP at a cutting frequency of 64 days; they report values of 3406, 3500 and 4200 kg DM ha⁻¹ at planting densities of 200 000, 100–000 and 50–000 plants ha⁻¹, respectively. these differences may be associated with the planting densities used; a study by Rodas-Barrios (2015) reports values for total green biomass of 65.8-ton ha⁻¹ that exceed those found in this research; this may be mainly because they are not on the same base and, possibly, due to environmental factors such as rainfall.

Pardo-Aguilar et al. (2021) found that successive cuts at 30 days of age in CHP plants increased total dry matter yield by up to 311%.

Camarillo-Castillo and Mangan (2020) report yields of 6293 kg ha⁻¹ of fresh biomass in CHP, with application of 40 kg N2 ha⁻¹. In another study, Ríos-Hilarario et al. (2022) reported average yield values of 527 and 28–363 kg DM ha⁻¹ at 30 and 75 days of plant age, respectively, for *Crotalaria juncea*, at 30 days, their results are lower than those found in this research in rain and dry. However, they are higher at 75 days; these differences may be due to a higher density of plants per hectare employed by them. Akanvou et al. (2001) reported yields for *Crotalaria juncea* of 9–000 kg DM ha⁻¹, which was higher than in this research. The observed differences may be since it is another species despite belonging to the same genus.

3.2 Leaf yield

The means of the dry matter yield of the leaf component are shown in Figure 2, and differences were found between seasons ($p < 0.05$), frequencies, and cutting intensities. The highest leaf yields 2433 and the lowest 643 kg DM ha⁻¹ occurred in the rainy season in the 70-days and 35-days frequency, respectively ($p < 0.05$); In the dry season, the highest production was 1904 kg DM ha⁻¹ with a frequency of 35 days; however, when comparing the averages between both seasons, it was higher at 1716 kg DM ha⁻¹ in the dry season and lower at 1425 kg DM in the rainy season. When observing the DM yields among cutting intensities in the rainy season, a reduction is noted as cutting intensity decreases. This may be due to leaving a good proportion of uncut leaves on the plants.

The higher DM yields observed during the dry season are since, because of cutting, the plants are stimulated to produce more branches, which in turn increases the DM yield. Pardo-Aguilar et al. (2021) found that when CHP plants being subjected to successive cuts every 30 days can increase leaf DM production by up to 311%; in this study, the 35-days cutting in the dry season was more significant than the one carried out in the rainy season. Maldonado-Peralta et al.

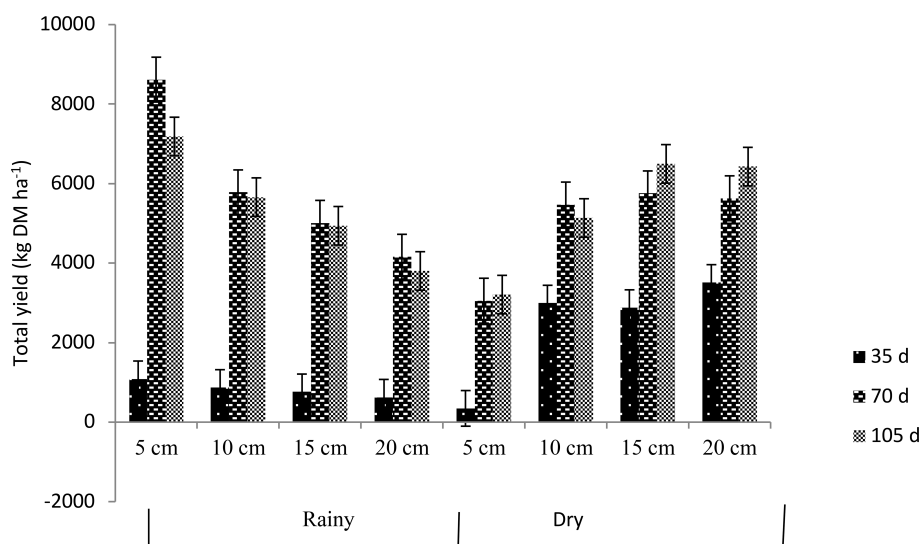


FIGURE 1

Total dry matter yield (kg ha⁻¹) of chipile during the rainy and dry seasons with varying cutting frequency and intensity in the dry tropics.

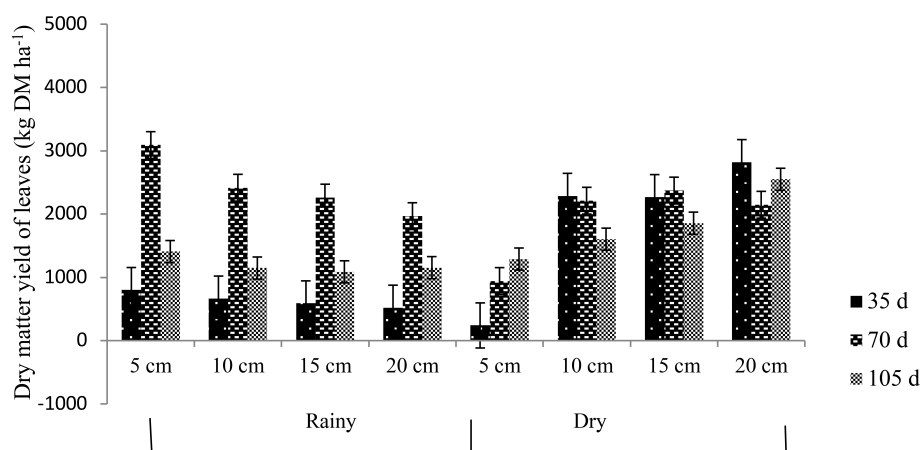


FIGURE 2

Dry matter yield of chipile leaf (kg ha⁻¹) during rainy and dry seasons with varying cutting frequency and intensity in the dry tropics.

(2023) found higher percentages of the leaf component in the cutting frequencies of 15 to 36 days regardless of the sowing density, which coincides with the findings of this study; the highest proportions of leaves occurred in the frequencies of 35 to 70 days.

Ríos-Hilario et al. (2022) reported that the leaf component represented 62 and 15% of the total biomass harvested at 30 and 75 days of age in *Crotalaria juncea* plants, respectively, Álvarez-Vázquez et al., 2020; Mendoza-Pedroza et al., 2010 they mentioned that the highest proportion of harvested leaf was recorded in the cuttings at 3 and 4 weeks of age. These findings confirm the behavior of the leaf component to decrease in the total yield of the CHP, with increasing age of the CHP crop.

3.3 Stem dry matter yield

Stem DM yields are shown in Figure 3; a difference was found ($p < 0.05$) between seasons; regardless of the cutting intensities the

highest yield 4199 kg DM ha⁻¹ occurred in the rainy season in the 105-days frequency; with a lower yield of 194 kg DM in 35 days. However, in the dry season the highest yield of 3061 kg DM occurred at 70 days and the lowest value 530 kg at 35 days. When comparing yields between cutting intensities during the rainy season, they decrease as the intensity decreases from 3866 to 5 cm and 1650 kg DM at 20 cm, this behavior did not occur in the dry season, possibly due to the effect of previous cuts made to the plants. This is due to a large number of stems remaining on the plants at the lower intensities.

A study by Maldonado-Peralta et al. (2023) reported that the proportion of stems increased at the frequency of 64 days with a value of 73% for CHP plants; these results coincide with what was found in this study, in which stem yields increased at cutting frequencies of 70 and 105 days, this may be because plants at that age pass from the vegetative to the reproductive phase, slowing the production of leaves and increasing the weight of the stems. Ríos-Hilario et al. (2022) reported that the proportion of stems in the biomass was 37 and 68% cut at 30 and 75 days of cultivation,

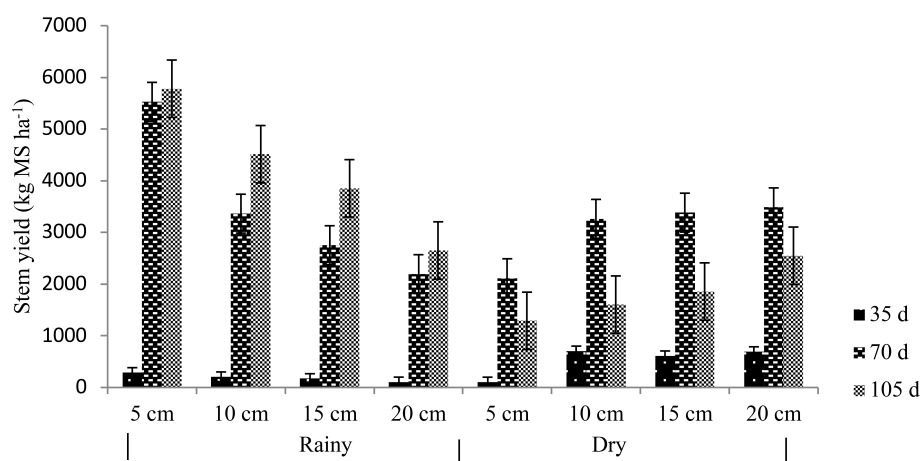


FIGURE 3

Dry matter yield of stem (kg ha⁻¹) of chipile in rainy and dry seasons, varying the frequency and intensity of cutting in the dry tropics.

respectively; similar behavior was found in this study in the rainy season for cutting frequencies of 35, 70 and 105 days where the contribution of stems to the total biomass yield was 23.2, 59 and 78% in that exact order; in alfalfa crop,

Álvarez-Vázquez et al. (2020) found that the stem component contributes in the total biomass yield in 41% when it is cut at 35 days of the age of the plants; in another research study conducted by (Mendoza-Pedroza et al., 2010) on alfalfa crop, they found that with increasing plant age at cutting, the contribution of stem to biomass yield was higher in all seasons of the year, this coincides with the results of this study in which, it is observed that stem yields increased with increasing age of CHP plants. Camarillo-Castillo and Mangan (2020) report 661 kg DM ha⁻¹ for stems in unfertilized plants without rhizobia inoculation.

3.4 Crude protein in leaves

Table 1 shows the means of crude protein in leaves (CP) by the rainy and dry seasons; for both seasons of the year, a difference ($p < 0.05$) was found in this variable. Higher values 37% were found in crude protein of leaves during the dry season than in the rainy season 32%, when comparing the averages within the period, the best values of 32.5% occurred at 35 and 70 days in rainy conditions and 37.5% in dry conditions at the same cutting frequencies; the protein content in dry conditions was 15.4% higher than that observed in rainy conditions. The values of crude protein among intensities, within the season, were not different ($p > 0.05$) for rainy or dry periods, indicating that the cutting height does not influence the protein content; however, the cutting frequency affects the protein level in the leaves of chipile. The protein content of these

plants indicates that they have good potential to be used as a protein food for humans and small ruminants.

The contents of CP found in this research maintain a similar trend to that found by Maldonado-Peralta et al. (2023). The highest contents of CP were presented in the cutting frequencies of 15 to 43 days after cutting, with values of 30 to 25%, respectively. However, they are lower than those found in this study, possibly the determination was made for the whole plant; on the other hand, they are like those reported by (Pardo-Aguilar et al., 2019) for *C. longirostrata* at 30 days of age, consigns 31.6% in leaves for both rainy and dry seasons; in another study by (Rodas-Barrios, 2015), he reports 37.6% of CP for CPH at the age of 75 days, which is equal to that found in this research at the frequency of defoliation of 70 days, in the dry period; for his part, Laguna-González (2016) reports CP content of 40.9% for CHP, a value higher than those obtained in this study in the rainy and dry seasons. For leaves with petiole in chipile plants,

3.5 Crude protein in stems

The means of crude protein content in stems (CPC) are shown in Table 2 for the rainy and dry seasons; differences were found ($p < 0.05$) in both seasons because of treatments, with the highest protein content of 14% at the frequency of 35 days in both rainy and dry seasons, and the lowest value of 7% occurred at 70 and 105 days in the rainy season. The protein values between intensities within the season showed no variations ($p > 0.05$) with average contents of 9% in the rainy season and 11.5% in the dry season. The average protein content of the rain represented 75% of that achieved in the dry period. The above indicates that, at an age greater than 70 days of the stems, they do not present attractive attributes as food for

TABLE 1 Crude protein content (%) in leaves of chipile plants in rainy and dry season with varying frequency and intensity of cutting in the tropics.

Total dry matter yield Intensity						
Rain						
Frequency	5 cm	10 cm	15 cm	20 cm	Average	CV
35 d	34Ba	33Ba	33BCa	33Ca	33	2.7
70 d	30Cc	31Cb	32CDB	33Ca	32	0.6
105 d	31Cab	32BCa	30DB	28Dc	30	1.9
Average	32	32	32	31	32	
Dry						
35 d	37Aa	38Aa	38Aa	37Aa	37	1.7
70 d	37Abc	40Aa	39Aab	36ABc	38	2.5
105 d	35Ba	34Ba	35Ba	35Ba	35	2.2
Average	36	37	37	36	37	
CV	1.8	2.2	2.4	1.9		

abcd, Means with the same lowercase literal in the same row, not different ($P > 0.05$); ABCD, Means with the same uppercase literal in the same column, not different ($P > 0.05$); CV, Coefficient of variation.

TABLE 2 Crude protein content (%) in stems of chipile plants in rainy and dry season by varying the frequency and intensity of cutting in the dry tropics.

Protein in stems Intensity						
Rain						
Frequency	5 cm	10 cm	15 cm	20 cm	Average	CV
35 d	13Bc	14Abc	14Aab	15Aa	14	3.4
70 d	7Ca	7Dbc	7Cc	7Dab	7	3.1
105 d	7Ca	8Da	7Ca	7Da	7	2.8
Average	9	10	9	10	9	
Dry						
35 d	13Aab	12ABb	14Aab	15Aa	14	7.5
70 d	15Aa	10Cb	9Bc	10Cbc	11	2.9
105 d	9Cb	11BCa	11Ba	11Aa	11	4.7
Average	12	11	11	12	12	
CV	4.9	5.4	5.8	4.0		

abcd, Means with the same lowercase literal in the same row, not different ($P > 0.05$); ABCD, Means with the same uppercase literal in the same column, not different ($P > 0.05$); CV, Coefficient of variation.

humans or ruminants, because the intracellular content is reduced by increasing the crude fiber, affecting the digestibility of the stems.

For stems of CHP plants cut at 30 days of age (Pardo-Aguilar et al., 2019) found a CP content of 20.17%; this value is higher than those obtained in this study in the rainy and dry season at any frequency of plant cutting, this same author reports higher CP content in leaves than in stems, like (Juárez-Fuentes et al., 2014). Torres-Salado et al. (2020) report a higher CP content in leaves at the highest frequencies and intensities of defoliation, regardless of plant genotype, being consistently lower in stems than in leaves.

4 Conclusions

In conclusion, *Crotalaria longirostrata* stands out for its high potential as a source of protein, representing a valuable alternative for strengthening food security in the Oaxacan coastal region. The best biomass yields are achieved at 70 days, while the optimal protein content occurs between 35 and 70 days, allowing for efficient use.

In addition to its significant nutritional contribution, this plant offers an opportunity to diversify the local food base, contributing to more sustainable and resilient agricultural systems in tropical areas. Promoting its cultivation and consumption in the coastal region of Oaxaca encourages the preservation of traditional practices, food sovereignty, and the nutritional well-being of local populations. In terms of regional development, the integration of this legume into intercropping systems can strengthen the food supply and the rural economy, promoting agroecological practices and strengthening the social fabric through the valorization of endogenous natural resources.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Author contributions

HA-A: Methodology, Validation, Formal analysis, Conceptualization, Supervision, Visualization, Writing – original draft, Writing – review & editing, Investigation. DM-P: Methodology, Conceptualization, Investigation, Writing – original

draft, Formal analysis. AP-S: Methodology, Formal analysis, Investigation, Conceptualization, Writing – original draft. MS-M: Conceptualization, Formal analysis, Methodology, Investigation, Writing – original draft. AR-G: Conceptualization, Formal analysis, Methodology, Writing – original draft, Investigation. MM-P: Formal analysis, Methodology, Writing – original draft, Conceptualization, Investigation. AC-M: Conceptualization, Formal analysis, Writing – original draft, Investigation. AA-C: Methodology, Investigation, Conceptualization, Writing – original draft. IG-M: Validation, Conceptualization, Methodology, Supervision, Writing – review & editing, Investigation, Writing – original draft, Formal analysis, Visualization.

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