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CUTTING POSITION ON THE QUALITY OF AMAZONIAN SPINACH (*Alternanthera sessilis* (L.) R.Br. ex DC.) SEEDLINGS

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Abstract

Brazil has a great biodiversity of unconventional food plants, such as spinach from the Amazon, an underutilized species, highly adaptable and with enormous food potential. The objective of this work was to evaluate the cutting position on the quality of Amazonian spinach (*Alternanthera sessilis* (L.) R.Br. ex DC.) seedlings. The experiment was carried out in the experimental garden of the Federal University of Acre under greenhouse conditions, from October to November 2022. The experimental design was completely randomized, with three treatments and ten replications, totaling 30 experimental units. Cuttings were collected from stem segments harvested from three positions along the shoot of mature donor plants: the lower third (LT), middle third (MT), and upper third (UT). There was a significant difference for stem diameter, leaf length, total number of shoots, shoot fresh mass, shoot dry mass, total fresh mass. There was no statistical difference for seedling quality index. Regardless of the part of the plant used as propagation material for the species, there was no difference in the quality of seedling formation. All seedlings produced, regardless of the part of the plant used, presented high quality and complete formation.

Keywords: Amaranthaceae; Seedling production; Unconventional vegetables.

POSIÇÃO DE CORTE DA ESTACA NA QUALIDADE DAS MUDAS DE ESPINAFRE-DA-AMAZÔNIA (*Alternanthera sessilis* (L.) R.Br. ex DC.)

Resumo

O Brasil apresenta grande diversidade de plantas alimentícias não convencionais, entre elas o espinafre da Amazônia, espécie ainda pouco explorada, com elevada capacidade de adaptação e expressivo potencial alimentício. O objetivo deste trabalho foi avaliar o efeito da posição de retirada das estacas na qualidade de mudas de espinafre-da-Amazônia (*Alternanthera sessilis* (L.) R.Br. ex DC.). O experimento foi conduzido na horta experimental da Universidade Federal do Acre, em casa de vegetação, entre outubro e novembro de 2022. Utilizou-se delineamento inteiramente

casualizado, com três tratamentos e dez repetições, totalizando 30 unidades experimentais. As estacas foram obtidas de segmentos caulinares retirados de três posições dos ramos de plantas matrizes adultas: terço inferior (TI), terço médio (TM) e terço superior (TS). Houve diferença significativa para diâmetro do caule, comprimento das folhas, número total de brotações, massa fresca da parte aérea, massa seca da parte aérea e massa fresca total. Não houve diferença estatística para o índice de qualidade das mudas. Independentemente da posição de retirada do material propagativo, não houve diferença na qualidade das mudas formadas. Todas as mudas produzidas apresentaram elevada qualidade e formação completa.
Palavras-chave: Amaranthaceae; produção de mudas; hortaliças não convencionais.

Introduction

Over the past few decades, there has been significant growth in the search for food alternatives that combine nutritional quality and low production costs. In this context, Brazil stands out for its broad plant biodiversity and high nutritional potential, which favors the appreciation of Unconventional Edible Plants. These species include native or exotic plants, cultivated or wild, which can be consumed both fresh and processed, contributing to dietary diversification and nutritional security (Kinupp; Lorenzi, 2014).

Both conventional and unconventional edible plants contain high levels of vitamins, fiber, and essential minerals, performing fundamental metabolic functions in the human body (Bhuyan *et al*., 2018; Alam *et al*., 2022). PANCs, in particular, have been gaining prominence due to their potential for integration into diversified food systems and sustainable agricultural practices, constituting viable alternatives for subsistence and expanding the variety of products available on the market (Liberato *et al*., 2019).

Among the most promising species is Amazon spinach (*Alternanthera sessilis* (L.) R.Br. ex DC.), a member of the Amaranthaceae family. Although popularly considered a weed by some, it is an unconventional vegetable with high nutritional value. Native to tropical regions of the Americas, it is highly adaptable and resilient, as well as having a high protein content, which can reach up to 25%, exceeding that of several vegetables traditionally consumed in Brazil (Kinupp; Lorenzi, 2014; Bhuyan *et al*., 2018).

The plant has rounded, slightly wrinkled leaves and small, white flowers. It is traditionally cultivated on a small scale, primarily sold in open-air markets in the states of Amazonas and Pará (Bassingthwaighte, 2022; Silva *et al*., 2022). It thrives in tropical environments with fertile soils rich in organic matter. Propagation occurs primarily asexually, through vegetative cuttings, a

characteristic associated with its hardiness, rapid growth, and ease of rooting (Kinupp; Lorenzi, 2014; Bhuyan *et al*., 2018; Alam *et al*., 2022).

Despite its high nutritional and agronomic potential, there is still a scarcity of scientific research focused on the cultivation and management of Amazon spinach, especially regarding seedling production. Obtaining high-quality seedlings is an essential step in establishing productive and sustainable crops, as it directly influences growth, vegetative development, and final crop yield. Morphological and physiological factors of the parent plants can significantly affect this process, making the adoption of appropriate propagation techniques essential (Taiz *et al*., 2019; Silva *et al*., 2022; 2023; Lima *et al*., 2025).

Therefore, this study aims to evaluate the quality of Amazon spinach (*Alternanthera sessilis*) seedlings produced from different cutting positions, aiming to identify agronomic strategies that enhance propagation efficiency and contribute to the consolidation of this species as an alternative food and production source in the Amazon region.

Materials and methods

The study was conducted at the experimental unit of the Federal University of Acre (Universidade Federal do Acre - UFAC), located in the municipality of Rio Branco, Acre State, Brazil, at geographic coordinates 67º42’18’’ W and 10º01’30’’ S, with an elevation of approximately 164 meters. The mean temperature in the region ranges from 21.6 °C to 28.5 °C (Climate-data, 2022). The experiment was carried out between October and November 2022 in a greenhouse structure.

The experimental design was completely randomized (CRD), with three treatments and ten replicates, totaling 30 experimental units. Treatments consisted of cuttings collected from different portions of the mother plant: lower third (LT), middle third (MT), and upper third (UT). Each cutting contained two nodes in all treatments.

The experiment was conducted using 200 cm^3 polystyrene containers. All containers were labeled and perforated for drainage, then filled with a commercial substrate. The cuttings were placed according to the defined treatments and arranged randomly in the greenhouse according to the pre-established randomization scheme.

The substrate used was the commercial product MecPlant®, composed of pine bark, vermiculite, acidity corrector, and macronutrients, with a water holding capacity (WHC) of 60%, cation exchange capacity (CEC) of 200 mmolc/kg, and maximum moisture content of 60%, according to the product label.

Cuttings were collected from the lower, middle, and upper thirds of adult *Alternanthera sessilis* (Amazon spinach) plants grown in experimental garden beds at UFAC. Each cutting was

approximately 10 cm long, 4 mm in diameter, with two internodes and four axillary buds. Branches were pruned from the mother plants using pruning shears and immediately stored in a shaded location to prevent desiccation and depletion of carbohydrate reserves prior to planting.

Subsequently, cuttings were measured with a precision caliper and separated according to their original position on the branch. To minimize environmental variability, the experiment was conducted in a greenhouse with controlled external conditions. Irrigation was performed manually on a daily basis, according to plant needs.

The cuttings were cultivated for 28 days, allowing complete seedling formation. Afterwards, the seedlings were transferred to the experimental unit of the Horticulture Laboratory of the Graduate Program in Agronomy/Plant Production at UFAC for evaluation.

The following variables were measured: plant height (PH), stem diameter (SD), leaf length (LL), leaf width (LW), total number of leaves (TNL), total number of shoots (TNS), number of leaves per shoot (NLS), root length (RL), fresh shoot biomass (FSB), dry shoot biomass (DSB), fresh root biomass (FRB), dry root biomass (DRB), total fresh biomass (TFB), total dry biomass (TDB), and Dickson Quality Index (DQI).

For each treatment, ten seedlings were evaluated. Plants were carefully removed from the containers with the substrate still attached to the roots. The root system was separated from the shoot, and roots were washed thoroughly to remove substrate residues. Excess water was blotted off with paper towels before measurements.

Leaf length, leaf width, stem diameter, and root length were measured using a precision caliper, while plant height was measured with a graduated ruler. Total leaf number and shoot number were determined by direct counting. Fresh and dry biomass of shoots and roots were determined using an analytical balance. All plant material was oven-dried at 65 °C in a forced-air circulation oven for 48 hours, until constant weight was achieved.

Seedling quality was assessed using the Dickson Quality Index (DQI), following Dickson *et al.* (1960), calculated as:

$$DQI = \frac{TDB\ (g)}{\frac{PH\ (cm)}{SD\ (mm)} + \frac{DSB\ (g)}{DRB\ (g)}}$$

After data collection and tabulation, outliers were verified using the Grubbs test (1969), error normality was assessed using the Shapiro-Wilk test (1965), and variance homogeneity was evaluated with the Cochran test (1941). Analysis of variance (ANOVA) was then performed, and when significant differences were detected, means were compared using Tukey's test (1949) at a 5% probability level.

To determine the influence of branch position on cutting propagation, multivariate analysis was conducted through principal component analysis (PCA) and multiple correlation analysis of the

evaluated variables. All statistical analyses were performed using the open-source software R (R Core Team, 2016).

Results and discussion

Significant effects ($p < 0.05$) were observed among treatments for stem diameter (SD), leaf length (LL), and total number of shoots (TNS), as well as for shoot fresh biomass (SFB), shoot dry biomass (SDB), and total fresh biomass (TFB). No statistically significant differences were detected ($p > 0.05$) for the remaining variables.

For the morphometric traits LL, TNS, SFB, SDB, and TFB, the cuttings from the middle third of the mother plant exhibited the highest mean values. The only exception was SD, where the lower third cuttings produced the thickest stems (Tables 1 and 2). This pattern indicates that middle cuttings are physiologically more balanced, resulting in improved early seedling performance.

Table 1. Plant height (PH), stem diameter (SD), leaf length (LL), leaf width (LW), total number of leaves (TNL), total number of shoots (TNS), and number of leaves per shoot (NLS) of *Alternanthera sessilis* seedlings produced from different cutting positions.

Cutting position	PH (cm)	SD (mm)	LL (cm)	LW (cm)	TNL	TNS	NLS
Upper third	9.90 a	3.36 c	4.05 b	4.08 a	25.0 a	3.70 ab	6.88 a
Middle third	9.91 a	4.30 b	4.59 a	3.96 a	27.4 a	4.40 a	6.30 a
Lower third	9.68 a	5.80 a	3.99 b	3.86 a	22.3 a	3.40 b	6.85 a
CV (%)	12.50	9.98	10.49	11.65	18.78	17.2	14.28

*Means followed by the same letter in the column do not differ by Tukey's test ($p > 0.05$).

The superior performance of seedlings derived from middle cuttings can be attributed to greater uniformity in seedling formation and more favorable physiological reserves, which promote both rooting and shoot development. Similar results have been reported for vegetative propagation of other tropical herbaceous species (Kinupp; Lorenzi, 2014; Bhuyan *et al*., 2018; Alam *et al*., 2022). This trend was further confirmed by principal component analysis (PCA), which revealed a lower dispersion and more compact grouping of data for middle cuttings compared with the upper and lower cutting positions (Figure 2).

Table 2. Root length (RL), shoot fresh biomass (SFB), shoot dry biomass (SDB), root fresh biomass (RFB), root dry biomass (RDB), total fresh biomass (TFB), total dry biomass (TDB), and Dickson Quality Index (DQI) of *Alternanthera sessilis* seedlings produced from different cutting positions.

Cutting position	RL (cm)	SFB (g)	SDB (g)	RFB (g)	RDB (g)	TFB (g)	TDB (g)	DQI
Upper third	19.80 a	11.19 ab	1.17 b	2.19 a	0.14 a	13.38 ab	1.31 a	0.12 a
Middle third	20.00 a	12.57 a	1.43 a	2.35 a	0.16 a	14.92 a	1.59 a	0.14 a
Lower third	20.65 a	9.50 b	1.30 ab	2.63 a	0.18 a	12.12 b	1.48 a	0.16 a
CV (%)	14.20	15.92	16.80	23.37	23.26	14.70	17.43	17.84

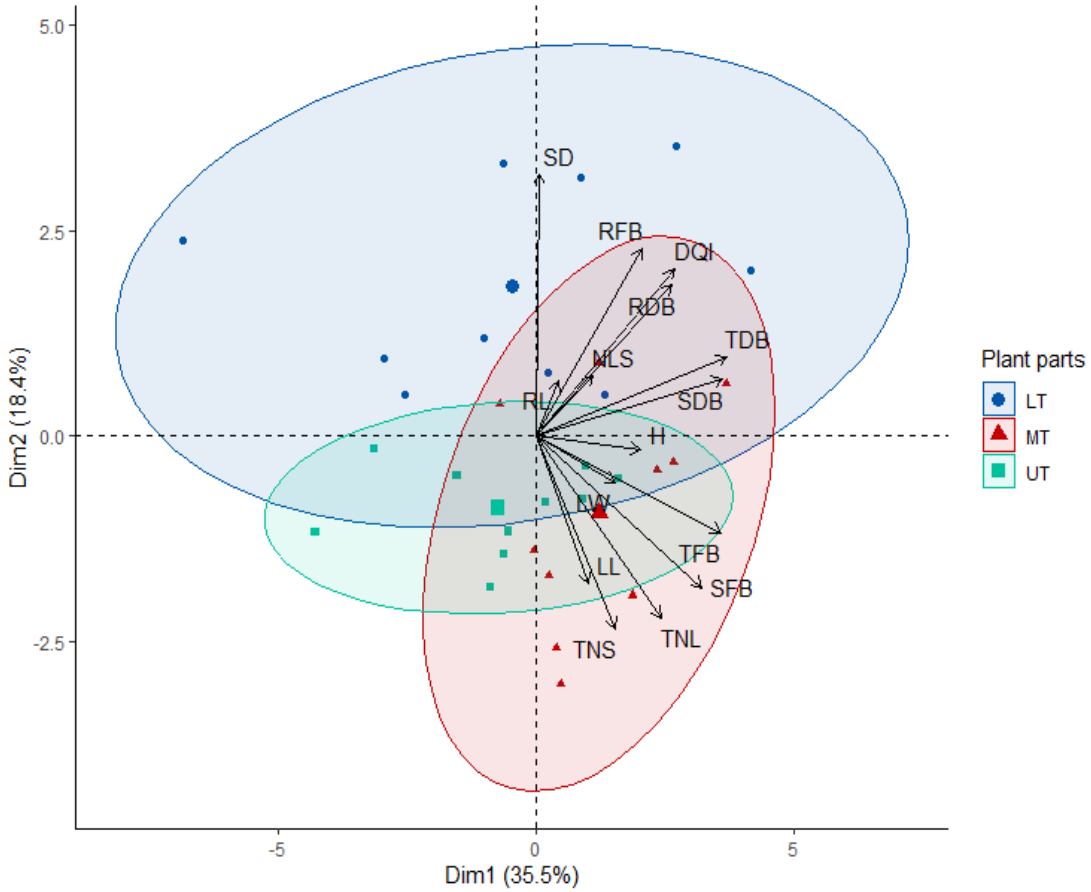
*Means followed by the same letter in the column do not differ by Tukey's test ($p > 0.05$).

Although some morphometric and biomass variables differed significantly, the Dickson Quality Index (DQI) showed values between 0.12 and 0.16, with no statistical differences among treatments ($p > 0.05$). This indicates that all cutting positions are suitable for vegetative propagation of *A. sessilis*, ensuring seedling quality for field establishment.

The DQI is widely recognized as one of the most robust indicators of seedling quality because it integrates morphological attributes related to biomass accumulation and plant architecture, including height, stem diameter, and biomass partitioning (Dickson *et al*., 1960). Although originally proposed for forest species, the index has been successfully applied to herbaceous and horticultural crops as an effective measure of seedling vigor and structural balance (Santos *et al*., 2017; Silva *et al*., 2022; 2023; Lima *et al*., 2025). The absence of significant differences in DQI reinforces the interpretation that variations observed in isolated growth variables did not compromise the overall quality of the seedlings produced.

The correlation matrix (Figure 1) revealed weak but positive associations among PH, LW, TNL, TNS, SFB, RFB, and RDB, with no significant correlation between RL and other variables, suggesting that root length did not substantially influence shoot development. Similarly, LL and LW were not significantly correlated, indicating independent patterns of leaf expansion in length and width a trend also reported in other herbaceous crops (Bhuyan *et al*., 2018; Alam *et al*., 2022).

Figure 1. Correlation analysis among the variables evaluated in *Alternanthera sessilis* (Amazon spinach) seedlings produced from different cutting positions.

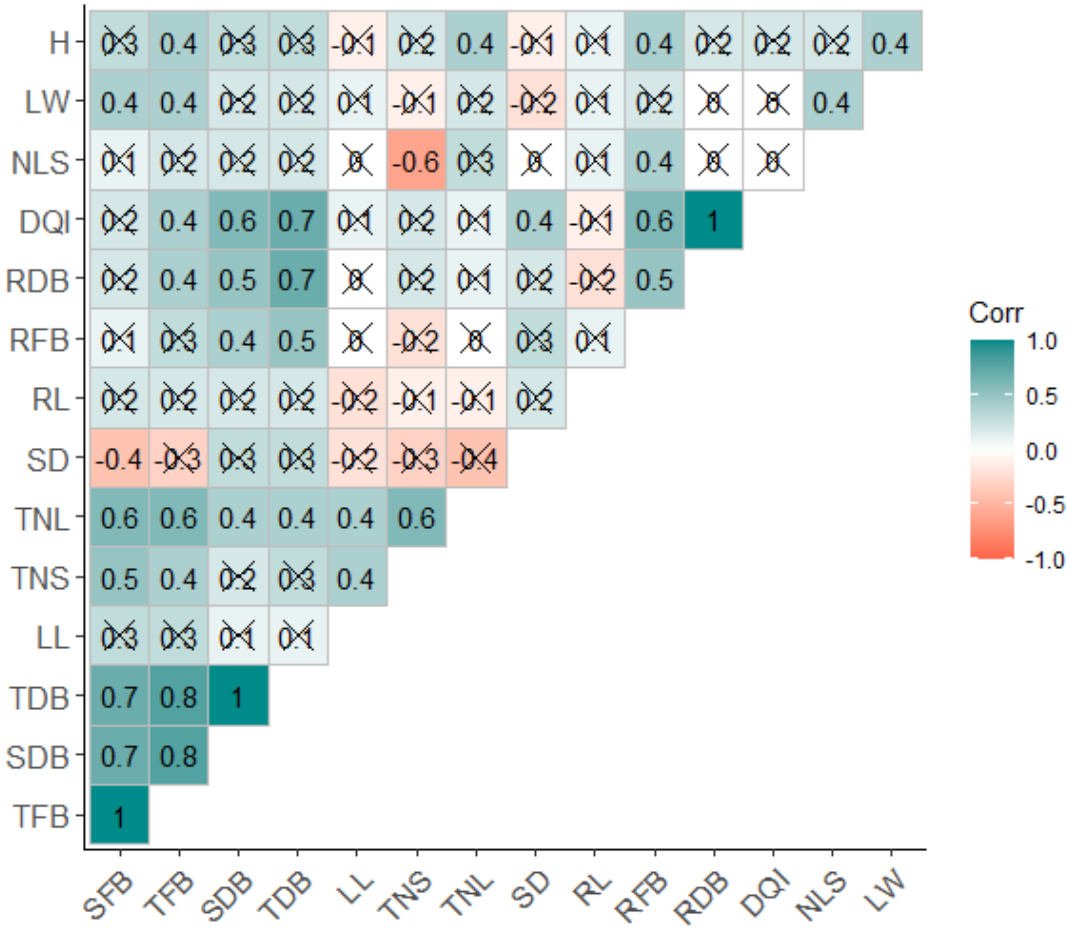


*Positive and negative correlations are represented in blue and red, respectively, with color intensity proportional to the correlation coefficient. Variables: plant height (H), stem diameter (SD), leaf length (LL), leaf width (LW), total number of leaves (TNL), total number of shoots (TNS), number of leaves per shoot (NLS), root length (RL), shoot fresh biomass (SFB), shoot dry biomass (SDB), root fresh biomass (RFB), root dry biomass (RDB), total fresh biomass (TFB), total dry biomass (TDB), and Dickson Quality Index (DQI).

Strong positive correlations were observed between DQI and biomass variables (TDB, SDB, and RDB). This reflects the central role of biomass accumulation in determining overall seedling quality. Higher shoot and root biomass indicate greater photosynthate allocation, which enhances survival and establishment potential in the field (Fachinello *et al*., 2005; Santos *et al*., 2017).

PCA explained 53.9% of the total variance in the data, with 35.5% attributed to Dim1 and 18.4% to Dim2 (Figure 2). Dim1 was primarily associated with SD, TNL, SFB, SDB, RFB, RDB, TFB, TDB, and DQI, while Dim2 represented residual variability. The PCA biplot clearly shows a more cohesive grouping for the middle third treatment, suggesting greater morphological stability and uniformity in seedling development compared with the upper and lower thirds.

Figure 2. Principal component analysis (PCA) of variables associated with *Alternanthera sessilis* (Amazon spinach) seedlings produced from different cutting positions.



*Plant height (H), stem diameter (SD), leaf length (LL), leaf width (LW), total number of leaves (TNL), total number of shoots (TNS), number of leaves per shoot (NLS), root length (RL), shoot fresh biomass (SFB), shoot dry biomass (SDB), root fresh biomass (RFB), root dry biomass (RDB), total fresh biomass (TFB), total dry biomass (TDB), and Dickson Quality Index (DQI).

Comparable responses have been reported for other vegetatively propagated species. Furlani *et al*. (2018) observed that cutting position influenced rooting and shoot emission in *Jatropha curcas*, emphasizing the importance of propagule origin for successful vegetative propagation. Classical propagation principles also indicate that physiological maturity and the distribution of endogenous reserves play important roles in determining rooting capacity and initial vegetative growth (Fachinello *et al*., 2005). In the present study, the superior performance of middle cuttings for some morphometric and biomass traits is consistent with these concepts, although the lack of differences in DQI demonstrates that all cutting positions were capable of producing seedlings of equivalent overall quality.

The results obtained for *A. sessilis* are also consistent with the biological characteristics of the species, which include high adaptability, vigorous vegetative growth, and efficient propagation

through stem cuttings (Kinupp; Lorenzi, 2014; Bhuyan *et al*., 2018; Alam *et al*., 2022). From a practical perspective, all stem portions can be successfully used for seedling production, while middle-third cuttings may provide greater uniformity and predictability during the propagation process, representing a useful strategy for commercial production systems.

Conclusions

The cutting position on the stem cuttings did not significantly influence the quality index of Amazon spinach (*Alternanthera sessilis*) seedlings, although some of the morphological characteristics evaluated differed among treatments.

Regardless of the plant part used as a cutting, all treatments produced fully developed and viable seedlings, indicating the species' high propagation potential.

Cuttings taken from the middle third of the plant showed less variability in morphological characteristics, suggesting greater uniformity and stability in seedling quality.

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