Anthropogenic disturbances stimulate natural regeneration of *Euxylophora paraensis*

Huber in a forest fragment in the eastern Amazon

Distúrbios antropogênicos estimulam a regeneração natural de *Euxylophora paraensis* Huber em um fragmento florestal na Amazônia Oriental

Los trastornos antropogénicos estimulan la regeneración natural de Euxylophora paraensis Huber

en un fragmento de bosque en la Amazonía oriental

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Abstract

In the Brazilian Amazon, forest fragments increase annually leading many species to local extinction. *Euxylophora paraensis* Huber is an endemic species whose natural habitat is restricted to the north of Brazil. The natural regeneration was evaluated using 10 circular plots of land each with a radius of 20 meters. The density of regenerating individuals at the edge and the interior of the fragment and the behavior in relation to the parent tree. The Periodic Annual Increment of the diameter and the height were calculated individually for each plant that remained alive during all four measurements and GLM (General Linear Model) was applied to verify if there were differences between environments and the size of the individuals. The results show that the density of natural regeneration increases as the distance from the parent tree increases and has a significant negative correlation with canopy area, the smaller the canopy area the higher the density of individuals. Larger diametric growth was observed in saplings and small trees at the edge and larger increments of height in small trees were observed both at the edge and interior of the fragments. Therefore, natural regeneration is influenced by the process of forest fragmentation where the germination of seeds and establishment of saplings is favorable, however, there were no adult trees at the edge. **Keywords:** Edge effect; Forest fragmentation; Extinction; Endemic specie.

Resumo

Na Amazônia brasileira, os fragmentos florestais aumentam anualmente, levando muitas espécies à extinção local. *Euxylophora paraensis* Huber é uma espécie endêmica cuja ocorrência natural é restrita apenas no norte do Brasil. A regeneração natural foi avaliada utilizando 10 parcelas circulares com 20 metros de raio cada. Avaliou-se a densidade de indivíduos regenerantes na borda e no interior do fragmento e o comportamento em relação à árvore matriz. O Incremento Periódico Anual do diâmetro e da altura foi calculado individualmente para cada planta que permaneceu viva durante todas as quatro medições e aplicado o GLM (Modelo Linear Geral) para verificar se houve diferença entre os ambientes e tamanho dos indivíduos. Os resultados mostram que a densidade da regeneração natural aumenta com o distanciamento da árvore matriz e tem correlação negativa e significativa com a área de copa, ou seja, quanto menor a área de copa maior a densidade dos indivíduos. Observou-se maior crescimento diamétrico em mudas e arvoretas em área de borda e maior incremento em altura nas arvoretas, tanto em borda quanto no interior do fragmento. Sendo assim, a regeneração natural é influenciada pelos processos de fragmentação florestal, onde há favorecimento da germinação das sementes e estabelecimento de mudas, porém na borda a regeneração não apresentou arvoretas até a fase adulta.

Palavras-chave: Efeito de borda; Fragmentação florestal; Extinção; Espécie endêmica.

Resumen

En la Amazonía brasileña, los fragmentos de bosque aumentan anualmente, lo que lleva a muchas especies a la extinción local. *Euxylophora paraensis* Huber es una especie endémica cuya presencia natural está restringida solo en el norte de Brasil. La regeneración natural se evaluó utilizando 10 parcelas circulares con un radio de 20 metros cada una. Se evaluó la densidad de individuos en regeneración en el borde y dentro del fragmento y el comportamiento en relación al árbol matriz. El Incremento Periódico Anual de diámetro y altura se calculó individualmente para cada planta que permaneció viva durante las cuatro mediciones y se aplicó el GLM (Modelo Lineal General) para verificar si había una diferencia entre los ambientes y el tamaño de los individuos. Los resultados muestran que la densidad de regeneración natural aumenta con la distancia del árbol padre y tiene una correlación negativa y significativa con el área del dosel, es decir, cuanto menor es el área del dosel, mayor es la densidad de individuos. Se observó mayor crecimiento diametral en plántulas y árboles en la zona de borde y mayor aumento de altura en los árboles, tanto en el borde como en el interior del fragmento. Así, la regeneración natural está influenciada por procesos de fragmentación del bosque, donde se favorece la germinación de semillas y el establecimiento de plántulas, pero en el límite, la regeneración no mostró árboles hasta la edad adulta.

Palabras clave: Efecto de borde; Fragmentación forestal; Extinción; Especies endémicas.

1. Introduction

Forest fragmentation in the Brazilian Amazon is driven by disorganized land use and occupation, besides populational growth (Haddad, et al., 2015; Laurance, et al., 2018). It significantly lowers the flow of animals, pollen, seeds and, in general, introduces anthropic barriers in the landscape (roads, cities, agriculture, pastures, etc.) that generate severe edge effects (Murcia, et al., 1995; Marques, et al., 2010; Razafindratsima, et al., 2018). Near the edges, fragments exhibit high temperatures, low soil humidity, high luminosity, and strong winds occur (Laurance, et al., 2009; Cheptou, et al., 2017), causing critical ecological changes in the fragments (Laurance, et al., 2018). At the edges of the fragments, intense demographic changes in the natural regeneration of forest species take place, increasing abundance of some (Laurance & Vasconcelos, 2009; Marques, et al., 2010; Magnago, et al., 2017; Duarte, et al., 2018) and declining population of others (Laurance, et al., 2006; Marini, et al., 2012). Variation in the intensity of disturbances causes substantial changes in the spatial distribution of natural regeneration (Schwartz, et al., 2017; Duarte, et al., 2018). Individuals and species can exhibit significant growth variation based on their genetic history and ecological alterations caused by the disturbances (Costa, et al., 2020).

In this context, the processes involved in the natural regeneration of fragmented areas allow the detection and prediction of changes in the next generations, even before the effects are established in the adult community needs investigation. This knowledge is essential for management and development of action plans to conserve biodiversity, avoiding irreparable environmental losses, such as the extinction of rare and threatened species (Marini, et al., 2012; Cheptou, et al., 2017), which is the case of *Euxylophora paraensis* Huber.

Euxylophora paraensis, commonly known as "pau-amarelo" (Brazilian yellowwood or yellowheart), is the only species of the genus *Euxylophora* in the family Rutaceae. It is an endemic species with its distribution restricted to three northern states of Brazil: Amazonas, Tocantins and Pará (Pirani, 2016). Brazilian yellowwood has a long history of use in Pará, where the wood's renowned beauty and structural robustness led to intense harvesting for use as flooring and finishing structures (Freitas, et al., 2019). The substantial exploitation of the species in the 1990s occurred without any planning and drastically reduced its natural populations (Brandão, et al., 2018). Consequently, the intense harvesting associated with restricted distribution and population reduction explained the inclusion of *E. paraensis* in the list of threatened species. In 1998 and 2014, it was classified as "Critically endangered" (Brasil, 2014).

Knowledge of tree species ecology disturbed by human activities forests, especially those of economic interest and threatened, is of utmost importance for sound management and conservation. In this sense, this study aimed to evaluate the

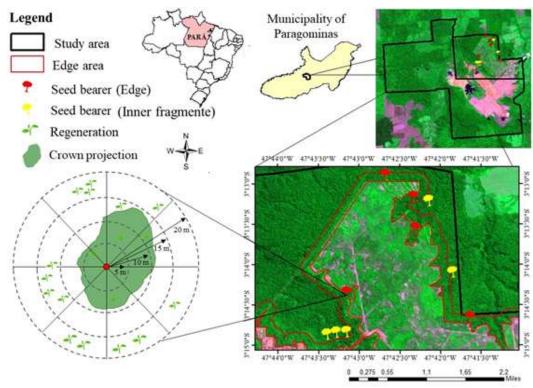
natural regeneration of *E. paraensis* in anthropogenically disturbed environments to answer the following questions: i) are there differences in density of seedlings and saplings between the edge and inside the fragment? ii) are there differences in diameter and height increments between seedlings and saplings of *E. paraensis* between the fragment edge and inner environments? iii) are there variations in the natural regeneration density regarding the distance from the seed bearer and its canopy area?

2. Methodology

2.1 Study area

The research was carried out in a forest fragment near a bauxite mining operation called Platô Miltônia III in the municipality of Paragominas – Pará State, Brazil, between the latitude of 3°15'0" S and longitudes 47°44'0" W (Figure 1).

Figure 1. Location of the research and sketch of the plots in the *Euxylophora paraensis* Huber seed bearers at the edge and in the inner fragment in the municipality of Paragominas, State of Pará, Brazil.





The size of the fragment is 1,989 ha. Several land-uses are found in the study area's vicinities, such as agriculture, cattle raising, and bauxite extraction (Duarte, et al., 2018). The natural vegetation is classified as Submontane Dense Ombrophylous Forest and Lowland Forest, according to IBGE (2012). According to Köppen's classification, the region's climate is "Awi" type, i.e., tropical humid, with an average annual rainfall of 1.800 mm, the average annual temperature of 26.3 °C and relative air humidity of 81% (Alvares, et al., 2013). The main soil types in the region are Yellow Latosols, Yellow Argysols, Plintosols, Gleissols, and Neossols (Rodrigues et al., 2003).

2.2 Data Collection

The methodology adopted was quantitative according Pereira et al. (2018). Ten seed bearers were selected using the following criteria: a) distance of the tree to the edge of the fragment and b) occurrence of regeneration near the seed bearer.

Each seed bearer was georeferenced to obtain the distance to the edge of the fragment. Individuals located within 150 m from the border of the fragment were considered as in the edge to evaluate the natural regeneration of *E. paraensis* in different environments. Individuals situated farther than 150 m from the edge were classified as belonging to the inner fragment (Nascimento & Laurance, 2006). Thus, five seed bearers were located in the edge (distance < 150 m) and five in the inner fragment edge (distance >150 m). The individuals of the natural regeneration were classified into seedlings (height < 300 cm) and saplings (height \geq 300 cm).

Four circular plots were set up in each seed bearer, with 5 m, 10 m, 15 m, and 20 m radii, defined by the crown projection. The center of each tree trunk was used as a reference point to estimate the crown projection area CPA. The CPA of each seed bearer was calculated by measuring the area of eight triangles formed from the center of the plot at 45° angles towards the crown projection border as shown in Figure 1 above. The CPA as well as the distances from the natural regeneration to each seed bearer were calculated by the ArcGis 10.1 software package.

The total sampled area was 1.26 h (0.63 ha in the edge and 0.63 ha in the inner fragment). A metric tape was used to obtain girths, later converted into diameters. The following criteria were considered for measurements: for individuals shorter than 150 cm in height, diameters were measured at the ground level; those with heights above 150 cm, diameters were measured at 1.30 m (DBH - Diameter at Breast Height). Heights were assessed for all sizes of individuals. Direct measurements of heights were carried out in individuals less than 300 cm high; those with heights above 300 cm were measured indirect.

In each plot, individuals of *E. paraensis* were identified and measured in December 2017; March 2018; June 2018, and September 2018. Dead and recruited individuals were also recorded, measured, and mapped in each measurement.

2.3 Data analysis

Absolute density of *E. paraensis* natural regeneration was recorded in each sample unit and converted to a hectare basis, For the diameter distribution we used 1.5 cm class intervals as follows: (1) 0.1-1.4 cm;(2) 1.5-2.9 cm;(3) 3-4.4 cm; (4) 4.5-5.9 cm; (5) 6-7.4 cm; (6) 7.5-8.9 cm; (7) 9-10.4 cm; (8) 10.5-12 cm, and one class for diameter over 12 cm (9).

Analysis of the effect of the environment (edge x inner fragment) and the natural regeneration size (seedlings and saplings) of *E. paraensis* on the Periodic Annual Increments in DBH and height was carried using a completely randomized design in a $2x^2$ factorial arrangement (environment x size of the individual), being each seed bearer a replication of the treatments.

The Periodic Annual Increments in diameter (PAI_{DBH}) and Height (PAI_{HT}) were calculated as the difference between the values of diameter/height at the initial and final measurement period divided by the length of the period (T): ($DBH_{final} - DBH_{initial}/T$) and $HT_{final} - HT_{initial}/T$.

To analyze the PAI_{DBH} and PAI_{HT} of seedlings and saplings at the edge and in the inner fragment, the GLM (Generalized Linear Model) followed by ANOVA (p < 0.05) was applied. Edge and inner fragment and size of regeneration were the factors and PAI_{DBH} , and PAI_{HT} were the dependent variables.

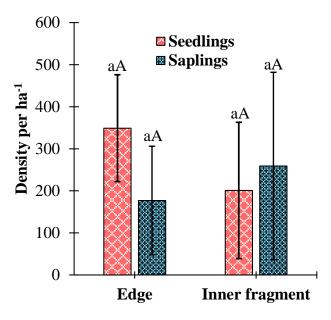
The data were first checked for the assumptions of the analysis of variance (ANOVA) by the following tests: a) normality by the Shapiro-Wilk test (p > 0.05), b) homoscedasticity by the Bartlett test (p > 0.05). Pearson's correlation coefficient between individuals' density and the crown area (m^2) was calculated for all seed bearers. Analysis of Variance was applied to the density of individuals. Then regression analysis was performed to fit equations to the data regarding the distance from the tree crown. The model was selected considering the significance of the parameters' coefficients and the largest coefficient of determination (R^2). Statistical analysis was performed employing the software R by version 3.6.3 (R Development Core Team, 2019), with the p < 0.05 significance level.

3. Results

3.1 Population structure

By the last assessment in September 2018, 525.2 natural regeneration individuals had been recorded in the edge and 420.17 in the inner fragment. There was no statistical difference in the density of individuals concerning the edge or inner fragment (p-value = 0.629) and similarly to the size of individuals (p-value = 0.490) (Figure 2). However, a considerable high density of seedlings occurred in the edge as well as saplings in the inner fragment (Figure 2). In more internal areas of the fragment, a significant reduction and even absence of natural regeneration were observed in one of the seed bearers (Figure 2).

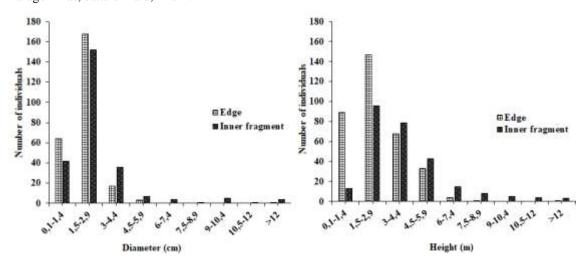
Figure 2. Density of natural regeneration of *Euxylophora paraensis* Huber in a forest fragment in the municipality of Paragominas – Pará, Brazil.



Notes: Averages followed by the same letter do not differ statistically among themselves; lowercase letters represent the comparison between sizes of individuals and capital letters represent the comparison between locations in the fragment by ANOVA and Tukey's post-hoc test at 5% probability. Bars represent the standard deviations1'. Source: Authors (2021).

The individuals with the largest diameters and height were better distributed inside the fragment. Those six centimeters in diameter and nine meters in height did not occur at the edge (Figure 3). The distribution of individuals in diameter and height classes revealed a higher density of individuals in the second diameter class (1.5-2.9 cm) for edge and inner fragment (Figure 3). The two first diameter classes and height together comprise 91.7% and 68.8% of the individuals in the edge and 76.9% and 40.9% in the inner fragment, respectively (Figure 3).

Figure 3. Diameter and height distribution of *Euxylophora paraensis* Huber in a forest fragment in the municipality of Paragominas, state of Pará, Brazil.

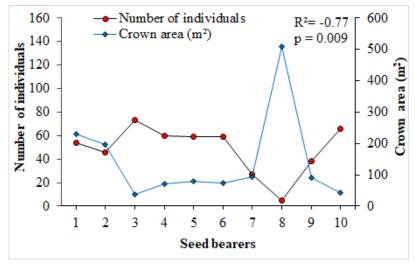


Source: Authors (2021).

3.2 Natural regeneration concerning the seed bearer

The crown areas ranged from 38.2 m² to 509.4 m². The seed bearers' distance to the edge of the fragment varied from 30 m (smallest crown area) to 600 m (largest crown area). A significant negative correlation was found between NR density and crown area ($R^2 = -0.77$; p < 0.009) (Figure 4). The density of NR was small under seed bearers with large crowns, whereas those with small crowns had a higher density of individuals (Figure 4).

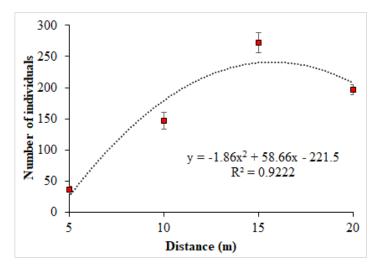
Figure 4. Density of NR under crowns of seed bearers of *Euxylophora paraensis* Huber in relation to the crown area in a forest fragment in the municipality of Paragominas – Pará, Brazil.



Source: Authors (2021).

The quadratic polynomial model provided the best fit to express how the density of natural regeneration per m^2 responds to the distance from the seed bearer (p = 0.001). As shown in Figure 5, the density increases with the distance, reaching the maximum value of 272 individuals at a 15.8 m distance.

Figure 5. Relationship between the density of *Euxylophora paraensis* Huber NR and distance from the seed bearer in a forest fragment in the municipality of Paragominas, state of Pará, Brazil.

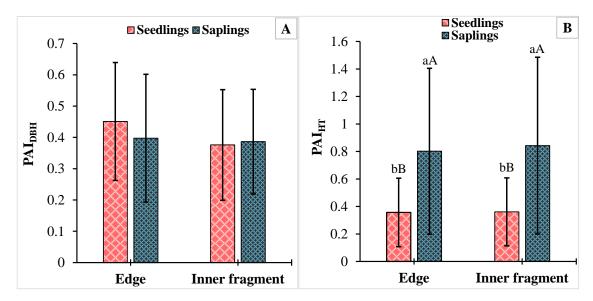


Source: Authors (2021).

3.3 Population dynamics

Fourteen ingrowths were registered in a single seed bearer, and 10 plants (2.3%) died during the measurement period. Periodic annual increment in diameter and height of *E. paraensis* was 0.41 cm year⁻¹ and 0.36 m year⁻¹ for seedlings and 0.39 cm year⁻¹ and 0.82 cm year⁻¹ for saplings, respectively (Figure. 6). No interaction was found between locations (edge or inner fragment) and the size of the NR (seedlings and saplings) for PAI_{DBH} and PAI_{HT} (p-value = 0.065). However, PAI_{DBH} showed a significant difference between the locations in the fragment (p-value = 0.006) (Figure 6A). The diameter increment of seedlings and saplings was higher at the edge of the fragment. PAI_{HT} was significant both for location (p-value = 0.001) and for the size of NR (p-value = 0.001). The higher increments occurred for the NR located both at the edge (0.81 m year⁻¹) and in the inner fragment (0.84 m year⁻¹) (Figure 6B).

Figure 6. Periodic Annual Increment in DBH (PAI_{DBH}) and height (PAI_{HT}) of the natural regeneration of *Euxylophora paraensis* Huber in the municipality of Paragominas – Pará, Brazil.



Notes: Averages followed by the same letter do not differ statistically among themselves, lowercase letters represent the comparison between the sizes of the NR and capital letters, the comparison between locations by ANOVA with Tukey's post-hoc test at 5% probability. The bars represent standard deviations. Source: Authors (2021).

4. Discussion

4.1 Population structure

The NR of *E. paraensis* proved to be sensitive to fragmentation, as seen by the differences in density and growth of individuals between the fragment locations. An increase in NR density in the early stages was observed at the edge of the fragment. Studies have shown that some species are more vulnerable to microenvironments, such as those at the edge of forest fragments, because of their ecological responses to environmental changes. This effect confers some competitive advantage in light requirements to secondary species (Nascimento & Laurance, 2006; Schwartz, et al., 2014; Duarte, et al., 2018). This is the case of *E. paraensis* which fits Budowski's (1965) classification of ecological groups as a light demanding species (Amaral, et al., 2009).

Individuals situated at the fragment border are smaller. High incidence of solar radiation in this environment may induce seedling germination and growth, especially for light-demanding species, which show higher photosynthetic rates than shade-tolerant ones. This difference explains the large abundance of seedlings in the fragment edge (see also Nascimento & Laurance, 2006; Razafindratsima, et al., 2018; Benitez-Malvido, et al., 2018).

The low frequency of saplings (height > 300 cm) in the edge may be explained by the high mortality caused by falling branches, high wind turbulence, and low humidity. These environment features interfere with establishing seedlings in forest fragments (Marques, et al., 2010) and their growth into adulthood (Laurance, et al., 2018; Benítez-Malvido, et al., 2018).

The higher density of saplings in the inner fragment may be explained the natural openings in the forest canopy before the fragment was created. Gaps caused by natural tree mortality are common small-scale disturbances that play an essential role in forest regeneration and dynamics (Whitmore, 1989; Schwartz, et al., 2014). On the other hand, openings created by major disturbances generally cause significant impacts on the forest environment and can modify the behavior of natural regeneration, growth, mortality, and recruitment of individuals (Schwartz, et al., 2014; Dionisio, et al., 2018).

The diameter distribution of *E. paraensis* natural regeneration was irregular, i.e., it did not follow the typical inverse J-curve distribution (Figure 5). At the species level, the diameter distribution may greatly differ from that shape due to the species' ecophysiological responses, and the changes occurred after disturbances over time (Reis, et al., 2014).

Fragmentation and habitat disturbance mainly affect younger individuals who have not yet been established. Instability in the fragments varies according to the nearby anthropic activities (Murcia, 1995; Blumenfeld, et al., 2016). The more intensive the disturbance, the greater the damage to the remaining vegetation (Blumenfeld, et al., 2016).

4.2 Natural regeneration under the seed bearers

The high negative correlation between the crown area and the density of NR of *E. paraensis* individuals revealed the species' need for environments with high solar radiation. The environment under the crown is influenced by its area and architecture, which interferes with the amount of light reaching the forest floor and, consequently, the germination and establishment of natural regeneration (Melo, et al., 2015).

The highest concentration of natural regeneration distant from the parent tree and outside the crown projection corroborates the escape hypothesis postulated by Janzen (1970) and Connel (1971). According to these authors, seeds under the crown or close to the parent tree suffer intense predation, and those far from it would have a better chance of survival. Seeds dispersal, far beyond the seed bearer, may represent high dispersive quality individuals, as they are more likely to survive into adulthood after escaping distance-dependent mortality (Bett & May, 2017).

Seed dispersion of *E. paraensis* is barochoric, but the seeds can also be found beyond the seed bearer, indicating a possible secondary type of dispersion (Melo, et al., 2015).

4.3 Population dynamics of E. paraensis

The short observation period of this study may explain low ingrowth and mortality. Longer-term monitoring periods would be necessary to understand the influence of forest fragmentation on the species natural regeneration.

The periodic annual increment of diameter and height of the natural regeneration varied according to the location in the fragment and size of individuals. The ecological survival strategies of the species in fragmented areas result in the highest diameter and height increments of seedlings and saplings in the edge. The increase in diameter of the smallest individuals is most affected due to the damage caused by the natural fall of tree branches from the upper stratum of the canopy. The rapid height increase of individuals > 300 cm is probably an ecological response of secondary species to the fragmented environment. After the opening of large clearings, individuals grow rapidly to reach the canopy (Schwartz, et al., 2014; Dionisio, et al., 2018).

Light-demanding species such as *E. paraensis* need a gap to reach the upper canopy. Gaps created in the forest canopy favor height growth but later, this growth decreases due to canopy closure by the lateral crown's expansion. *E. paraensis* occupies the upper stratum requiring full sunlight to grow (Brandão, et al., 2018).

Despite the positive response to changes in its natural habitat, it is crucial to evaluate the species' long-term ecological responses. Notwithstanding the increase in density in the fragment's edge, it is necessary to know if these individuals reach adulthood by resisting the fragmentation. Species resistant to fragmentation have typical ecological characteristics, including non-selective pollinators, high reproductive capacity, successful germination, both in fragments and closed forest, tolerance to stress in the seedling stage, resistance to natural enemies, and physical damage.

Forest fragments act as drivers for recovering degraded landscapes. They are shelters for animals and allow movements between fragmented areas promoting the dispersion of fruits, seeds, and keeping soil humidity. As sources of diaspores, they are considered key factors in the vegetal colonization of degraded areas (Rodrigues, et al., 2004; Ribeiro, et al., 2019). Thus, mitigation and conservation of forest fragments associated with management efforts in fragmented tropical forests become necessary to promote actions such as natural regeneration management, enrichment planting, and reforestation of degraded landscapes, as is the case of mining areas.

5. Conclusions

Forest fragmentation has impacted the responses of natural regeneration of *E. paraensis* regarding the density of individuals and their growth dynamics. Seedlings were more abundant in the fragment's edge and showed high diameter and height increments. Saplings were denser in the inner fragment and had improved height increment independent of the fragment environment.

There was variation in the abundance of the species' natural regeneration regarding the canopy area and distance from the seed bearer. Trees with a smaller canopy area had higher abundance of regenerating individuals, and the distance from the mother tree favored an increase in the density of regeneration.

The results demonstrate that the forest fragmentation influences the natural regeneration of *E. paraensis* by favoring seedlings establishment. However, the disturbed environment hinders the development of saplings into the adult stage, therefore, we recommend studies with long-term monitoring of natural regeneration, especially of forest species at risk of extinction.

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