

ENVIRONMENTAL BIOTECHNOLOGY APPLIED TO ARECÁCEAS VIA SUSTAINABLE BIOFERTILIZER, AN ALTERNATIVE TO CARBON SEQUESTRATION



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ABSTRACT

The greenhouse effect causes several environmental problems, such as rising global temperatures, extreme climate change, rising sea levels, melting glaciers, ocean acidification, and loss of biodiversity. In this context, environmental biotechnology applied to arecaceous trees, especially palm trees, emerges as an innovative and sustainable alternative for carbon sequestration. This work aims to evaluate the applications of biofertilizers in arecaceous plants, using environmental biotechnology as a promising strategy to optimize carbon sequestration in this plant family. To achieve this objective, the following methodologies were adopted: literature review, extraction and characterization of specific genes in Arecaceae species, analysis of the species with the highest efficiency in carbon sequestration, monitoring the impact of biofertilizer application on palm trees, evaluation of the effects of biotechnology on plant-environment interactions, monitoring of carbon sequestration in genetically improved palm trees, and monitoring the photosynthetic enhancement and potentiation of young palms through EMGPALM. The

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results showed that the average chlorophyll per species was $2.09 \text{ mg/ml} \pm 0.41$. The average biomass per ton per year was 5.02 ± 0.32 , while the carbon was 2.26 ± 0.29 . The average fresh weight biomass per palm tree was 4.23 kg/m^2 , and the dry weight biomass was 2.58 kg/m^2 . After the application of biofertilizer in six palm species, the annual growth ranged from 40 cm to 90 cm. The number of leaves was from 9 to 12, with an average biomass of $7.51 \pm 0.37 \text{ t/ha}$ and a carbon sequestration of $4.54 \pm 0.4 \text{ t/ha}$. In comparison, the control group, which did not receive the biofertilizer, showed an annual growth between 17 cm and 40 cm, with the number of leaves varying between 7 and 9. The average biomass was $1.52 \pm 0.02 \text{ t/ha}$, and the carbon sequestration was $0.56 \pm 0.1 \text{ t/ha}$. Therefore, environmental biotechnology stands out as a promising, innovative, and sustainable alternative to maximize the role of Arecaceae in carbon sequestration.

Keywords: Enhanced Photosynthesis. Carbon Fixation. Carbon credit.

INTRODUCTION

The impacts of deforestation and fires include the loss of opportunities for sustainable forest use, both for the production of traditional commodities, such as timber and non-timber products, and for capturing the value of forest environmental services. Deforestation also sacrifices the opportunity to take advantage of the benefits of forest ecosystem services. The unsustainable nature of virtually all land uses in deforested areas results in significant long-term losses by preventing forest maintenance (PHILIP, T. K.; ITODO, I. N, 2006).

The fires associated with deforestation affect the amount of gases emitted not only from the biomass that burns, but also from the biomass that does not burn. During a fire, in addition to the release of carbon dioxide (CO₂), gases such as methane (CH₄), carbon monoxide (CO) and nitrous oxide (N₂O) are also released. Biomass that does not burn in the initial phase, with intense flames, will also eventually be oxidized (PHILIP, 2002).

The main cause of drastic and sometimes irreversible climate change is the accumulation of greenhouse gases in the atmosphere. Trees have an exceptional ability to capture and accumulate gases, helping to prevent the planet from overheating (DAVID, 2018).

However, large-scale and uncontrolled deforestation increases the concentration of greenhouse gases contributes significantly to rising temperatures and intensifying climate change. Therefore, it is essential to reduce forest loss and greenhouse gas emissions, as well as focus on restoring forest cover. These actions can be carried out using modern technologies to analyze the situation from different angles and make informed decisions about how to save forests and reduce greenhouse gas emissions globally (SILVA JUNIOR, 2020).

According to Gregory P. Asher (2006), greenhouse gases (GHG) trap and emit infrared radiation, further warming the Earth's atmosphere and surface, which results in the greenhouse effect. The main greenhouse gases are carbon dioxide (CO₂), methane (CH₄), and nitrous oxide (N₂O).

Plants absorb excess carbon dioxide under normal conditions, and when forests are burned or cut down, the accumulated CO₂ is released. Thus, deforestation and the greenhouse effect cause a substantial increase in the concentration of these gases. The main source of other gas emissions is deforestation for the creation of agricultural land and

the additional use of land for agriculture and food production (GREGORY P. ASHER, 2006).

In view of the problem presented, the guiding question is: How can the application of environmental biotechnology optimize the capacity of areaceae in carbon sequestration, offering an innovative and sustainable alternative to face the challenges of climate change?

In this context, growing concerns about climate change have stimulated the search for innovative solutions that can mitigate environmental impacts. Environmental biotechnology emerges as a promising tool, offering new possibilities to face global challenges. This research explores the application of environmental biotechnology in areaceae, through biofertilizers, with a focus on maximizing carbon sequestration as a fundamental strategy to combat greenhouse gas emissions.

Environmental biotechnology, based on advanced techniques such as gene editing and cloning, offers the ability to modify organisms in a precise manner. These tools have the potential to improve crucial characteristics in plants, making them more efficient in carbon absorption, resistant to adverse conditions and contributing to environmental sustainability (FERREIRA et al., 2005; SOARES-FILHO et al., 2004, 2005).

Areaceae, known as palm trees, play a crucial role in carbon sequestration. With their ability to absorb large amounts of CO₂, these plants contribute significantly to the reduction of atmospheric carbon levels. The application of environmental biotechnology aims to enhance this natural capacity, allowing areaceae to play an even more prominent role in climate change mitigation (SU et al., 2020).

The application of environmental biotechnology in areaceae can be directed to increase efficiency in carbon absorption, ensure pest resistance, and adapt to different climatic conditions. By genetically enhancing the areaceae, we can create more robust organisms, capable of facing emerging environmental challenges (SOARES-FILHO et al., 2004, 2005).

The benefits resulting from the application of environmental biotechnology in areaceae by means of biofertilizers are diverse. In addition to contributing to the reduction of carbon emissions, this approach can generate sustainable economic opportunities, such as the production of biomass and products derived from genetically enhanced palm trees.

Environmental biotechnology applied to areaceae, such as palm trees and other plants of this family, emerges as an innovative and sustainable alternative in the context of carbon sequestration. The strategic application of biotechnological techniques to this group

of plants has great potential to mitigate climate change and promote environmental sustainability.

Areaceae play a crucial role in absorbing carbon dioxide (CO₂) from the atmosphere, thus contributing to the global climate balance. However, factors such as deforestation, soil degradation and climate change threaten the ability of these plants to effectively fulfill this vital function Carvalho et al., (2010). This is where environmental biotechnology, through biofertilizers, comes in as a promising tool.

Through genetic manipulation and other advanced techniques, scientists can enhance the characteristics of areaceae to optimize their carbon sequestration potential. This can include modifying genes related to the rate of photosynthesis, increasing water use efficiency, and resistance to harsh environmental conditions. By enhancing these characteristics, environmental biotechnology creates more robust and adaptable palm trees, capable of playing an even more vital role in reducing CO₂ concentrations in the atmosphere (FERRERIRA et al., 1997).

In addition, research into environmental biotechnology through biofertilizers paves the way for the development of areaceae varieties that can be grown more efficiently in different environments, contributing to the restoration of degraded ecosystems and promoting biological diversity. This approach not only strengthens the carbon sequestration capacity of these plants but also drives conservation and environmental restoration efforts.

In summary, the application of environmental biotechnology in plants of the Arecaceae family, through the use of biofertilizers, represents an innovative and promising strategy to face the challenges of carbon sequestration. By enhancing the photosynthesis of these plants, it is expected to strengthen their natural capacity to absorb carbon, contributing significantly to climate change mitigation. This approach exemplifies the intersection between scientific advances and sustainable practices, highlighting the essential role of biotechnology in building a more resilient and ecologically balanced future. Thus, the research aims to evaluate the use of biofertilizers in Arecaceae, using environmental biotechnology as an effective alternative to optimize carbon sequestration in this plant family.

MATERIALS AND METHODS

It is a field and experimental research that allowed a strict control of experimental conditions, facilitating the validation of hypotheses and obtaining accurate data. According

to Fonseca (2002, p. 11-12), experimental research is characterized by the direct manipulation of the variables related to the object of study and aims to test the hypotheses formulated by the researcher.

THE IMPORTANCE OF FIELD RESEARCH AS A SUBSIDY FOR EXPERIMENTAL RESEARCH

According to Fonseca (2002), field research plays a crucial role in the development of scientific investigations, as it allows the collection of data directly from the environment or the reality studied. This type of research provides empirical and detailed information that enriches the foundations of experimental research, enabling the formulation of more accurate and contextualized hypotheses.

In addition, the interaction with the object of study in its natural context facilitates the identification of relevant variables, promotes the understanding of complex phenomena and expands the possibilities of experimentation.

The research was carried out in three different locations: in a field area at Rancho Água Fria, where an experimental site was created; at the Mahogany Farm, where the palm trees were analyzed in four quadrants, both located in the municipality of Ananás, state of Tocantins, and in the laboratories of the State University of the Tocantina Region of Maranhão – UEMASUL, for the analysis of the collected materials.

The analysis of the efficiency in carbon sequestration in Arecaceae (palm) species adopted a multidisciplinary approach, which included field observations, counting the palms in four quadrants of 25 x 100 m, material collection and laboratory analyses.

Data were collected on the species present, recording information such as palm size, DBH (diameter at breast height), above-ground biomass and population density. The instruments used included a tape measure, GPS, camera, digital scale and electric oven. To measure the biomass of palm trees and calculate the carbon stock per hectare/year in different species, the methods described by Arevalo (2002) and Oliveira (2007) were applied, according to the formula.

$$BA = 0.1184 \times DBH^2 \times 0.02 \text{ t/ha.}$$

Where: **BA = Biomass**, **0.1184** = constant, **DBH** = Diameter of breast height (cm), **0.02** = conversion factor for quadrants (plot of 25m x 100m) and **t/ha** = ton per hectare year.

Preliminary studies were also carried out on the photosynthetic efficiency of palm trees, according to Golding and Smith (2007), with the objective of evaluating the capacity of the leaves to capture carbon dioxide and convert it into biomass. Leaf length, fresh weight of biomass and dry weight of biomass were considered (see Figure 6). It has been observed that these traits can influence carbon sequestration in different palm species.

The data collected were compared between different species of Arecaceae to identify those with the greatest efficiency in carbon sequestration, considering the costs associated with the cultivation and maintenance of the various palm species compared to the benefits generated by carbon sequestration

EXTRACTION AND CHARACTERIZATION OF SPECIFIC GENES IN ARECACEOUS (PALM) SPECIES ASSOCIATED WITH CARBON SEQUESTRATION

The choice of palm species of the Arecaceae family for the study included: açai (*Euterpe oleracea*), buriti (*Mauritia flexuosa*), buritirana (*Mauritiella aculeata*), babassu (*Attalea speciosa* Mart.), bacaba (*Oenocarpus bacaba*), coco-da-praia (*Cocos nucifera* L.), coco-najá (*Maximiliana maripa*), macaúba (*Acrocomia aculeata*), piassava (*Attalea funifera* Martius) and P. trepadeira (*Desmouçus sp.*). The selection of these species took into account factors such as their geographic distribution, carbon sequestration capacity, and sample availability.

Samples were collected from young and adult palm leaves at different stages of development. For DNA extraction, the methods described by Thames and Kosmos (2023), in the *Genetics & DNA Experiment*, and by Costa and Moura (2001), in the *DNA Extraction Manual: Embrapa*, were used. Small fragments of leaves from different palm trees were analyzed to investigate gene expression and identify genes involved in carbon sequestration.

Microscopy and staining techniques with methylene blue and lugol were applied to identify the morphological and structural characteristics of the chloroplasts, where photosynthesis occurs, and to verify the chloroplastic DNA. The objective was to visualize the genetic information essential for the synthesis of proteins involved in photosynthesis, highlighting the direct relationship between the genetic material and the plant's ability to perform photosynthesis and carbon sequestration.

Bioinformatics tools such as ChloroSeq and Geneious were used, which offer a variety of resources for the analysis of genomic sequences, including assembly, annotation, and comparison.

To perform the statistical tests and evaluate the significance of the genetic differences observed, Analysis of Variance (ANOVA) was applied to verify the variability between the groups.

BIOFERTILIZER PRODUCTION

The biofertilizer production process involved the use of banana peels, bean peels, corn husks, buriti fruit peels, citrus fruit peels, cashew nut kernel shells, old wood ashes, eggshells, beach coconut mesocarp powder, cow manure, avocado peel, papaya peel and composting of litter decomposition, that have been turned into dust.

The peels were purchased at the Central Market of Imperatriz, Maranhão; at Rancho Água Fria, Tocantins; and at the Mahogany Farm. To process the material, scissors, a knife, an aluminum basin, an electric oven, a digital scale and a mill were used. The material was cut into small pieces, taken to the oven for drying at 80°C and then crushed in the mill until it was turned into powder. After this process, the composition of the biofertilizer was carried out, using weighing on a digital scale. For each 500 g of material, it was diluted in one liter of water and applied to the palm trees, according to the methodology of (MEDEIROS 2002).

Borges (2021) states that banana peels, beans, corn, and cow manure contain a variety of nutrients useful for soil fertilizers. Banana peel is an excellent source of potassium, an essential macronutrient for plant growth. Potassium plays a vital role in regulating water and nutrient absorption, root development, and plant resistance to stress. In addition, banana peels also contain phosphorus, another important nutrient for healthy plant growth.

Beans are an excellent source of nitrogen, one of the main micronutrients needed for plant growth. Nitrogen is a vital component of proteins, enzymes, and chlorophyll, playing a key role in photosynthesis and overall plant growth. Just like banana peels, beans also contain phosphorus in significant amounts.

Corn is also a good source of nitrogen, providing an additional boost of this important nutrient for plant growth. In addition, corn contains potassium, which is essential for various metabolic functions of plants.

Cow dung is a rich source of organic nitrogen, slowly released into the soil as it decomposes, providing a steady supply of nitrogen to plants over time. It also contains phosphorus in significant amounts, helping with root development and overall plant health. In addition, cow manure contains potassium, completing the triad of macronutrients essential for plant growth (MEDEIROS, 2002).

The remaining nutrients, such as eggshells, dried leaf remains, citrus peels, avocado peels, papaya peels, and coconut waste from the beach, were used as ingredients to balance the proportions and provide a full range of nutrients necessary for healthy plant growth. In addition, calcium, magnesium and other micronutrients were found, which were considered when formulating this organic matter.

BIOFERTILIZER EFFICACY TEST

To test the efficacy of the biofertilizer produced, six species of palm trees (4 individuals per species) were used as the object of study, each one being identified according to its species. This stage was carried out in an experimental area at Rancho Água Fria, municipality of Ananás, state of Tocantins.

To ensure the validity of the results and minimize biases, a control group was established, consisting of 6 species and 20 individuals, which did not receive the application of biofertilizer, and an experimental group, also with 6 species and 20 individuals, which received the application of biofertilizer.

After the application of the biofertilizer, data collection and monitoring of the physiological performance of the palm trees began. Parameters such as photosynthesis rate, transpiration and water use efficiency were analyzed, as well as growth measurements, including height, trunk diameter and number of leaves. The total biomass was evaluated based on fresh and dry leaves, and the carbon sequestration capacity was determined by techniques such as total carbon analysis and quantification of the carbon content in the leaves, according to the methodologies of (AREVALO, 2002 and OLIVEIRA, 2007).

In the environmental monitoring, relevant data such as temperature, humidity and light intensity were recorded, in addition to the diameter (in cm), height (in m) and density of the palm trees, to better understand the conditions that can influence the results. These data were recorded in cards and later transformed into tables and graphs.

For the statistical analyses, appropriate methods were used, such as analysis of variance (ANOVA) and Tukey's test for multiple comparisons.

STRUCTURE TO ENHANCE PHOTOSYNTHESIS IN YOUNG PALMS (EMGPALM)

To improve photosynthesis in young palm trees, the EMGPALM (Structure for Group Improvement of Young Palm Trees) was built. The structure consists of an acrylic chamber 50 cm long, 40 cm high and 8 cm wide, as well as a transparent acrylic plate. The chamber was divided into three modules: two for experimentation and one for control.

Two carbon capture batteries were installed to measure the carbon uptake by palm trees, both in the experimental modules and in the control module. Two aquariums were used to induce CO₂ and liquid chlorophyll, promoting growth and allowing the analysis of palm trees. In addition, CO₂ and chlorophyll cylinders were employed to manipulate and adjust the environmental conditions inside the chamber, simulating different scenarios of CO₂ exposure and nutrient supply.

For the experimentation, two acrylic tanks were attached to EMGPALM, with the following dimensions: 15 cm high, 16.5 cm wide and 13 cm long. Each tank is equipped with a luminaire and an LED biovolt pump. These tanks are used to receive diluted CO₂, produced from biological materials, which is applied in the process of enhancing the photosynthesis of palm trees.

The first tank was equipped with two CO₂ generator kits and a Vigoar A300 submersible aquarium pump, with a capacity of 450 L/h, intended for tanks up to 100 L and 110V. The second tank was equipped with a CO₂ generator kit and a Vigoar A300 submersible aquarium pump, also with a capacity of 450 L/h, for tanks up to 100 L and 110V.

MONITORING OF PALM TREES

The activity was conducted using specimens of the palm species (leaves) that are part of the study, both those that received biofertilizer and those that did not, ensuring that they were representative of the selected environment. Palm growth was monitored over 12 months, with the quantification of parameters such as height, diameter, circumference, number of leaves, amount of chlorophyll, biomass volume and carbon sequestration.

The materials used included drone, GPS, machete, palm leaves, digital scale to weigh the collected samples, laboratory equipment, reagents (salt, distilled water, ethanol,

neutral detergent), camera, magnifying glass, microscope, tape measure and field notebook. During the monitoring, it was possible to quantify the amount of carbon sequestered by analyzing the fresh and dry biomass of each species.

QUANTIFICATION OF CARBON IN THE SAMPLES

To analyze the amount of carbon in the samples, the methods described by Arevalo (2002) and Oliveira (2007) were used, which quantified the total organic carbon (TOC). Carbon sequestration was calculated by determining the amount of carbon per unit of biomass and per area occupied by palm trees. Then, using EMGPALM, the biofertilizer was applied to the young palms and to the palms of the experimental nursery, using a specific form to identify the parameters (palm, height, diameter, chlorophyll, number of chloroplasts, fresh biomass, dry biomass, dilute biomass and carbon sequestration. For each palm tree, 100 grams of nutrient dissolved in one liter of water were applied, and each individual received two liters of solution with the biofertilizer over the course of a month.

During this process, the four types of biofertilizers produced were analyzed. Palm trees that received the refined biological fertilizer showed a faster increase in chlorophyll, that is, they demonstrated higher biomass in a shorter period. The other biofertilizers (granulates) showed a more gradual effect, but were also effective.

The variables were controlled to avoid drastic changes in the results, including growing conditions, plant age, chlorophyll quantification, biomass expression, carbon sequestration, and other environmental factors. The experiments were repeated eight times to obtain more reliable results and reduce experimental variability, according to the methods described by Quintanilha et al. (2023).

RESULTS AND DISCUSSION

The role of palm trees in ecosystems may be significantly more relevant than estimates made to date suggest. A comprehensive survey in several databases, gathering information collected by 223 researchers from different countries, revealed that forest biomass measurements can vary by up to 15% more or less (KAHN, F. and MEJIA, K., 2020).

ANALYSIS OF ARECÁCEOUS (PALM) SPECIES WITH GREATER EFFICIENCY IN CARBON SEQUESTRATION.

The concept of carbon sequestration was formulated at the Kyoto Conference in 1997, an event that brought together authorities from more than 160 countries. During this conference, an international treaty was created establishing targets for the reduction of greenhouse gas emissions and encouraging the development of sustainable technologies to convert and reverse the accumulation of atmospheric CO₂, with the aim of mitigating the greenhouse effect (HOUGHTON, R. A., 1994).

Based on this assumption, the amount of biomass made available by the palm trees in four quadrants of 25 x 100 meters was analyzed, totaling 109 palm trees belonging to eight different species. The mean diameter of palm trees at breast height (DBH) was 34.96 cm. The total biomass per palm tree, in tons per hectare (BTP/t/ha), was estimated at 3.37 t/ha, while the biomass carbon per palm tree, in tons per hectare (CBP/t/ha), was 1.6 t/ha.

According to Houghton (1994), the diameter of the stem is directly related to the leaf surface area available for photosynthesis. A larger leaf area generally implies a greater photosynthetic capacity, the process by which plants convert sunlight into energy. In addition, the stem of palm trees can act as a nutrient storage site. A larger diameter suggests a greater capacity to store resources, which favors the healthy growth and development of the plant.

To quantify the biomass, the palms with the highest number of individuals were selected. This method allowed the determination of fresh biomass per square meter, dry biomass per square meter, fresh biomass per dilute gram and biomass per dry weight of diluted grass, as shown in Table 1.

Table 2: Indicates four possibilities of the amount of biomass per palm tree, with the first two parameters indicating green leaves and dry leaves; the second, wet ashes and dry ashes Source of information Luís Gustavo Neres Ferreira Soares.

Palm trees DAP	Biomass fresh weight kg/m²	Biomass dry weight kg/m²	Biomass Fresh Weight per Gram/Dilute (g)	Biomass dry weight per gram/dilute (g)
Babassu	10,1	5,5	198	190
Moriche palm	5,7	2,1	119	110
Coca Najá	9,2	4,1	118	109
Acai	3,7	1,2	62	58
Tucum	3,6	1,4	52	43
Buritirana	3,1	1,2	15	8

The relationship between the amount of biomass and the amount of carbon sequestered is essential to understand the role of plants in capturing and storing atmospheric carbon. In general, the biomass of a plant is predominantly composed of carbon, which is a fundamental component of the organic compounds produced during photosynthesis. Thus, the increase in biomass in palm trees is directly linked to the increase in the amount of carbon sequestered, contributing to the reduction of CO₂ levels in the atmosphere and playing a crucial role in the global carbon cycle.

ANALYSES OF THE AMOUNT OF CHLOROPHYLL PER PALM TREE

Palm trees play a crucial role in environmental balance, contributing significantly to carbon sequestration. This ability makes them essential elements in the fight against climate change, as they help to substantially reduce CO₂ levels in the atmosphere. This process is favored by the uniform structure of the palm canopy, which optimizes carbon uptake during photosynthesis and promotes efficient chlorophyll accumulation, (KAHN AND MEJIA, 2020).

Each of these species has distinct genetic characteristics, which suggests that the ability to sequester carbon may vary between them. Identifying specific genes in this context is essential to understand and optimize the carbon sequestration potential of palm trees. Table 2 illustrates the amount of chlorophyll extracted from 200 grams of crushed leaves in different palm species, comparing areas of dry soil and flooded areas.

Palm trees	Scientific name	>Mv	>Mv	Average	mg/ml
Acai	<i>Euterpe oleracea</i>	0.08	0.05	0.13	2.3± 0.4
Moriche palm	<i>Mauritia flexuosa</i>	0.09	0.05	0.14	2.2±0.5
Buritirana	<i>Mauritiella aculeata</i>	0.07	0.04	0.11	2.1±0.4
Babassu	<i>Attalea speciosa</i> Mart.	0.08	0.05	0.13	2.3±0.4
Bacaba	<i>Oenocarpus bacaba</i>	0.08	0.05	0.13	2.2±0.5
Beach Coconut	<i>Cocos nucifera</i> L.	0.06	0.02	0.08	1.2±0.1
Coco najá	<i>Maximiliana Maripa</i>	0.04	0.01	0.05	2.6±0.6
Macaúba	<i>Acrocomia aculeata</i>	0.04	0.02	0.06	2.0±0.5
Piassava	<i>Attalea funifera</i> Martius	0.05	0.04	0.09	1.0±0.3

* Chlorophyll collection results analyzed in eight experiments from each palm tree.

>Mv = Highest value collected. > Mv = Lowest amount collected.

Mg/ml = Milligram per milliliter

± Variation.

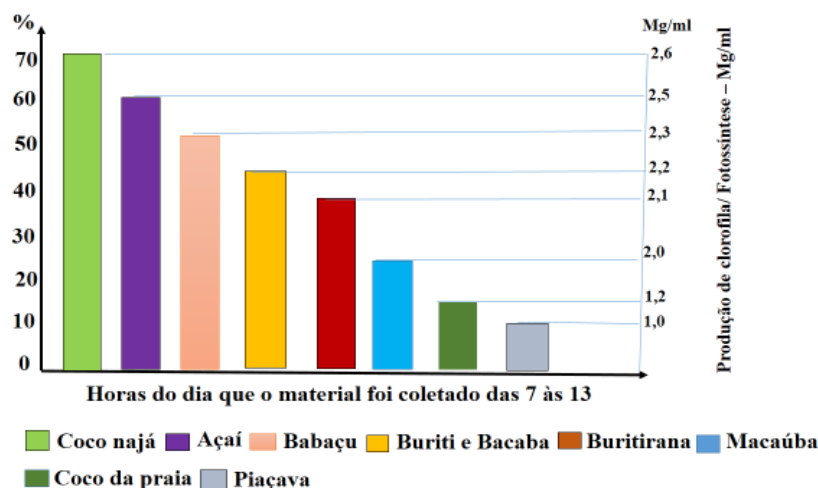
Source: Author.

Genes play a key role in how efficiently palm trees carry out photosynthesis, the process by which they convert carbon dioxide (CO₂) and water into glucose and oxygen,

using solar energy. Genetic variation can influence the rate and efficiency of photosynthesis in different plant species, directly affecting the amount of carbon they are able to capture from the atmosphere (MESQUITA et al., 2001).

Recent research has identified specific genes in several palm species, directly related to carbon sequestration. These genes are involved in processes such as carbon fixation, metabolite transport, and the regulation of photosynthesis. Graph 1 illustrates the performance of palm trees in photosynthesis (carbon sequestration), based on chlorophyll extraction in seven experiments carried out. The collection of the material took place between 7 am and 1 pm.

Graph 1: Photosynthetic performance of 9 palm species, 4 of which are wetlands and 5 are arid zones.



Source: Luís Gustavo Neres Ferreira Soares.

The green column in the graph represents the photosynthetic performance of the najá coconut palm, a species adapted to dry areas, while the lilac column depicts the açaí palm, which grows in wetlands and flooded areas. Both species had the highest amount of chlorophyll per milliliter, with the average of the seven experiments carried out between 7 am and 1 pm. The najá coconut palm obtained the best results (2.6 mg/ml – 70%) due to its uniform crown, long straws, with more than 2 meters, and, on average, 12 leaves per stem.

When analyzing the graph, it is observed that the other palm trees presented satisfactory results, with the piassava palm being the one that obtained the lowest results in all experiments (1.0 mg/ml – 20%). This result is related to the average size of the palm tree (2m), which receives shade from the other trees, especially in the morning. According to Goulding & Smith (2007), the presence of shade can affect the ability of trees to perform

photosynthesis efficiently, as the reduction in the intensity of the available light can lead to a decrease in chlorophyll production and, consequently, in the efficiency of photosynthesis.

The palm trees that presented non-uniform results (oscillations) were those located in flooded areas or close to flooded regions (coco najá, açai, buriti, buritirana and bacaba). The curvature in chlorophyll analyses can depend on several factors, such as the specific species of palm tree, soil conditions, climate, among others. Chlorophyll, a substance responsible for photosynthesis in plants, can have its production influenced by several environmental factors.

In wetter zones, plants may face more intense competition for sunlight due to the lush growth of surrounding vegetation. This can lead plants, including palm trees, to develop strategies to optimize light absorption. One of these strategies may be the curvature of the leaves, which allows a more efficient exposure to sunlight, favoring the production of chlorophyll and, consequently, increasing the efficiency of photosynthesis (GOULDING & SMITH, 2007).

FREQUENCY OF GENES RESPONSIBLE FOR CARBON SEQUESTRATION IN PALM TREES

In view of the results obtained, the frequencies of genes responsible for carbon sequestration in six palm species that had the highest incidence in the studied area (buriti, açai, buritirana, coco-najá, babassu and bacaba) were analyzed. In the microscopic analyses, it was observed that the palm trees analyzed belong to the C4 group. C4 plants have special adaptations in the photosynthesis process, optimizing efficiency in hot and dry environments.

The term "C4" refers to the fact that these plants utilize a four-carbon cycle for initial carbon fixation during photosynthesis, in contrast to C3 plants, which utilize a three-carbon cycle. This adaptation helps C4 plants to overcome photoinhibition in conditions of high luminosity and high temperatures (CARVALHO et al., 2010).

When analyzing the collected material (chlorophyll) using an electron microscope with a 100x/1.25 objective, the presence of the enzyme considered more efficient in carbon fixation, PEP carboxylase, was observed in all palm trees. PEP carboxylase (phosphoenolpyruvate carboxylase) is a crucial enzyme in photosynthetic metabolism known as the crassulaceous acid cycle, or simply C4 metabolism.

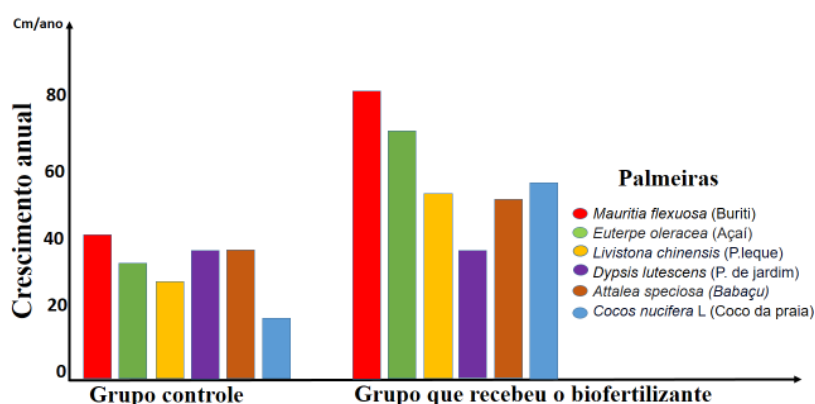
C4 metabolism allows palm trees to optimize carbon fixation under high temperatures and bright light conditions, reducing water loss through transpiration and increasing photosynthetic efficiency compared to C3 plants in certain environments. PEP carboxylase works in conjunction with other enzymes in specific cells called mesophyll cells to fix CO₂ in oxaloacetic acid molecules. These molecules are transported to cells in the vascular bundle, where CO₂ is released and utilized by RuBisCO (enzyme ribulose-1,5-bisphosphate carboxylase/oxygenase) in the Calvin-Benson cycle.

THE IMPACT OF BIOLOGICAL FERTILIZER APPLICATION ON PALM TREES

The use of biofertilizers on palm trees has significant benefits for increasing carbon sequestration. These fertilizers provide essential nutrients to plants, such as nitrogen, phosphorus, and potassium, which, when assimilated, promote healthier growth of palm trees. This development favors the increase of biomass and, consequently, the capacity for carbon sequestration.

After the application of the biofertilizer, the performance of the palm trees in terms of growth, increase in diameter, number of leaves, accumulated biomass and carbon sequestration was analyzed. Comparatively, the control group showed slower growth compared to palm trees that received biofertilizer twice a month (DBFT/month) and irrigation with water three times a week (APS/L). The results of these analyses are presented in Graphs 02 and 03.

Graph 02: Comparison of the annual growth (in centimeters) of the palm trees between the control group, which did not receive biofertilizer, and the group treated with regular doses of biofertilizer.



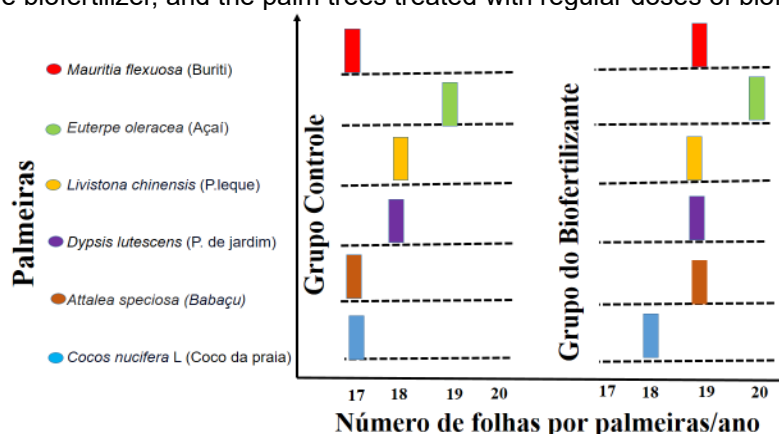
Source: Luís Gustavo Neres Ferreira Soares.

Analysis of the significant difference in palm growth between the two groups is essential. The group treated with biofertilizer showed a higher growth than the control

group, indicating that the biofertilizer had a positive impact on plant development. Interpreting the graphs provides valuable insights into the potential of biofertilizer to promote palm tree growth, with implications ranging from improvements in agriculture to strategies to promote environmental sustainability. However, it is important to interpret the results with caution, considering the validity of the study and the need for further experiments to validate the findings.

Graph 03 illustrates the number of leaves in each group. The control group maintained an average of 17 to 19 leaves per palm tree over the course of a year, while the group treated with biofertilizer showed an increase, recording between 18 and 20 leaves per palm tree. These results suggest that the application of biofertilizer had a positive effect on palm leaf production.

Graph 03: Comparison of the number of leaves over a year between the palm trees in the control group, which did not receive biofertilizer, and the palm trees treated with regular doses of biofertilizer.



Source: Luís Gustavo Neres Ferreira Soares.

The growth rates were based on the results followed over the course of a year, considering three main factors: the dosage of biofertilizer, the amount of water applied to each palm tree, and the differences between the species. The control group had an average number of leaves per palm tree between 17 and 19 over a one-year period, while the experimental group, which received biofertilizer, had an average number of between 18 and 20 leaves per palm tree. This difference suggests that the biofertilizer had a positive effect on the leaf cells of palm trees, resulting in an increase in the number of leaves.

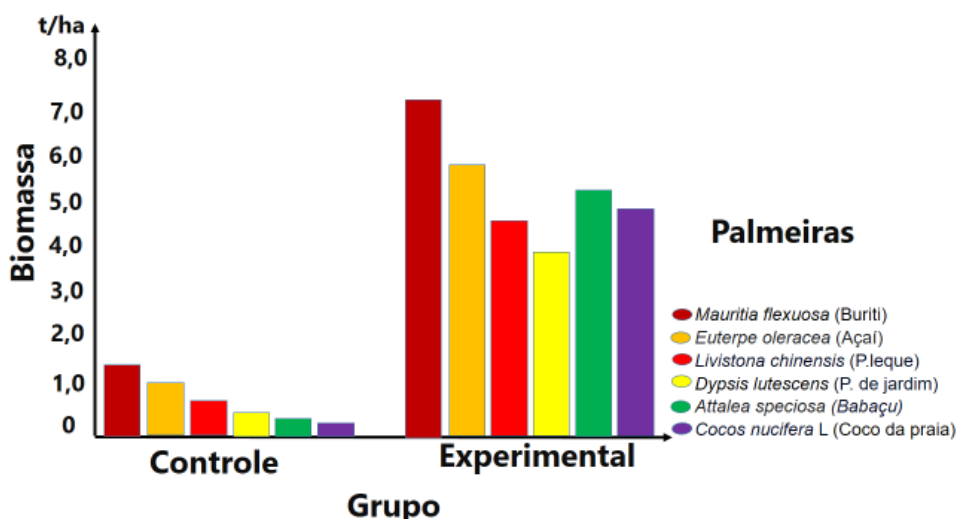
However, it is important to consider that there is natural variation in the number of leaves between different species of palm trees. Therefore, the difference observed between the two groups can be attributed both to the effect of the biofertilizer and to the

inherent variation between the species and the natural habitat of each palm tree. In addition, it has been observed that young palms tend to grow faster than older ones. Ideal growing conditions, such as fertile soil (with biofertilizer), proper irrigation, and protection from strong winds, significantly increase the growth rate. In contrast, the palm trees in the control group, which did not receive adequate irrigation or biofertilizer, showed slower growth, yellowing leaves, and low chlorophyll content.

Biofertilizers are products derived from organic sources and microorganisms that improve soil fertility and plant health. They contribute to the growth of palm trees and the increase of their biomass in several ways. They improve soil health, increase the availability of essential nutrients, and stimulate plant growth through natural mechanisms. Biofertilizers play a crucial role in the sustainable development of agriculture, including palm cultivation, resulting in healthier plants, with better growth and higher biomass production (CARVALHO et al., 2010).

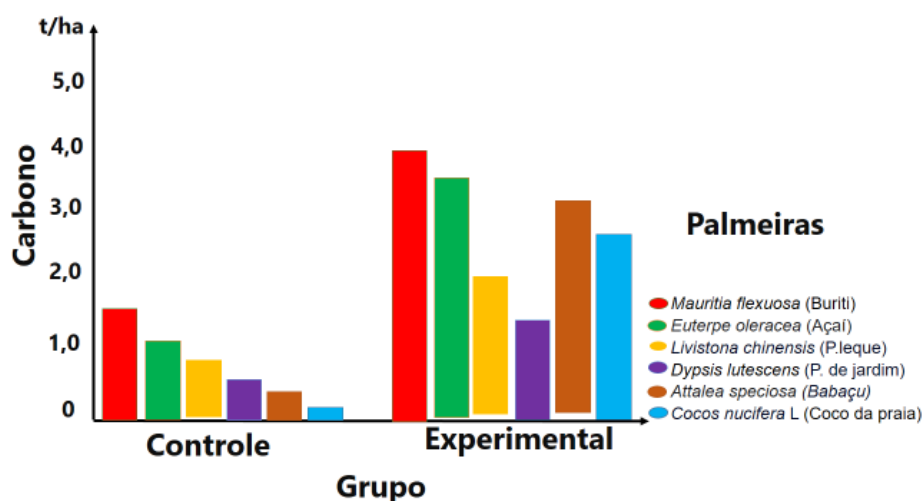
Graphs 04 and 05 show positive results for a group of six palm species, which showed an increase in biomass production and carbon sequestration after receiving biofertilizer and irrigation three times a week. These results were obtained in a controlled experimental study, comparing with a group that did not receive these same variables.

Graph 04: Shows the number of tons of biomass per hectare per year, comparing the palm trees in the control group, which did not receive biofertilizer, with those in the experimental group, which received doses of biofertilizer.



Source: Luís Gustavo Neres Ferreira Soares.

Graph 05: Shows the number of tons of carbon per hectare per year, comparing the palm trees in the control group, which did not receive biofertilizer, with those in the experimental group, which received doses of biofertilizer.



Source: Luís Gustavo Neres Ferreira Soares.

The results of these graphs clearly demonstrate that biofertilizer application and regular irrigation had a positive impact on biomass production and carbon sequestration in palm trees. According to Borges (2021), these results are consistent with the existing literature, which highlights the benefits of using biofertilizers and efficient irrigation practices in agriculture and forestry.

One of the most significant observations was the substantial increase in palm biomass in the experimental group compared to the control group. The application of biofertilizer provided essential nutrients in a more bioavailable way, promoting more vigorous plant growth. In addition, regular irrigation ensured an adequate water supply, optimizing the physiological processes of palm trees and maximizing the growth rate.

Another notable aspect was the increase in carbon sequestration in the palm trees of the experimental group. Biofertilizer and regular irrigation not only stimulated palm growth, but also increased their ability to capture and store atmospheric carbon. This is a highly desirable outcome, considering the crucial role carbon sequestration plays in mitigating climate change.

Importantly, the observed results can be attributed not only to the application of biofertilizer and water, but also to the complex interplay between various factors, such as soil type, climatic conditions, and the genetic characteristics of the palm trees. However, this study provided solid evidence that the use of these sustainable agricultural practices can play a significant role in increasing plant productivity and reducing environmental impact.

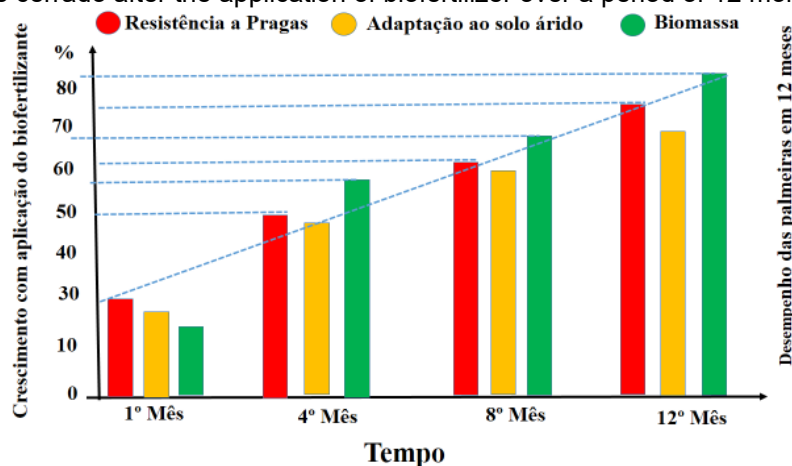
Despite the promising results, it is important to recognize that there are still issues to be addressed. For example, it is necessary to investigate the long-term effects of the continuous application of biofertilizers and water on soil health and local biodiversity (cerrado). In addition, economic and logistical aspects must be considered in the implementation of these practices on a commercial agricultural scale.

Thus, the results show the potential of sustainable agricultural practices, such as the use of biofertilizers and efficient irrigation, to promote healthy palm growth and mitigate carbon emissions. These results provide a solid basis for future research and for the implementation of environmentally responsible management strategies in agricultural and forest ecosystems.

ANALYSIS OF THE BIOTECHNOLOGICAL EFFECTS OF BIOFERTILIZER ON PLANT-ENVIRONMENT INTERACTIONS, CONSIDERING FACTORS SUCH AS RESISTANCE TO ENVIRONMENTAL STRESSES, ADAPTATION TO DIFFERENT CLIMATIC CONDITIONS, AND INTERACTIONS WITH SOIL MICROORGANISMS

The use of biotechnology, through the application of biofertilizers (which provide macronutrients), has made it possible for palm species to become more resistant to pests, diseases, and adverse environmental conditions. This contributed to the increase in the number of leaves. The palm cells showed a notable increase in chlorophyll concentration, resulting in biomass growth and greater efficiency in carbon sequestration. In addition, genetic manipulation, facilitated by the controlled application of biofertilizer, improved the nutritional composition of palm trees, making them more nutritious and adapted to different soil and climate conditions in the cerrado region (see Graph 06).

Graph 06: Highlights the growth of palm tree biomass, its resistance to pests and adaptation to the arid soil of the cerrado after the application of biofertilizer over a period of 12 months.



Source: Luís Gustavo Neres Ferreira Soares.

Biotechnological methodologies can give plants greater resilience to abiotic stresses, such as water deficit, edaphic salinization, and thermal oscillations, which are determining factors for agricultural sustainability in different ecosystems (Cirino, 2021). In this way, genetic engineering can also be used to enhance biomass in Arecáceas, enabling its conversion into biofuels and promoting renewable energy sources.

The biotechnological applications of the biofertilizer developed in this study enable the selected palm trees to adapt better to adverse environmental conditions, such as drought, substrate salinity and thermal variations. This effect is evidenced by the increase in the greenish hue of the leaves, indicative of a higher concentration of chlorophyll, which intensifies carbon fixation and improves the fertility of the soil in which these species are inserted.

The administration of the biological input resulted in the structural optimization of the chloroplasts, making them more tolerant to environmental stresses. For example, genes associated with drought resistance, from species adapted to arid biomes, have been incorporated into crops susceptible to water deficit. Among the palm trees evaluated, the babassu (*Attalea speciosa*), the coconut-najá (*Acrocomia intumescens*), the tucum (*Astrocaryum vulgare*) and the macaúba (*Acrocomia aculeata*) stood out as the most adaptable, species that demonstrated greater aptitude to the acidic soil characteristic of the Cerrado of Maranhão and Tocantins.

This strategy provided innovative tools to increase the tolerance of the Arecácea to adverse environmental factors, promoting agroecological sustainability and boosting the resilience of these species in different biomes.

EFFECT OF IMPROVED PHOTOSYNTHESIS RATE ON YOUNG PALMS (EMGPALM)

Increasing the concentration of CO₂ can improve the rate of photosynthesis, resulting in faster growth of palm trees. Biofertilizers, in turn, have shown significant benefits, such as improving the resistance of palm trees to diseases and pests, as well as increasing the overall health of the plants. CO₂ contributes to the greater robustness of palm trees, making them more resistant to environmental stresses.

A nutritional balance was observed between the palm trees of modules two and three, which showed higher growth than the palm trees of module one, with an additional height of 3 cm and five leaves, two more than the palm trees of the control module. The use of biofertilizers and CO₂ was carried out in a balanced way to avoid excess nutrients or CO₂, preventing possible damage to the palm trees in the experimental module.

The costs of production and application of biofertilizers and CO₂ were evaluated to ensure economic viability and long-term sustainability. The use of sodium bicarbonate, citric acid and biological yeast to generate CO₂ was done in a controlled manner, to avoid negative environmental impacts, which was confirmed by the capture of diluted carbon in the EMGPALM batteries.

The use of biofertilizers and additional CO₂ can bring many benefits to the genetic improvement of young palms, promoting faster growth, greater resistance to diseases and better soil quality. After one year of experiment, the palm trees showed a growth of up to 2 meters more compared to the palm trees in the control group.

MONITORING CARBON SEQUESTRATION IN PALM TREES THAT RECEIVED BIOFERTILIZER DOSES, COMPARED TO THOSE THAT DID NOT (CONTROL)

Throughout the year, the species selected to receive the biofertilizer and those that were not treated were monitored. It was found that the palm trees submitted to the application of the biological input presented a dark green color, indicating a higher concentration of chlorophyll, which suggests an increase in the density of chloroplasts per cell.

During the monitoring, the palm trees treated with the biofertilizer recorded an average monthly growth of 7.5 cm and an average chlorophyll concentration of 2.3 mg/ml. In contrast, the specimens in the control group exhibited an average growth of 3.6 cm and an average chlorophyll content of 0.9 mg/ml.

The quantification of chlorophyll in the leaves was essential, since this pigment is directly correlated to the photosynthetic rate of the plants. During photosynthesis, carbon fixation (CO_2) and the release of molecular oxygen (O_2) occur, a continuous process throughout the plant cycle. The monitoring made it possible to estimate the amount of carbon assimilated through the analysis of the fresh and dry biomass of each species, measured in tons per hectare annually.

In addition, it was found that palm trees play a fundamental role in maintaining local biodiversity, attracting diverse species of fauna and flora and promoting ecological balance. This effect is associated with the biogeochemical cycle of palm trees, which capture atmospheric CO_2 during photosynthesis and incorporate it into their biomass. In this process, the assimilated carbon is converted into structural carbohydrates, such as glucose and starch, which are essential for plant metabolism and for the sustenance of other species. During the decomposition of leaves, fruits and trunks, nutrients are reintegrated into the soil, enriching the availability of essential compounds for the surrounding vegetation.

Some palm species play a crucial role in carbon sequestration, contributing to climate change mitigation. This process occurs due to the ability of these plants to store carbon in their plant tissues throughout growth. The unique shape of the canopy, characterized by the absence of branches, optimizes the absorption of solar radiation, favoring photosynthetic efficiency. In addition, its wide and compact leaves maximize biomass production, expanding its CO_2 fixation capacity.

With an average occupation of 160 cm^2 per individual on the soil surface, these species are highly adaptable to extensive agricultural areas, as long as the spacing allows the mechanization of crops. In addition, it was found that its cultivation can be integrated into agricultural systems without harming animal husbandry, keeping the soil enriched in nutrients through the dynamics of carbon sequestration. This interaction promotes environmental conservation and contributes to the reduction of greenhouse effect impacts.

According to Grattapaglia (2023), biofertilizers play a strategic role in increasing carbon sequestration and agricultural sustainability. Composed of organic waste, these inputs provide essential macro and micronutrients for plants, stimulating vegetative growth and promoting the accumulation of biomass, especially carbon, which is assimilated during photosynthesis.

From a genetic perspective, the application of biofertilizers can modulate the expression of genes involved in plant metabolism and development. Houghton (1994) points out that the edaphic microbiota exerts a significant influence on the gene regulation of plants, impacting physiological processes such as nutrient absorption, resistance to phytopathogens and root architecture. In this way, by favoring the microbial diversity of the soil through the addition of biofertilizers, it is possible to optimize the genetic performance of young palm trees, making them more efficient in capturing and storing atmospheric carbon.

CONCLUSION

The application of biofertilizers demonstrated high efficacy in optimizing the functionality of chloroplasts, resulting in a significant increase in the photosynthetic rate of the Arecaceous species selected for the experiment. This physiological improvement favored a more robust vegetative development, reflected in the increase of leaf biomass and the strengthening of nutritional reserves, driven by carbon sequestration.

In conclusion, the integration of environmental biotechnology into the Arecácea, combined with the use of biofertilizers and the controlled manipulation of chlorophyll and dilute CO₂, proves to be an innovative and highly promising strategy to maximize atmospheric carbon capture. This approach not only contributes to reducing the impacts of climate change, but also fosters environmental sustainability in agroecological systems. In addition, the expansion of carbon sequestration in genetically enhanced plants can enable the generation of additional carbon credits, which can be negotiated by companies and nations to offset their CO₂ emissions, in line with the guidelines of international agreements, such as the Paris Agreement.

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