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Development of Anadenanthera colubrina Seedlings Subjected to Different Doses of Coconut Powder and Arbuscular Mycorrhizal for Rehabilitation of Mining Areas

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Authors' contributions

This work was carried out in collaboration among all authors. The authors FMMS and MMF conducted the study, collected the analyzes in the field, did the statistical analysis of the data, wrote the protocol and wrote the first draft of the manuscript. The author RFP designed, monitored and supervised all of this study. The authors DSL and RNAF assisted in literature searches, writing in the manuscript and discussing the data. The authors VCP and MCF helped in the search of the literature and in the translation of the same into English language. All authors read and approved the final manuscript.

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Original Research Article

ABSTRACT

The objective of this work was to evaluate the development of angico [Anadenanthera colubrina (Vell.) Brenan] seedlings produced with different doses of coconut powder and inoculated with arbuscular mycorrhizal fungi (AMF) in nursery and mining degraded area in the state of Sergipe (Brazil). The experimental design was in randomized blocks, with the following proportions of mineral substrate and coconut powder, with and without AMF. Angico growth was evaluated in nursery after 150 days, and height and diameter were evaluated. Later the seedlings were planted in a degraded area by sand and gravel mining and after 120 days of planting the height, diameter and survival rate were measured. In the nursery phase, the addition of coconut powder promoted a reduction in height and diameter of angico seedlings. Inoculation with AMF significantly increased the height and diameter of *Anadenanthera colubrina* seedlings in the nursery. However, the field phase results show that the addition of coconut powder to the substrate promotes better development in height and diameter of *Anadenanthera colubrina* seedlings in the mining degraded area. The conjugation of a low proportion of coconut powder with the inoculation of AMF on *Anadenanthera colubrina* seedlings resulted in higher survival in the planting of degraded area by mining.

Keywords: Arbuscular mycorrhizal fungi (AMF); angico; survival rate; ecological restoration.

1. INTRODUCTION

Coconut cultivation stands out in Sergipe (Brazil) as a segment of agribusiness that generates more income, tax collection and play an important social role. In the Northeast highlight as the second largest producer, second only to the state of Bahia [1].

The increase in coconut water consumption generates about 6.7 million tons of bark.year⁻¹, becoming a serious environmental problem, especially for large cities. About 70% of the waste produced on the coast of Brazil's large urban centers is made up of coconut shells, a material that is difficult to degrade and which, in addition to focus and proliferation of diseases, has been shortening the life of landfills [1].

In much of the world, the plant substrate industry is seeking materials that are more elaborate and that meet the needs of plants and producers. Agro-industry residues such as coconut shell and decomposed organic materials appear as promising alternatives to mixtures [2].

Coconut shell has a large percentage of lignin (35-45%) and cellulose (23-43%) and a small amount of hemicellulose (3-12%), which is the fraction vulnerable to attack by microorganisms. These characteristics give the coconut shell substrate great durability and are therefore recommended for long cycle crops such as ornamental and forest species to recover degraded areas [3]. According to [4] the structure of coconut fiber associated with its

physicochemical properties makes it particularly suitable for use as a substrate.

According to Klein et al. [5], the residue or powder of ripe coconut shell has been indicated as an agricultural substrate, mainly because it has an advantageous physical structure providing high porosity, high moisture retention potential and being biodegradable.

Edaphic microorganisms have key functions in terrestrial ecosystems, and among these, arbuscular mycorrhizal fungi (AMF) stand out [6]. AMF can be present in the most varied types of soils and are considered critical organisms for plant succession within a process of revegetation of degraded areas. Currently, its applicability as a biological input has still been challenging for scientific research [6].

Soils degraded by mining activity often have severe nutritional limitations such as N and P, as AMF increase the absorption of these nutrients, this symbiosis contributes to improving soil fertility and improving the edaphic environment for the establishment of new species on site [7]. If the effects of AMF on N uptake by plants are as widespread as those found for P, the role of AMF in the functionality of degraded environments will be greater than currently considered.

Early studies using fast-growing native leguminous species revealed that double inoculation with AMF and nitrogen-fixing bacteria is an important strategy for revegetation of degraded areas, providing improvements in soil physical, chemical and biological attributes [7], favoring growth and anticipating the planting of seedlings in the field, promoting greater survival of seedlings in nurseries and after transplanting in the field and in dry periods [8].

Choosing correctly the plant community that will start the succession process in a degraded area is one of the most critical points of the ecological restoration process. The definition of key species, those that control the structure of the community due to its abundance, spatial distribution, biomass, size or cover and which influence the occurrence of other associated species, being of fundamental importance for the success of ecological restoration in degraded areas [9].

In this sense, this work aimed to evaluate the development of angico seedlings produced with different doses of coconut powder and inoculated with AMF in nursery and mining degraded area in the state of Sergipe (Brazil).

2. MATERIALS AND METHODS

In the nursery phase, the experiment was started in September 2018 in a greenhouse of the seedling biofactory of SERGIPETEC, São Cristovão, SE. Different dosages of coconut powder and with and without AMF were tested for seedlings production of angico Table 1.

The substrate used for seedling production was composed of black earth and washed sand, in a 3: 1 ratio. To produce the coconut powder, empty coconuts were collected from trades in the city of Aracaju and São Cristovão. Subsequently, the external fibers were removed and sun-dried, then minced in a chopper / forage harvester.

The different proportions of substrates were packed in 800 cm³ polyethylene bags. In each plastic bag were placed 3 seeds and at this time in the treatments with AMF were inoculated with inoculants of Glomus clarum. After seedling emergence, thinning was performed leaving only one seedling per bag. The seedlings were kept under roof 50% until the emergency occurred and then transferred to an area in full sun.

The seedlings were measured in the nursery phase at 5 months with the following growth parameters for each treatment: the stem diameter (D) and shoot height (H). The diameter was measured with a digital caliper at 10 cm from the ground and the height was measured

with a tape measure, taking as default the last pair of leaves, being H in cm and D in mm.

The treatments were arranged in a randomized block design in factorial 3 (coconut fiber doses) x 2 (with AMFand without AMF), with 50 seedlings per treatment. The results were submitted to analysis of variance by F test, 1% significance and Tukey's test for comparison between means, with the aid of the statistical program Sigma Plot.

After the nursery phase, the seedlings were planted in a mining degraded area, located on Fazenda Itália owned by the company CAL TREVO, located in the municipality of Itaporanga da A'juda, SE. The study area had a degradation process by removing the topsoil and subsequently removing the subsoil layers for gravel collection. This withdrawal took place in 2007 and was later abandoned.

In the degraded area were planted 50 individuals produced in the nursery phase, for each treatment in spacing of 3 x 3 m and the dimensions of the pit 40 x 40 cm, making a sample area per treatment (planting) of 450 m².

The seedlings were measured in the field at 4 months in the following growth parameters for each treatment: the stem diameter (D) and the shoot height (H). The diameter was measured with a digital caliper at 10 cm from the ground and the height measured with a tape measure, taking as default the last pair of leaves, being H in cm and D in mm. In the field, a randomized block design with the same treatments and repetitions as in the nursery phase was used. The results were submitted to analysis of variance by F test, 1% significance and Tukey's test for comparison between means, with the aid of the statistical program Sigma Plot.

3. RESULTS AND DISCUSSION

Height and diameter measurements showed statistically higher in the treatments 4 and 5 in the nursery phase Table 2.

Regarding height and diameter, the treatment 5 seedlings presented the highest values Table 2. In this sense, it is observed that the addition of coconut powder to the mineral substrate in the nursery phase would not be recommended, and inoculation with AMF would be sufficient to produce *Anadenanthera colubrina* seedlings for revegetation of mining degraded areas. The authors [10,11] found that colonization by

Treatments	Proportions	Mineral substrate	Coconut powder	AMF
1	1:1	1	1	With AMF
2	1:1	1	1	Without AMF
3	2:1	2	1	With AMF
4	2:1	2	1	Without AMF
5	1:0	1	0	With AMF
6	1:0	1	0	Without AMF

Table 1. Formulated substrates (v/v) for the different treatments

AMF = Mycorrhizal fungi

AMF can cause food in the allocation of carbon to the shoots than when not mycorrhized, which explains better seedling diameter and height developments found in this work.

The fact that height and diameter are not favorable for the addition of coconut powder to the mineral substrate in the nursery phase was explained by [12], who noted that coconut powder-based substrates reduce the height and diameter of forest seedlings, due to the high content of healthy tannins that when concentrated are phytotoxic. These tannins promote root tip inhibition by reducing nutrient uptake by forest seedlings, reducing growth in height and diameter of the stem [12].

According Delarmelina et al. [13], evaluating seedlings production of Sesbania virgata (Cav.) Pers. observed that the addition of coconut powder above 60% promoted a reduction in diameter. Another factor to influence height and diameter negatively, according to Santos et al. [14] is that the increase of coconut powder to the substrate for forest seedling production promotes a reduction of P, due to the low content of this nutrient in coconut powder Table 2.

Table 2. Height and diameter measurements of angico seedlings with different proportions of mineral substrate: Coconut powder and inoculated with AMF in the nursery phase after 150 days

Treatments	Height (cm)	Diameter (mm)
1	7,32b	1,45a
2	6,64c	0,74c
3	7,90b	1,40a
4	8,70a	1,44a
5	9,81a	1,48a
6	7.77b	1.24b

Averages followed by different lower case letters mean that the treatments differ from each other by the Tukey's test

Thus, treatment 2 with the highest percentage of coconut powder had the lowest diameter and

height and treatment 2 was lower than treatment 1 which has the same proportion of coconut powder, but treatment 1 was inoculated with fungi. mycorrhizal plants which promotes a higher absorption of P. In an experiment for seedling production of *Moringa oleifera* [15], observed that the use of green coconut fiber in the substrate should be used in proportions less than 25%, as they reduce the growth of *Moringa oleifera* seedlings and promote nitrogen deficiency.

The highest values of height in the field phase were observed in treatments 1 and 4, differing statistically in relation to the other treatments Table 3. Regarding diameter, the highest values were observed in treatments 1, 3 and 4, differing significantly Table 3.

Table 3. Mean height and diameter of angico seedlings with different proportions of mineral substrate: Coconut powder and inoculated with AMF in the field phase

Height (cm)	Diameter (mm)
22,25a	1,93a
10,45b	1,81b
10,76b	2,01a
17,62a	2,05a
9,61b	1,70b
3,00c	1,02c
	22,25a 10,45b 10,76b 17,62a 9,61b

Averages followed by different lower case letters mean that the treatments differ from each other by the Tukey's test

Thus, *Anadenanthera colubrina* seedlings from treatments 1 and 4 showed the best development when planted in the degraded area by mining Table 3. This shows that the addition of coconut powder to the substrate in treatments 1 and 4 promoted a better development of *Anadenanthera colubrina* seedlings in the field. The addition of other residues such as sewage sludge and tree pruning also demonstrated that *Anadenanthera colubrina* seedlings showed better growth in height and diameter compared to mineral substrates [16].

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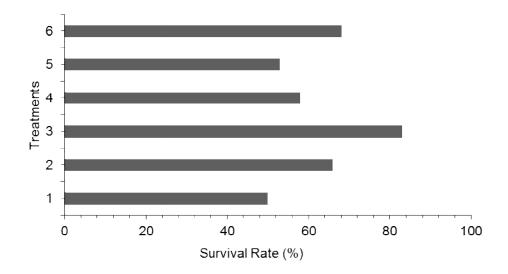


Fig. 1. Survival rate of angico with different proportions of mineral substrate: Coconut powder and inoculated with AMF in the field phase

The Anadenanthera colubrina seedlings were planted in an area degraded by sand and gravel mining, with the soil in the area being a sandy subsoil with low water retention capacity. In this sense, the seedlings produced in coconut powder substrate in its composition can retain larger amount of water, having a better development in the degraded area in the evaluated period. Thus, treatment 6 presented the smallest diameter and height, being the treatment in which no AMF were inoculated and no coconut powder was added, compromising the development in the degraded mining area.

In an experiment for seedling production of two eucalyptus species [17], observed that the seedlings produced in coconut fiber substrate presented better development in height and diameter, due to the large microporosity of coconut fiber, conferring greater water retention capacity.

Survival rates may be considered low because rate of 80% or less except treatment 3 (Fig. 1).

In reforestation projects in degraded areas, a survival rate above 80% can be considered a high plantation survival rate [18,19].

Treatment 3 presented the highest survival rate, that is, the conjugation of seedlings produced with coconut powder as a source of organic material in the substrate and the inoculation with AMF can increase the survival of *Anadenanthera colubrina* seedlings in areas degraded by mining. The importance of the association of AMF in *Anadenanthera colubrina* seedlings for field survival was observed by [8], in which seedlings inoculated with AMF and without fertilization presented higher survival rate.

4. CONCLUSION

The results showed that the addition of coconut powder to the substrate promoted a better development in height and diameter of *Anadenanthera colubrina* seedlings in the mining degraded area. The treatment 4 with coconut powder and inoculation of AMF on *Anadenanthera colubrina* seedlings resulted in higher survival in the planting of degraded area by mining.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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