

## SOIL FUNGI PROMOTE VIGOR IN SEEDS FROM BRAZILIAN DRY SEASONAL FOREST

FUNGO DE SOLO PROMOVE VIGOR EM SEMENTES DO BIOMA CAATINGA

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### ABSTRACT

The Brazilian Dry Seasonal Forest (Caatinga Biome) is poorly studied about possible interactions between seeds, seedlings, and *Trichoderma harzianum* strains to promote vegetal vigor in seedlings. So, this research aims to study the *T. harzianum* strain promotion of vigor in different species from Caatinga Biome in nursery. For assays, we used: *Aspidosperma pyrifolium*, *Cnidocolus quercifolius*, *Handroanthus impetiginosus*, *Pseudobombax marginatum*, and *Tabebuia aurea* species and the *T. harzianum* IBLF 006 based-product-inoculant. We tested these assays in nursery conditions. *T. harzianum* solution (0.5% v/v) was used in the nursery assay (applied over soil after the sowing). For this research, we evaluated variables of vigor and germination and there was a control treatment (non-inoculated). About nursery assay results, there was a positive interaction for all species between seeds and seedlings and *T. harzianum*, except for *T. aurea* and *C. quercifolius*. For all species tested in this research, we suggest that other seed or soil treatments can be tested in environmental Caatinga Biome conditions to expand this knowledge about the interaction between *Trichoderma harzianum* and seedlings and seeds from Caatinga biome.

KEYWORDS: *Trichoderma harzianum*, Caatinga Biome, Brazilian Semiarid, Seed Technology.

### RESUMO

A Floresta Estacional Seca Brasileira (Bioma Caatinga) é pouco estudada sobre as possíveis interações entre sementes e mudas com isolados de *Trichoderma harzianum* para promoção de vigor vegetal. Assim, esta pesquisa tem como objetivo estudar a promoção do vigor de *T. harzianum* em diferentes espécies do bioma Caatinga em condições de viveiro. Para os ensaios, foram utilizadas sementes das espécies *Aspidosperma pyrifolium*, *Cnidocolus quercifolius*, *Handroanthus impetiginosus*, *Pseudobombax marginatum* e *Tabebuia aurea* e um produto comercial a base de *T. harzianum* IBLF 006. O experimento foi realizado em condições de viveiro. A solução de *T. harzianum* (0,5% v / v) foi aplicada sobre o solo após a semeadura. Sendo avaliadas as variáveis de vigor e germinação, houve um tratamento controle (não inoculado). Sobre os resultados dos ensaios no viveiro, houve interação positiva entre sementes e plântulas com o isolado de *T. harzianum* para todas as espécies estudadas, exceto para *T. aurea* e *C. quercifolius*. Para todas as espécies testadas neste trabalho, pode ser sugerido que outros tratamentos de semente ou solo possam ser testados em condições ambientais do Bioma Caatinga, para expandir o conhecimento sobre a interação entre isolados de *Trichoderma harzianum*, mudas e sementes provenientes do bioma Caatinga.

KEYWORDS: *Trichoderma harzianum*, Bioma Caatinga, Semiárido Brasileiro, Tecnologia de Sementes.

## INTRODUCTION

*Trichoderma* is a common gender soil-borne fungi found in some different biomes around the world. Species of *Trichoderma* have several ecological characteristics such as biological control of other organisms, vegetal growth promotion, breaking of soil substances, and symbiosis (SOUZA et al., 2016). Due to its diversity, some different researchers approach these characteristics for sustainability; some strains are studied nowadays, but for farm production, the *T. harzianum* species is the most studied (SILVA et al., 2020). Zhang et al. (2021) affirm that this gender is an important symbiotic and endophytic, mainly to ecosystem regulation around the world.

Carvalho et al. (2021), Michelen-Álvarez & Martínez-Hernández (2021), and Silva et al. (2021), describe the importance of this fungi gender in symbiosis with plants from the Caatinga, the Brazilian Dry Seasonal Forest, such as a promoter of resistance to the dry season, biological control of pathogens and vegetal growth promotion. On the other hand, the Caatinga is characterized by severe drought events, average luminosity, and high annual temperatures, being adapted to xerophilic conditions, endemism, and high factor of anthropization, and desertification (FERREIRA et al., 2021 a).

Meiado et al. (2012) also describe the need for research with plants from the Caatinga, to improve technologies of conservation and recuperation. And, following Ferreira et al. (2021 b), it can be described that this biome is poorly studied and it's breaking down before it's understood. Then, the studies for conservation and recuperation of Caatinga are important for promoting sustainability in this area, and the sustainable production of native species seedlings (FERREIRA et al., 2021a; FERREIRA et al., 2021 b).

Caatinga wood is used to timber and landscaping is the Bignoniaceae group, in this paper. it represented *Handroanthus impetiginosus* (Mart. ex DC.) Mattos (Bignoniaceae), as described by Almeida et al. (2020) and Araújo et al. (2020), and *Tabebuia aurea* (Silva Manso) Benth. & Hook. f. ex S. Moore (OLIVEIRA et al., 2012; SANTO et al., 2012; BRITO et al., 2021).

*Cnidocclus quercifolius* Pohl is an important species in the semiarid region because it is used for ethnobotanical (medicament) and forage (DIAS JÚNIOR et al., 2019). *Pseudobombax marginatum* (A. St.-Hil. Juss. & Cambess.) A. Robyns (Malvaceae) is a tree species found in several Brazilian biomes, commercially used to produce fibers, medicines, and timber (AMORIM et al., 2009). *Aspidosperma pyriforme* Mart. & Zucc. is an important plant species from Caatinga Biome used to timber,

cosmetic, and medicinal (FERREIRA & CUNHA, 2000; PEREIRA et al., 2016; ANDRADE JÚNIOR et al., 2020).

The production of seedlings in nurseries has an important role in the revitalization of Caatinga areas and supports commercial forest production. These facts are supported by the perspective of the production of vigorous seeds and seedlings (DANTAS et al., 2020; OLIVEIRA et al., 2020). Then, this research aims to study the *T. harzianum* strain promotion of vigor in different species from Caatinga Biome (*Aspidosperma pyriforme*, *Cnidocclus quercifolius*, *Handroanthus impetiginosus*, *Pseudobombax marginatum* and *Tabebuia aurea*).

## MATERIAL AND METHODS

### Seeds and experimental localization

The assays were realized in the nursery from Semiarid Nacional Institute (Campina Grande, Paraíba State, Brazil) in the second semester of 2021. The region shows the BSh climate classification, with temperature variables between 20 to 32°C, for research occasion (FERREIRA et al., 2021a; FERREIRA et al., 2021 b). For these assays, we used *Aspidosperma pyriforme*, *Cnidocclus quercifolius*, *Handroanthus impetiginosus*, *Pseudobombax marginatum* and *Tabebuia aurea* seeds ceded by NEMA (Núcleo de Ecologia e Monitoramento Ambiental) for UNIVASF (Universidade Federal do Vale do São Francisco) (Table 1).

### Nursery assay

We sowed the seeds in the nursery of soil (Neossolo regolith soil), to protect the environment (sombre 50%) (Table 2). The treatments used were *T. harzianum* IBLF 006 SC based-product-inoculant (TRICHODERMIL®, Koppert) was used in nursery assay (applied over the soil after the sowing) in solution (0.5% v/v), according to methodologies describes to Ferreira et al. (2020). In this assay, we evaluated variables of vigor and germination and there was a control treatment (non-inoculated).

Due to the experimental assay, we have the variables in the third day after sowing, the variables: emergence speed index (ESI), average emergence time (AET), average emergence speed (AES), and dry weight variable (DW), according to Ferreira et al. (2021 a). The percentage variables of emergence in 14 (14 E), 21 (21 E), and 28 (28 E) days of sowing were analyzed according to Ferreira et al. (2021 b).

**Table 1.** Family botanical, genus, species, geographic parameters, and germination of seeds used in this research.

Family	Gender	Species	Localization	Latitude	Longitude	Altitude (m)	Germination (%)
Apocynaceae	<i>Aspidosperma</i>	<i>pyrifolium</i>	Pernambuco  Petrolina	-8.77	-40.53	418	97
Bignoniaceae	<i>Handroanthus</i>	<i>impetiginosus</i>	Ceará  Brejo Santo	-7.59	-38.88	409	91
Bignoniaceae	<i>Tabebuia</i>	<i>aurea</i>	Pernambuco  Petrolina	-9.32	-40.55	390	83
Euphorbiaceae	<i>Cnidocolus</i>	<i>quercifolius</i>	Pernambuco  Floresta	-8.55	-38.04	457	71
Malvaceae	<i>Pseudobombax</i>	<i>marginatum</i>	Pernambuco  Salgueiro	-8.12	-39.19	441	86

Fonte: NEMA.

**Table 2.** Chemical characters from substrate of nurseries (0-20 cm deep).

pH	P	K <sup>+</sup>	Na <sup>+</sup>	H <sup>+</sup> + Al <sup>+3</sup>	Al <sup>+3</sup>
H <sub>2</sub> O (1:2,5)	mg.dm <sup>3</sup>		cmolc.dm <sup>3</sup>		
5.4	3.3	98.09	0.11	3.37	0,2
Ca <sup>+2</sup>	Mg <sup>+2</sup>	V%	CTC	OM	
cmolc.dm <sup>3</sup>				g.kg	
3.34	0.32	54.4	7.39	6.46	

P, K, Na: Extractor 1 of Mehlich; H + Al: Extrator Ca acetatate 0,5 M, pH 7,0; pH 7,0; Extractor Al, Ca, Mg: KCl 1 M; OM: Organic Matter - Walkley-Black; V% (FERREIRA et al., 2021).

### Statistical analyses

The treatments were analyzed using the Chi-square analysis ( $\chi^2$ ), at 5% significance, the test was performed at a significance level of 5%, and in this case, H<sub>0</sub> is rejected when p-value  $\leq$  0.05 and accepts H<sub>0</sub> otherwise. Also, the means obtained were treated using non-parametric statistics (mean per treatment, general mean, coefficient of variation, standard error, and deviation).

### RESULTS AND DISCUSSION

About the emergence at 14 days after sowing variable (14 E) results we would observe that in all vegetal species there were significant differences between the treatments, except *A. pyrifolium* species. The *T. harzianum* presence promotes the best average to *C. quercifolius*, and there were positive Pearson Correlations to *T. aurea* species for this variable (Table 3).

Regarding the percentage of emergence at 21 days after sowing variable results (21 E), for all vegetal species there were significant differences between the treatments, except to *A. pyrifolium* seeds; The *T. harzianum* presence promotes the best average all species, except to *A. pyrifolium* seeds (Table 3).

About the percentage of emergence at 28 days after sowing variable (28 E), for all vegetal species there were significant differences between the treatments and the *T. harzianum* presence promotes the best average for *C.*

*quercifolius* and *P. marginatum* seeds (Table 3).

About the Emergence Speed Index variable (ESI), for all vegetal species, there were significant differences between treatments, except to *Tabebuia aurea* seeds. *T. harzianum* presence promotes the best average to *Tabebuia aurea* and *C. quercifolius* seeds, and there were positive Pearson Correlations to all species for this variable (Table 3).

Regarding the Average emergence time Index variable (AET), for all vegetal species, there were significant differences between the treatments and the *T. harzianum* presence promotes the best average of all species, except to *H. impetiginosus* and *T. aurea* seeds (Table 3).

Concerning the average time index (AES), for all vegetal species studied there were significant differences following between the treatments and the *Trichoderma harzianum* presence promotes the best average for all species, except to *Tabebuia aurea* (Table 3).

And about the dry weight variable (DW), in all the species there were statistical differences between the treatments, and the promotion of biomass production to *Trichoderma* present were better than the control. Regarding the variable dry weight, all species, differences between treatments; the presence of *Trichoderma* promoted better mean in the species *Anadenanthera colubrina*, *Handroanthus impetiginosus*, and *Cnidocolus quercifolius*. The importance of contextualized studies about the forest production in Caatinga biome areas in its summary to the sustainable development of semi-arid regions, in accordance with Carvalho et al. (2021) (Table 3).

**Table 3.** Averages and nonparametric statistic to variables Emergence Speed Index (ESI), Average Emergence Time (AET), Average Emergence Speed (AES), Dry Weight Variable (DW), And Percentage Variables of Emergence in 14 (14 E), 21 (21 E), and 28 (28 E) on seeds of *Aspidosperma pyriformium*, *Cnidoscopus quercifolius*, *Handroanthus impetiginosus*, *Pseudobombax marginatum*, and *Tabebuia aurea* sowing on nursery with soil treated or non-treated with *Trichoderma harzianum*.

	E14	E21	E28	ESI	AET	AEI	DW
	%			Index			g.plant <sup>-1</sup>
<b><i>Cnidoscopus quercifolius</i></b>							
Control	55.00	62.00	66.00	2.31	16.49	0.06	1.70
<i>Trichoderma harzianum</i>	49.00	60.00	61.00	2.13	15.25	0.07	1.58
<b>Chi-square test</b>	0.34	0.76	0.39	0.30	0.36	0.33	0.80
<b>Standard deviation</b>	9.56	6.68	7.84	0.22	2.32	0.01	0.51
<b>Coefficient of variation (%)</b>	18.39	10.94	12.34	10.01	14.62	12.56	30.88
<b>Average</b>	52.00	61.00	63.50	2.22	15.87	0.06	1.64
<b>Pearson Correlation</b>	0.52	-0.48	0.26	0.21	0.67	0.71	-0.21
<b><i>Handroanthus impetiginosus</i></b>							
Control	52.00	68.00	65.00	2.32	17.33	0.06	0.68
<i>Trichoderma harzianum</i>	66.00	57.00	69.00	2.65	15.28	0.07	0.62
<b>Chi-square test</b>	0.07	0.15	0.39	0.01	0.32	0.32	0.53
<b>Standard deviation</b>	11.66	10.24	10.20	0.34	2.57	0.01	0.36
<b>Coefficient of variation (%)</b>	19.77	16.38	15.22	13.49	15.76	14.57	55.60
<b>Average</b>	59.00	62.50	67.00	2.49	16.30	0.06	0.65
<b>Pearson Correlation</b>	0.65	0.25	0.74	0.92	0.08	0.26	0.99
<b><i>Pseudobombax marginatum</i></b>							
Control	70.00	81.00	84.00	2.96	20.08	0.05	0.54
<i>Trichoderma harzianum</i>	73.00	71.00	74.00	3.17	17.21	0.06	0.61
<b>Chi-square test</b>	0.75	0.28	0.19	0.56	0.19	0.18	0.57
<b>Standard deviation</b>	10.13	11.11	9.97	0.36	2.61	0.01	0.12
<b>Coefficient of variation (%)</b>	14.16	14.62	12.62	11.73	14.02	13.04	20.29
<b>Average</b>	71.50	76.00	79.00	3.07	18.64	0.05	0.58
<b>Pearson Correlation</b>	-0.29	-0.04	0.18	-0.42	-0.14	-0.08	-0.82

**Table 3.** Averages and nonparametric statistic (Cont...).

	<b>E14</b>	<b>E21</b>	<b>E28</b>	<b>ESI</b>	<b>AET</b>	<b>AEI</b>	<b>DW</b>
	<b>%</b>			<b>Index</b>			<b>g.plant-1</b>
<b><i>Tabebuia aurea</i></b>							
Control	63.00	71.00	71.00	2.96	18.14	0.06	0.50
<i>Trichoderma harzianum</i>	65.00	71.00	73.00	2.80	16.00	0.06	0.70
<b>Chi-square test</b>	0.88	1.00	0.80	0.62	0.29	0.30	0.18
<b>Standard deviation</b>	11.51	8.21	8.28	0.31	1.95	0.01	0.22
<b>Coefficient of variation (%)</b>	17.99	11.57	11.50	10.85	11.45	11.21	36.24
<b>Average</b>	64.00	71.00	72.00	2.88	17.07	0.06	0.60
<b>Pearson Correlation</b>	-0.90	0.39	-0.39	-0.79	-0.95	-0.91	0.66
<b><i>Aspidosperma pyrifolium</i></b>							
Control	52.00	67.00	62.00	2.60	16.61	0.06	0.49
<i>Trichoderma harzianum</i>	69.00	83.00	74.00	3.27	16.62	0.06	0.49
<b>Chi-square test</b>	0.02	0.21	0.34	0.00	1.00	0.99	0.98
<b>Standard deviation</b>	10.13	14.46	13.35	0.41	3.05	0.01	0.16
<b>Coefficient of variation (%)</b>	16.74	19.28	19.64	13.91	18.33	19.94	32.05
<b>Average</b>	60.50	75.00	68.00	2.94	16.61	0.06	0.49
<b>Pearson Correlation</b>	0.00	-0.24	-0.56	0.70	-0.70	-0.66	-0.73

Michelena-Álvarez & Martínez-Hernández (2021) affirm that the actions of recuperations and wood production in Caatinga area areas are incipient because technologies adapted to the conditions, achievements, and possibilities of the region are lacking in the literature. Nevertheless, Silva et al. (2021), the use of environmental management that promotes the symbiosis organism and native plants from the Caatinga biome is poorly studied (DANTAS et al., 2019; DANTAS et al., 2020). In this paper (Table 3), we argued about the interaction between the *Trichoderma harzianum* strain and important species from the Caatinga biome.

Ferreira et al. (2021 a), declare that nursery production supports forest production, since continuing with extractivism is not sustainable for the Earth. Meiado et al. (2012), Ferreira et al. (2021 b), and Silva et al. (2021) are necessary for the conservation and recuperation of Caatinga and the promotion of seed technologies adapted for this land (Table 3). Adding to this idea, Ferreira et al. (2021 b) affirm that the Caatinga biome is an emerging location in the recovery of its areas, due to the loss of biodiversity, low development economical, and social and environmental problems.

For Carvalho et al. (2021), Michelena-Álvarez & Martínez-Hernández (2021) the presence of *Trichoderma* species such as symbiotic in the Caatinga can viabilize the utilization this gender in projects of forest production. Carvalho et al. (2021) describe that *Trichoderma* strains from the Caatinga can be an evolutionary ratio between the local vegetation. *Trichoderma harzianum*, used in this paper is an important species found in forest (SOUZA et al., 2016), with the importance of the control of other organisms, vegetal growth promotion, breaking of soil substances, and symbiosis, according to writings of Silva et al. (2020). The *T. harzianum* IBLF 006 is a strain described in the literature such as a vegetal growth promoter in lettuce (PEREIRA et al., 2019), *Jacaranda mimosifolia* (FERREIRA et al., 2021 c), and *Moringa oleifera* (FERREIRA et al., 2020). Oliveira et al. (2019), describe that the *T. harzianum* IBLF 006 strain is resistant to high temperatures and ultraviolet radiation, a factor important to be considered in Caatinga conditions, such as for Silva et al. (2021). The results for each of the species studied in this research will be discussed below (Table 3).

About the *Handroanthus impetiginosus* the interaction is positive (Table 3), possibly a great emergence and vigor, as described by Almeida et al. (2020) and Araújo et al. (2020). The *H. impetiginosus* seeds are light and with great cellulose content, what it confers for then the possibility to dispersion more saturated. Almeida et al. (2020) and this

may be the proposal that promotes the positive interaction between its *T. harzianum* strain.

The results obtained to *Pseudobombax marginatum* are so positive to interaction and use of *Trichoderma* on nursery soil (TableS 2 and 3), probably the mucilages produced for these seeds in the emergence (AMORIM et al., 2009) promote a good place for colonization of *T. harzianum* and this microorganism promotes a positive interaction, according to Michelena-Álvarez & Martínez-Hernández (2021).

Also, for *Aspidosperma pyrifolium* the presence of *T. harzianum* is positive, probably to the low tanines counts, and the morphology of seeds taht help to *Trichoderma* to open to internals tissue of seeds (FERREIRA & CUNHA, 2000; PEREIRA et al., 2016; ANDRADE JÚNIOR et al., 2020) (Table 3).

About *Tabebuia aurea* and *Cnidoscolus quercifolius* species (Table 3) the interactions were negative, probably for interactions between these seeds, inherent to the natural constitution (DANTAS et al., 2019; DANTAS et al., 2020). For example, the seed of *Tabebuia aurea* produces a pigment in emergence to repel insects and other microorganisms, it can be dangerous to *T. harzianum* too (OLIVEIRA et al., 2012; SANTO et al., 2012; BRITO et al., 2020). Seeds of *Cnidoscolus quercifolius* are made up of oil content (DIAS JÚNIOR et al., 2019). Although the microbiome present in the seeds of this lot may repel some microorganisms, because there was an intimate ecological action between species from Caatinga and other microorganisms, such as describe Carvalho et al. (2021) and Michelena-Álvarez & Martínez-Hernández (2021).

## CONCLUSIONS

About the results, there was a positive interaction for all species between seeds and seedlings and *T. harzianum*, except for *T. aurea* and *C. quercifolius*. For all species tested in this paper, we suggest that other seed or soil treatments can be tested in environmental conditions to expand this knowledge about the interaction between *Trichoderma harzianum* and seedlings and seeds from Caatinga.

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