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MILENA PUPO RAIMAM

RECICLAGEM DE RESÍDUO SIDERÚRGICO PARA A PRODUÇÃO DE MUDAS DE *Corymbia* ASSOCIADA À BIOESTIMULAÇÃO COM *Bacillus* sp. CCMD862

BELÉM
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Tese de doutorado apresentada à Universidade Federal Rural da Amazônia, como parte das exigências do Curso de Pós-Graduação em Agronomia: área de concentração Produção Vegetal, para a obtenção do título de doutora.

Orientadora: Profa. Dra. Gisele Barata da Silva.
Co-orientador: Prof. Dr. Gledson Luiz Salgado de Castro

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“*Uma resposta aproximada ao problema certo vale mais do que uma resposta exata para um problema aproximado.”*

John Wilder Tukey

RESUMO

Esta tese é o resultado de uma pesquisa experimental que focaliza a reciclagem da escória de aciaria de forno elétrico a arco (EAE), um subproduto do refino do aço, combinada a rizobactérias promotoras de crescimento vegetal (PGPR) para uso como bioinsumo na agricultura. Neste estudo, trabalho na perspectiva de compreender os efeitos sinérgicos desta combinação sobre o desenvolvimento vegetal de clones de *Corymbia torelliana x Corymbia citriodora*, um hídrido utilizado na produção de carvão vegetal entre outros fins. Para o alcance deste objetivo, etapas estruturadas foram definidas, as quais orientaram a obtenção de isolados bacterianos com capacidade de promoção de crescimento de plantas e simultânea tolerância ao subproduto siderúrgico bem como a investigação das respostas fisiológicas, bioquímicas, morfo-anatômicas, nutricionais e de crescimento apresentadas pela espécie vegetal selecionada. A tese está dividida em quatro capítulos. O Capítulo 1 traz a contextualização sobre a origem, caracterização, gestão, desafios e potencialidades de uso do resíduo siderúrgico EAE na agricultura, o potencial da interação planta-PGPR, incluindo exemplos da interação com o gênero *Corymbia*. O Capítulo 2 apresenta a prospecção de PGPR em área de depósito de resíduo siderúrgico, a caracterização do isolado *Bacillus* sp. CCMD862 e o resultado da inoculação deste sobre o crescimento de mudas clonais híbridas de *Corymbia*. O Capítulo 3 apresenta os efeitos da combinação de EAE e *Bacillus* sp. CCMD862 sobre parâmetros fotossintéticos, bioquímicos, nutricionais e de crescimento de *Corymbia* sp. O Capítulo 4 traz as alterações morfo-anatômicas em raízes de *Corymbia* sp. desencadeadas pela combinação da rizobactéria *Bacillus* sp. CCMD862 com EAE relacionadas aos ganhos de crescimento do vegetal. Por fim, nas considerações finais apresento as principais conclusões deste estudo e as implicações para pesquisas futuras.

Palavras-chave: escória siderúrgica; PGPR; bioinsumo; eucalipto.

ABSTRACT

This thesis is the result of experimental research focusing on the recycling of electric arc furnace slag (EAE), a by-product of steel refining, combined with plant growth-promoting rhizobacteria (PGPR) for use as a bioimput in agriculture. In this study, I am working to understand the synergistic effects of this combination on the plant development of clones of *Corymbia torelliana x Corymbia citriodora*, a hybrid used to produce charcoal, among other things. In order to achieve this objective, structured stages were defined, which guided the obtaining of bacterial isolates with the capacity to promote plant growth and simultaneous tolerance to the steel by-product, as well as the investigation of the physiological, biochemical, morpho-anatomical, nutritional and growth responses presented by the selected plant species. The thesis is divided into four chapters. Chapter 1 provides background on the origin, characterization, management, challenges and potential for using EAE steel waste in agriculture, the potential of the plant-PGPR interaction, including examples of interaction with the *Corymbia* genus. Chapter 2 presents the prospecting of PGPR in a steel waste deposit area, the characterization of the *Bacillus* sp. CCMD862 isolate and the results of its inoculation on the growth of hybrid Corymbia clonal seedlings. Chapter 3 presents the effects of the combination of EAE and *Bacillus* sp. CCMD862 on the photosynthetic, biochemical, nutritional and growth parameters of *Corymbia* sp. Chapter 4 presents the morpho-anatomical changes in *Corymbia* sp. roots caused by the combination of the rhizobacterium *Bacillus* sp. CCMD862 with EAE, related to the plant's growth gains. The last section presents the main conclusions of this study and the implications for future research.

Keywords: steel slag; PGPR; bioinput; eucalyptus.

SUMÁRIO

1 CONTEXTUALIZAÇÃO

A industrialização é um processo crítico para o desenvolvimento econômico e para a transformação de um país, porém, traz consigo a geração de resíduos que conferem riscos ao ambiente e à saúde humana. Nesse contexto, é necessário repensar os processos produtivos, buscando alternativas inovadoras e sustentáveis para minimizar o impacto dos resíduos gerados.

1.1 O aço

O Brasil é o nono produtor mundial de aço e a América Latina lidera esse *ranking*, tendo produzido 36 milhões de toneladas em 2021 (WORLD STEEL ASSOCIATION, 2022). O saldo comercial do setor foi de US $ 4,4 bilhões, no mesmo ano, com 6 milhões de toneladas de produtos siderúrgicos exportadas. Considerando a grande versatilidade do aço e sua aplicação em incontáveis produtos e processos, cada habitante do país consumiu 123 kg de produtos siderúrgicos em 2021, produzidos por uma cadeia que atualmente emprega 73.606 colaboradores no país (INSTITUTO AÇO BRASIL, 2023).

O estado do Pará é a maior planta produtora de ferro-gusa, aço e ferro-ligas do norte do país, graças a produção do município de Marabá, com capacidade produtiva de 380 mil toneladas de aço ao ano, gerando aproximadamente 1.300 empregos diretos, o que evidencia a importância da atividade ao PIB do estado (INSTITUTO AÇO BRASIL, 2021).

O aço é uma liga de ferro e carbono que pode ser fabricado por dois modelos produtivos considerando a matéria prima: a base de minério de ferro reduzido a ferro-gusa e/ou a base de sucata de aço (BRANCA *et al.*, 2020a). Além do tipo de insumo dominante, as usinas podem ser integradas realizando todas as operações de produção do aço (redução, refino e laminação), semi-integradas quando operam duas fases (refino e laminação) e não-integradas quando realizam apenas uma destas operações, atuando muitas vezes como um fornecedor para plantas siderúrgicas de maior escala (MOURÃO, 2011).

Nas usinas integradas, a redução do minério de ferro em ferro-gusa ocorre em alto-forno com o consumo de carvão vegetal, oriundo do manejo de florestas de eucalipto. O gusa resultante é refinado em fornos no setor de aciaria e convertido a diferentes tipos de aço, mediante a queima e eliminação de impurezas, etapa que inclui algumas adições à mistura com a finalidade de melhorar as características do aço, proteger os fornos e otimizar a separação das impurezas (MARTINS, 2019).

Ao longo dessas etapas, diferentes subprodutos são gerados, denominados genericamente de escórias, com composição química variável. Em 2020, o Brasil produziu 622 kg de subprodutos por tonelada de aço bruto, destes 155 kg eram de escórias de aciaria (INSTITUTO AÇO BRASIL, 2021).

A escória de aciaria elétrica (EAE), recebe este nome por ser gerada em aciarias com fornos elétricos a arco ou oxigênio, apresenta características físico-químicas atreladas à composição do minério de ferro, à qualidade da sucata, dos fundentes adicionados e do ambiente químico onde é produzida, resultando em elevados valores de pH e com predomínio de óxidos de ferro básicos (FeO), cálcio (Ca), magnésio (Mg), silício (Si), manganês (Mn) e fósforo (P) além de outros elementos como zinco (Zn), molibdênio (Mo), selênio (Se), potássio (K), chumbo (Pb), cromo (Cr), cádmio (Cd), mercúrio (Hg) entre outros (Branca et al., 2020).

Atualmente, o uso das escórias siderúrgicas vem crescendo em diversos países ao redor do mundo. A taxa de reaproveitamento é expressiva, atingindo 98% no Japão, 85% na Europa, 80% nos EUA em 2018 (O'CONNOR *et al.*, 2021). No Brasil, a destinação dos agregados de aciaria é pouco diversificada, demonstrando o pequeno reaproveitamento e baixa valorização destes materiais. Em 2020, do montante gerado, 23% foram vendidos, 27% doados, 13% reciclados internamente nas usinas, 1% teve disposição final em aterros industriais e o estoque passivo representou 36% (INSTITUTO AÇO BRASIL, 2021). Esses números revelam a necessidade de desenvolvimento de tecnologias e incentivo para aplicação desses resíduos, resultando em menor impacto ambiental como poluição, ocupação de terras e a inutilização dos estoques passivos nos pátios das usinas.

1.2 Escória de aciaria de forno elétrico a arco

O subproduto da transformação do minério de ferro e/ou sucata de aço em aço na rota de produção em forno elétrico a arco (FEA) é denominado como escória de aço ou escória de aciaria e compreende um material complexo originado pela adição de fundentes, sendo os mais comuns a cal ($CaCO_3$) e cal dolomítica [$Ca\text{-}Mg(CO_3)_2$], que extraem as impurezas do ferro e aço fundidos no decorrer da fabricação deste (ALMEIDA *et al.*, 2017; YANG et al., 2018).

As características físico-químicas desse subproduto dependem das impurezas químicas originais no minério de ferro, da sucata, da natureza dos fundentes e do ambiente químico sob o qual a escória é produzida (HOSSEINI *et al.*, 2016). Sendo esses parâmetros que determinam a condutividade elétrica, área superficial, porosidade, pH, variedade e teores de elementos químicos.

A composição química primária das escórias de aciaria são óxidos de ferro (FeO e Fe_2O_3), sílica (SiO_2) e aluminia (Al_2O_3), que se originam do minério de ferro ou sucata, e óxidos de cálcio (CaO) e de magnésio (MgO) originários da cal ou dolomita utilizada e pH básico (± 12,0) (CAI *et al.*, 2022b). Grandes quantidades do macronutriente fósforo (P) e micronutrientes estão presentes, com destaque para ferro (Fe), enxofre (S), manganês (Mn), cobre (Cu), zinco (Zn), molibdênio (Mo), silício (Si) além de Ca e Mg (YILDIRIM; PREZZI, 2011). Metais pesados como arsênico (As), cromo (Cr), chumbo (Pb), cádmio (Cd), mercúrio (Hg) e flúor (F) presentes nos insumos podem ser, eventualmente, transferidos para a escória siderurgica (WANG *et al.*, 2021).

Essas características associadas ao baixo custo deste material vêm acarretando em maior utilização das escórias de aciaria por diferentes segmentos (SHU; SASAKI, 2022). Em maior escala na construção civil como aditivos de cimento (KURNIATI et al., 2023; GUO et al., 2018), na construção de pavimentação e rodovias (KUMAR; SHUKLA, 2023), na construção hidráulica e em materiais para tratamento de águas residuais ou gases (YI *et al.*, 2012).

O uso agronômico das escórias siderúrgicas, incluindo a escória de aciaria, teve seu início em 1878 na Europa e logo se difundiu como um recurso valioso que resultava em benefícios nutricionais às plantas e à qualidade do solo, aumentando o rendimento das culturas (CHAND et al., 2015).

Por outro viés, a escória de aciaria apresenta potencial para a remediação de solos contaminados por metais potencialmente tóxicos como cobalto, mercúrio, cádmio e chumbo (DÍAZ-PILONETA *et al.*, 2022; NING *et al.*, 2016). A escória atua como agente imobilizador de íons de metais pesados para solos ácidos e contaminados com estes elementos. A escória de aciaria é solta e porosa, e as principais fases minerais são silicato de cálcio, ferrita de cálcio e óxidos alcalinos. Estas fases minerais podem não apenas formar precipitados de hidróxido entre os grupos OH^- produzidos pela hidrólise da substância alcalina com íons de metais pesados mas também gerar silicatos e ferritas de metais pesados estáveis por troca iônica entre Ca^{2+} no silicato de cálcio e na ferrita de cálcio e outros íons de metais pesados, removendo assim efetivamente os íons de metais pesados da solução contaminada (CHEN *et al.*, 2019).

As escórias de aciaria também são úteis na redução da emissão de gases de efeito estufa pela presença de Fe, suprimindo a metanogênese (GWON *et al.*, 2018), como corretivos da acidez de solos (CASTRO; CRUSCIOL, 2013; HAYNES; ZHOU, 2018; ISLAM *et al.*, 2022) e como fertilizantes agrícolas (PIATAK et al., 2015; OZA *et al.*, 2018; DAS *et al.*, 2020; SILVA *et al.*, 2021).

No solo, a ação do silicato (SiO_3^{2-}) ao reagir com a água (H_2O), libera íons OH^- neutralizando o H+ e o Al^{3+} presente na solução do solo, consequentemente, ocorre a elevação do pH e dos teores de cálcio (Ca), magnésio (Mg) e saturação por bases (V%), com diminuição da acidez potencial (H+Al) (HOSSEINI *et al.*, 2016; GAO *et al.*, 2016) confere maior qualidade ao solo e disponbilidade de cargas. As escórias de aciaria são fontes de silício (Si) e os teores de micronutrientes como Cu, Zn, B, Mo atendem às demandas vegetais (ITO, 2015). Além disso os óxidos de Fe e Mn intensificam a produção de ácidos húmicos que auxiliam no armazenamento de nutrientes, no crescimento vegetal e na agregação do solo (TU *et al.*, 2017). A nutrição vegetal também é favorecida pela disponibilização de fósforo, visto que os íons SiO_3^{2-} saturam os sítios de adsorção pelo fosfato nos sesquióxidos de Fe e Al (PRADO; FERNANDES, 2001).

Além de liberar nutrientes às plantas, as escórias também fornecem nutrientes aos microrganismos rizosféricos promotores de crescimento vegetal, favorecendo o crescimento da microbiota, (GWON *et al.*, 2018). Acréscimos de cerca de 25% na taxa fotossintética observadas em plantas fertilizadas com escória de aciaria acarretam maior exsudação radicular que por sua vez impacta positivamente o crescimento e a atividade microbiana no solo (DAS *et al.*, 2019).

Os fertilizantes de escória devem fornecer os nutrientes necessários sem causar efeitos negativos ao ambiente nem à saúde humana, animal e vegetal. A presença de contaminantes metálicos tóxicos como cromo (Cr), cádmio (Cd), chumbo (Pb), vanádio (V), potenciais poluentes do meio ambiente, representa um desafio ao uso extensivo deste resíduo como fertilizante (ITO, 2015). Uma vez no solo estes elementos potencialmente tóxicos não são biodegradáveis e em concentrações elevadas podem ser absorvidos pelas plantas, influenciando o metabolismo e causando redução no crescimento e desenvolvimento vegetal (JING et al., 2007; RAJKUMAR & FREITAS, 2008). No entanto e as concentrações de metais pesados aparentam não ser suficientes para representar riscos ambientais (GWON et al., 2018).

Estudos demonstram efeitos positivos da aplicação de escória siderúrgica sobre a qualidade do solo e o crescimento vegetal. O aumento na absorção de macro e micronutrientes como N, P, K, Ca, Mg, Si, Fe, Mn e Zn ao mesmo tempo em que os teores de metais pesados como Cd, Cr, Cu, Pb, Ni são reduzidos nos tecidos vegetais e/ou indisponíveis no solo são frequentemente descritas em arroz (HE *et al.*, 2016; MATSUMOTO *et al.*, 2015), pimenta (OZA *et al.*, 2018), trigo (WHITE *et al.*, 2017), milho (HU *et al.*, 2019), cana-de-açucar (SOBRAL *et al.*, 2011).

Ainda é escassa a literatura que destaque os os efeitos da fertilização com escória de aciaria ao longo do tempo. Alguns estudos demonstram que a contaminação do solo por metais pesados e a absorção pelas plantas não é aumentada com o uso desse tipo de fertilizante em curto prazo (ALI; OH; KIM, 2008; GWON *et al.*, 2018). A longo prazo os resultados são contraditórios e divergentes para diferentes metais (HILTUNEN, R., AND HILTUNEN, 2004; KUHN et al., 2006) e carecem de estudos amplos para uma melhor compreensão dos efeitos sobre diversos tipos de solo e práticas agronômicas aliadas a utilização de escória siderugica no manejo do solo.

1.3 Rizobactérias promotoras de crescimento de plantas

As rizobactérias promotoras de crescimento de plantas - PGPR (*Plant Growth Promoting Rhizobacteria*), foram primeiramente descritas por Kloepper e Schroth (1978). Sendo representado por um diversificado grupo de bactérias de importantes gêneros como *Azotobacter, Serratia*, *Azospirillum*, *Bacillus, Chromobacterium, Agrobacterium, Erwinia, Pseudomonas, Burkholderia, Bradyrhizobium* e *Rhizobium*, que habitam a rizosfera, capazes de colonizar o sistema radicular das plantas e realizar uma gama de funções que resultam em melhor crescimento e desenvolvimento da planta, com maior competência comparadas às bactérias do solo não rizosférico (OLEŃSKA *et al.*, 2020).

Considerando a interação estabelecida com a planta, as PGPR podem ser extracelulares (de vida livre), encontradas na rizosfera, no rizoplano e nos espaços intercelulares superficiais do córtex radicular, como *Pseudomonas, Bacillus, Erwinia*, ou intracelulares (simbióticas) quando encontradas no interior de estruturas especializadas ,os nódulos localizados nas raízes, como *Rhizobium* e *Bradyrhizobium* (MARTÍNEZ-VIVEROS *et al.*, 2010).

Em 1950, com os estudos iniciais sobre bactérias fixadoras de nitrogênio, inúmeras PGPR foram descritas com aplicações biotecnológicas na agricultura, horticultura, silvicultura e proteção ambiental (RODRÍGUEZ-DÍAZ et al., 2008; SANTOS; OLIVARES, 2022) e embora o modo de ação das PGPR ainda não seja totalmente esclarecido, sabe-se que envolve mecanismos diretos e indiretos. De forma direta, as rizobactérias estimulam o crescimento das plantas através da fixação de nitrogênio, solubilização de nutrientes (principalmente ferro, enxofre e fosfatos inorgânicos e insolúveis), produção de fitohormônios (ácido indol acético (AIA), giberelinas e citocininas) e exopolissacarídeos (ELSAYED *et al.*, 2022; GROBELAK *et al.*, 2018; KHODAVERDILOO *et al.*, 2020; MEENA *et al.*, 2017b).

A fixação biológica de nitrogênio é realizada exclusivamente por procariotos, os quais transformam o nitrogênio atmosférico em amônia, usando um sistema enzimático complexo conhecido como nitrogenase (SINGH *et al.*, 2015). Já os principais mecanismos de solubilização de fosfato incluem a liberação de compostos complexantes ou dissolventes como ácidos orgânicos, íons H^+ e OH^- que alteram os valores de pH rizosférico modificando a disponibilidade dos íons, a liberação de enzimas extracelulares (solubilização bioquímica) e a liberação de fosfato oriundo da decomposição da matéria orgânica (AHKAMI *et al.*, 2017).

A produção bacteriana de substâncias complexantes de Fe, denominadas sideróforos, auxilia na assimilação deste micronutriente que se encontra em maior quantidade na forma Fe^{3+}, pouco solúvel aos vegetais (PII *et al.*, 2015). Os exopolissacarídeos (EPS) são importantes na formação de biofílmes que auxiliam na colonização das raízes pelas rizobactérias, promovem um ambiente favorável a circulação de nutrientes, protegem as plantas sob estresse hídrico, salino e tóxico, bem como contra o ataque de fitopatógenos, através de moléculas sinalizadoras de resposta de defesa no momento da infeção (PANDIT *et al.*, 2020).

Após a facilitação nutricional descrita nos mecanismos acima, a produção de fitormônios vem sendo considerada o mais importante modo de ação na promoção de crescimento de plantas por rizobactérias. Hormônios como AIA (ácido 3-indolacético), citocininas, giberelinas produzidos por PGPR podem alterar a proliferação celular, o desenvolvimento da parte aérea e radicular, a arquitetura da raiz, com maior produção de raízes laterais e pelos radiculares, culminando com aumento da captação de nutrientes e água e, consequentemente, melhorando o crescimento da planta (JI; GURURANI; CHUN, 2014; LIMA *et al.*, 2021; PERALTA *et al.*, 2012).

A modulação hormonal, principalmente sobre os níveis de AIA, estabelecida após a interação *planta x PGPR* também pode promover modificações anatômicas em diferentes órgãos do vegetal, como mesofilo foliar e raiz (AGARWAL *et al.*, 2019). Aumentos de área foliar relacionados aos maiores níveis de auxina, resultam em maior crescimento da parte aérea (folhas) para captação de luz, associado a maior condutância estomática que pode resultar na elevação da taxa fotossintética. Esses efeitos corroboram como as alterações no mesolfilo foliar, como aumento da espessura de parênquima paliçadico, aumento do número de estômatos bem como suas aberturas, como observado em fotínea (*Photinia x fraseri*) inoculada com *Azospirillum brasilense* e *Azotobacter chroococcum* (LARRABURU; APÓSTOLO; LLORENTE, 2010).

As alterações na arquitetura radicular decorrentes da inoculação de PGPR também podem vir junto a alterações anatômicas. Aumentos de diâmetro radicular, número de vasos de xilema ou ampliação do lumem são associados a alterações positivas na condutância hidráulica da planta (PAN *et al.*, 2023). PGPR estimulam a expansão do cortex, a espessura da exoderme, ampliam aerênquima e dessa forma conferir níveis adequados de nutrientes, especialmente K^{+}, Ca^{2+} e Mg^{2+}, redução na perda de água e maior resistência a escassez de água, como observado em arroz de terras altas inoculado com isolados de *Trichoderma, Pseudomonas* e *Bacillus* (RÊGO *et al.,* 2014),

De forma indireta, PGPR podem reduzir os danos causados por fitopatógenos através da produção de sideróforos, quitinases, glucanases e antibióticos (FILIPPI *et al.*, 2011; GOSWAMI; DEKA, 2020) e a mitigação de efeitos negativos da presença de metais tóxicos (e.g. Pb, e Cr) e xenobióticos como diesel e piretróides (ETESAMI, 2020; MEENA *et al.*, 2017a).

Embora os elementos potencialmente tóxicos (EPT`s) no solo possam afetar negativamente a respiração, o metabolismo e as atividades das comunidades microbianas, muito microrganismos apresentam genes plasmidiais como *cadCA, ars, pco, sil* e que conferem resistência a metais pesados (NJOKU; AKINYEDE; OBIDI, 2020). (NJOKU; AKINYEDE; OBIDI, 2020). Esses genes são relacionados com sistemas de bombas quimiosmóticas de íons/prótons e ATPases, responsáveis pelo efluxo do metal, como observados para a resistência de As, Pb, Cd e Cr em muitas bactérias (AHEMAD; KIBRET, 2014; VERMA; KUILA, 2019).

As PGPR podem imobilizar, ligar, oxidar, transformar e volatilizar metais no solo (MA *et al.*, 2016; PISHCHIK *et al.*, 2016; VERMA; KUILA, 2019) através de mecanismos que incluem sequestro físico, exclusão e complexação ou desintoxicação que podem gerar formas metálicas mais estáveis e menos móveis, podem precipitar ou ainda quelar os metais. Esses mecanismos imobilizam os metais na rizosfera, diminuindo a disponibilidade para absorção pelas plantas, resultando na redução da fitotoxicidade (RAJKUMA et al., 2013; MA, Y. *et al.*, 2011; RAJKUMAR & FREITAS, 2008).

As rizobactérias desempenham mecanismos para mitigação de estresse químico que muitas vezes são acompanhados de promoção de crescimento da planta (DHALI *et al.*, 2022; VOCCIANTE *et al.*, 2022; WANG *et al.*, 2023). A redução da biodisponibilidade de metais pode ocorrer através da produção de EPS e substâncias quelantes como sideróforos e ácidos orgânicos (LI *et al.*, 2023; SHARMA, 2022; WANG, XIAOHAN *et al.*, 2017; ISLAM, FAISAL *et al.*, 2014), através de enzimas bacterianas que desintoxicam o meio através da precipitação e/ou metilação dos metais (SHIN *et al.*, 2012) além da biosorção e bioacumulação de metais através do sequestro extracelular e acumulação intracelular, como realizado por *Bacillus* em *Alnus firma* (MA *et al.*, 2016).

A redução da absorção e posterior translocação de metais ou a redução dos efeitos tóxicos do metal absorvido, associados a promoção de crescimento por PGPR são comprovados em diferentes culturas. Isolados de *Alcaligenes fecalis* e *Bacillus cereus* reduziram a toxicidez de Cd, Cu, Zn e Pb em sorgo com melhoria em altura e biomassa das plantas (EL-MEIHY *et al.*, 2019), *Bacillus subtilis* promoveu ganhos de biomassa em repolho cultivado em altos teores de Cd (GE *et al.*, 2022), *Methylobacterium oryzae* e *Burkholderia* sp. reduziram os efeitos de Ni(II) e Cd(II) em tomate (*Lycopersicon esculentum*), com aumento significativo nos atributos de crescimento das plantas em comparação com o controle não tratado (MADHAIYAN; POONGUZHALI; SA, 2007). *Bacillus* sp. melhorou os atributos fisiológicos e de crescimento em grão de bico (WANI; KHAN, 2010) e milho (AFZAL *et al.*, 2020), na presença de Cr e Pb, respectivamente.

O conhecimento existente até o presente momento subsidia e dissemina para utilização tecnológica de microrganismos promotores de crescimento de plantas e as rizobactérias que possuem vários mecanismos para a promoção de crescimento são desejáveis (multicompetentes), visando produtos que melhorem a produção agrícola e contribuam para a proteção ambiental, de forma mais econômica e sustentável.

1.4 A eucaliptocultura e o gênero *Corymbia*

O setor de florestas plantadas no Brasil ocupa lugar de destaque na economia nacional, alcançando 9,5 milhões de hectares plantados e movimentando R$ 30,1 bilhões em 2021 (INDÚSTRIA

BRASILEIRA DE ÁRVORES, 2022). De acordo com o IBGE (2022), a eucaliptocultura representou 77% da área plantada, abrangendo todas as regiões geográficas do país, com predomínio nos estados de Minas Gerais, São Paulo e Mato Grosso do Sul, colocando o Brasil na posição de líder mundial na produtividade da espécie.

A produção de eucalipto no Brasil atende a alta demanda de madeira requerida pelas indústrias de papel e celulose, carvão vegetal e lenha, prioritariamente (IBGE, 2022). Ao longo de aproximadamente 50 anos de pesquisas com eucalipto, muitos progressos foram realizados com o aprimoramento de técnicas de cultivo como programas de melhoramento genético e clonagem que proporcionaram avanços em adaptação fisiológica, resistência a doenças e pragas, melhorias de densidade e produtividade de madeira (ASSIS; ABAD; AGUIAR, 2015).

Espécies como *Eucalyptus urophylla, Eucalyptus grandis, Eucalyptus saligna, Eucalyptus camaldulensis, Eucalyptus viminalis* e seus híbridos representam os materiais genéticos mais cultivados no país (REIS *et al.*, 2014). Todavia a expansão da cultura para áreas geográficas não tradicionais e a busca por melhorias na qualidade da madeira e dos produtos gerados, tem ampliado o horizonte para o uso de espécies pouco exploradas (CUNHA *et al.*, 2021).

Nesse cenário, espécies do gênero *Corymbia* e alguns de seus híbridos interespecíficos vêm sendo consideradas matéria prima para usinas siderúrgicas, na fabricação de carvão. O gênero *Corymbia (Myrtaceae*) compreende 113 espécies, as quais eram pertencentes ao gênero *Eucalyptus* até 1990. A diferenciação botânica foi realizada em relação às pétalas e sépalas de suas flores. Nas espécies de *Eucalyptus* elas são fundidas e um ou dois opérculos cobrem os estames e o ovário. Nos representantes de *Corymbia*, o desenvolvimento das flores demonstra que a evolução do opérculo ocorreu independente do que ocorre em *Eucalyptus* (ROZEFELDS, 1996).

As espécies de *Corymbia* apresentam alta densidade básica (acima de 600 kgm^{-3}), elevados teores de carboidratos e extrativos e seu uso é adequado à construção civil, movelaria, produção de carvão e geração de energia e sua madeira é considerada de alta qualidade e durabilidade (SEGURA, 2015). No geral apresentam resistência a pragas e doenças, bem como à distúrbios fisiológicos, com ênfase ao estresse hídrico (DE ARAUJO *et al.*, 2021; LEE, 2013).

As espécies tradicionais desse gênero, plantadas ou introduzidas no Brasil, são *Corymbia citriodora*, *Corymbia maculata* e *Corymbia torelliana.* O sucesso da exploração comercial dessas espécies está intimamente relacionado a obtenção de híbridos, cruzamentos entre esses indivíduos que resulta em heterose, um fenômeno muito comum nesse grupo. Os híbridos apresentam qualidades biológicas superiores aos genitores, produzindo ganhos substanciais em um curto espaço de tempo, como maior produtividade, maior resistência e precocidade (ASSIS; ABAD; AGUIAR, 2015).

Por outro lado, os representantes do gênero apresentam baixa receptividade à propagação vegetativa, com baixas taxas de enraizamento, inferiores a 5% em *C. citriodora* (REIS *et al.*, 2013) com plantios sendo tradicionalmente estabelecidos por via seminal (SHEPHERD *et al.*, 2008). A espécie *C. torelliana* apresenta melhor capacidade de enraizamento, cerca de 30% maior em relação a outros representantes de *Corymbia* e o uso desta espécie na composição de híbridos interespecíficos dentro do gênero é bastante explorado, pois vários clones destes são enraizáveis pelo efeito positivo da participação materna de *C. torelliana* (ASSIS, 2000). Esta técnica tem ajudado substancialmente, porém a taxa de enraizamento é variável e ainda baixa, variando de 16,5% para clones *C. citriodora* x *C. torelliana* a 43,6% para clones *C. torelliana* x *C. citriodora* (ASSIS, 2014).

Nos últimos anos houve aumento das pesquisas no Brasil sobre a propagação de espécies de *Corymbia* e seus híbridos (AVELAR *et al.*, 2020; TRUEMAN, 2018) mas ainda hoje ocorrem variações na capacidade rizogênica com formação de mudas com baixa qualidade de sistema radicular na propagação vegetativa deste grupo (ABIRI *et al.*, 2020).

Desse modo, a utilização de rizobactérias promotoras de crescimento surgiu como um recurso valioso em face das possibilidades de incremento no índice de enraizamento, no crescimento e no controle biológico de doenças (MAFIA, 2004) com estudos demonstrando que mudas produzidas com rizobactérias podem apresentar maior taxa de sobrevivência, em razão da melhoria da qualidade do sistema radicular, além da possibilidade de redução de doenças nos primeiros anos (GONÇALVES MAFIA *et al.*, 2009; MAFIA *et al.*, 2009; MAFIA; ALFENAS; FERREIRA; *et al.*, 2007; TEIXEIRA; ALFENAS; *et al.*, 2007).

As alterações sobre o sistema radicular decorrentes da interação planta-PGPR podem ser observadas sobre a arquitetura radicular com aumento do comprimento da raiz principal e raízes laterais, no número e posicionamento de raízes laterais e no estímulo ao desenvolvimento de raízes adventícias em espécies propagadas vegetativamente (BELLINI; PACURAR; PERRONE, 2014; LÓPEZ-BUCIO *et al.*, 2007), que por sua vez estão relacionados com a produção de enzimas e fatores de crescimento, principalmente auxinas como o ácido 3-indolacético bacterianos ou com o aumento da fixação de N e solubilização de P (CHOUDHARY *et al.*, 2022; PAZ *et al.*, 2012).

Aumento na emissão de raízes com melhoria global do sistema radicular e crescimento de mudas de *Eucalyptus* foram observados com a inoculação da *Bacillus subtilis* (PAZ *et al.*, 2012; RAASCH; BONALDO; OLIVEIRA, 2013), *Burkholderia* e *Azotobacter* (ENEBAK et al., 1998; TEIXEIRA; COUTO ALFENAS; *et al.*, 2007), refletindo em maior ganho de biomassa de parte aérea, conferindo maior vigor às plantas.

Poucos trabalhos abordam a aplicação de PGPR na promoção de crescimento de *Corymbia*. Recentemente, o efeito da inoculação de sementes de *C. citriodora* e de estacas de um híbrido de *C.*

citriodora x *C. torelliana* com as bactérias endofíticas *Priestia megaterium*, *Exiguobacterium sibiricum*, *Pantoea vagans* foi avaliado e incrementos sobre altura e massa seca, diâmetro de coleto, teores de clorofila, além de boa colonização rizosférica foram obtidos (OLIVEIRA, 2023). O mesmo autor sugere a interação entre PGPR e auxina sintética (ácido indol burítico) sobre o crescimento de estacas de um híbrido de *C. citriodora* x *C. torelliana* e verificou resposta sinérgica positiva da associação das rizobactérias e 1.000 mg kg^{-1} de AIB sobre o desenvolvimento das mudas (OLIVEIRA, 2023).

Ainda não existe um inoculante para *Corymbia* disponível no mercado e considerando a importância da espécie e seus hídridos para o setor florestal brasileiro, investigar o potencial de PGPR para a promoção do crescimento e obtenção de mudas mais robustas, representa um avanço com reflexos ecológicos, econômicos e sociais.

O conjunto de informações expostas até aqui sustentam a proposta para subsidiar uma estratégia de economia circular, através da reciclagem de escória siderúrgica, contribuindo para a redução do passivo ambiental e somada ao emprego da tecnologia microbiana para mitigar possíveis efeitos negativos e melhorar a qualidade final das mudas de *Corymbia*. Portanto, o objetivo geral deste trabalho foi avaliar os efeitos da fertilização com escória de aciaria de forno elétrico à arco combinado à inoculação de *Bacillus* sp., sobre o crescimento de mudas de *Corymbia* sp.

2 INOCULATION OF *Bacillus* sp. IMPROVES ROOT ARCHITECTURE, GAS EXCHANGE AND EFFICIENCY OF NUTRIENT USE IN *Corymbia* SEEDLINGS

ABSTRACT

The inoculation of forest species with rhizobacteria growth promoters is a technology increasingly capable of improving growth and also final quality, obtaining more robust and resistant seedlings. The objective was to evaluate growth, root architecture, gas exchange, chlorophyll a fluorescence, chlorophyll content and nutrient use efficiency in seedlings of *Corymbia* inoculated with the growth promoting rhizobacteria *Bacillus* sp. CCMD862. The growth promotion experiment was carried out in greenhouse, using hybrid clonal seedlings of *Corymbia torelliana* x *Corymbia citriodora.* The experimental design was completely randomized with 2 treatments and 5 replicates per treatment (*Bacillus* sp. CCMD862 and Control - not inoculated). The inoculation improved efficiency in gas exchange, chlorophyll fluorescence, nutrient use, biomass variables and in all root architecture parameters in relation to control. Biometric and biomass improvements resulted in higher Dickson quality index (DQI) and a global growth promotion rate 30% higher than control plants. The strain *Bacillus* sp. CCMD862 is able to biostimulate seedlings of *Corymbia* favoring the development of the root system, gains in the photosynthetic apparatus, increases in absorption and efficient use of nutrients that resulted in robust seedlings. The future development of an inoculant could aid rooting in *Corymbia*, helping to reduce mortality in the field and obtain homogeneous commercial plantations.

Keywords: Eucaliptus; Rooting; *Bacillus*; Auxin; Photosynthesis.

2.1 Introduction

The planted forests of eucalyptus occupy 7.53 million hectares distributed in all geographic regions of the country, attributing to Brazil the position of largest producer of the species worldwide, with productivity of 38.9 m^3/ha/year, moving R$ 30.1 billion in 2021 in the Brazilian trade balance (Brazilian Tree Industry, 2022).

The characterization and identification of some representatives of the genus Eucalyptus underwent changes from morphological studies led by Hill and Johnson (Hill and Johnson, 1995), culminating in the creation of the genus *Corymbia*, currently with 113 species as *C. citriodora*, *C. maculata*, *C. torelliana*. *Corymbia* species have aroused increasing economic interest, due to lower costs of wood production by volume compared to the genus *Eucalyptus*, because they have greater tolerance to biotic and abiotic stresses, in addition to higher wood density, important for coal production (SOUZA et al., 2020).

A challenge faced in the establishment of commercial plantations with *Corymbia* species is the recalcitrance to rooting, which can be defined as the low capacity of mini-cuttings to form adventitious roots from non-specialized tissues during clonal propagation (de Lima et al., 2022) The development of hybrids such as *C. torelliana x C. citriodora* that have a good percentage of rooting has been explored, but significant variability in the pattern of development of clonal seedlings is observed, growth of the root system, which impacts the final quality of the seedlings as well as adaptability in different environmental conditions of the field (Peralta et al., 2012; Filho et al., 2018; Wendling et al., 2021).

Plant growth promoting rhizobacteria (PGPR) can act freely or associated with plant roots (Kaushal et al., 2023; Glazebrook & Roby, 2018) promoting increases in biomass and root architecture, optimizing the absorption of water and nutrients, improving photosynthetic performance and energy conversion and also increasing stress tolerance (Abdelaal et al., 2021), providing significant improvements in plant species development. The use of PGPR is very promising, with satisfactory results on growth, with increase of fine roots or increase of average diameter of the root system, root and shoot dry matter increase in seedlings of *Eucalyptus* and *Corymbia* with the inoculation of bacteria of the genera *Bacillus, Pseudomonas, Azotobacter, Azospirillum*, among others (Mafia et al., 2009; Teixeira et al., 2007; Zarpelon et al., 2016; González-Díaz et al., 2019).

Even with so many benefits of inoculation recognized and the importance of species of the genus *Corymbia*, the number of inoculants for species of this genus is still negligible. Isolating growth promoting microorganisms with high performance in *Corymbia* species will allow the obtaining of more vigorous and resistant clonal seedlings to transplanting, contributing to greater survival in the field, which may reflect productivity gains and reduction of production costs.

Therefore, the objective of this study was to prospect isolates of PGPR, track the biochemical mechanisms of plant growth promotion presented by isolates and evaluate the effects of inoculation on gas exchange and fluorescence of chlorophyll *a*, chlorophyll content, root architecture, nutrient use efficiency and growth of *C. torelliana* x *C. citriodora.*

2.2 Material and Methods

2.2.1 Obtaining of microorganisms

Six individuals of *Ricinus communis* (castor bean), a species that occurs spontaneously in an area of steel waste deposit in the state of Pará/Brazil (524'35.3"S 4904'42.6"W), were carefully collected along with the soil adhered to the roots. Immediately after collection, for the isolation of rhizobacteria, 10 grams of rhizosphere soil were suspended in 90 mL of sterile saline solution (0.85% NaCl), kept under agitation for 5 minutes, diluted in series until 10^{-6} and spread in Nutrient Agar - NA (composition g/L: meat extract 1.0; yeast extract 2.0; peptone 5.0; sodium chloride 5.0, agar 15.0 with pH 6.8). To obtain endorrizospheric bacteria, the roots collected were washed in running water, rinsed with deionized water, disinfected superficially according to Tirry et al. (2018). Subsequently, aseptically, 1 g of root was collected, suspended in 10 ml of sterile saline in a test tube and macerated with the help of a sterile glass stick, in order to cause injury and facilitate the exit of bacterial cells to the solution. This suspension was diluted at 10^{-4} and 100 µl of each dilution were sown in NA medium. All plates were incubated at 30 ºC for 48 h. All procedures were performed in triplicate. Phenotypically distinct colonies were purified and characterized for morphology and Gram reaction under immersion optical microscopy and stored in sterile distilled water at room temperature (Castellani, 1939).

2.2.2 Selection of isolates

The selection of growth promoter isolates was performed in rice plants according to Cardoso et al. (2021), in greenhouse. The inoculum was prepared according to Tirry et al. (2018) with the final concentration adjusted to 10^8 cfu mL^{-1} and seed microbiolization was performed as proposed by Filippi et al. (2011). Two tests were performed. Test 1 consisted of 37 treatments (36 rhizobacteria isolates and 1 control, without rhizobacteria) in a completely randomized design, with three replicates for each treatment. Twenty-one days after germination, the plants were evaluated in height, root length, root biomass and total biomass. The data were submitted to analysis of variance and comparison of means using the Scott-Knott test ($p < 0.05$). A cluster analysis generated 4 groups of isolates, using the similarity matrix with standard Euclidean distance. The isolates with better performance in test 1 were again evaluated in test 2 and the results submitted to analysis of variance and comparison of means using the Tukey test ($P < 0.05$). The isolate CCMD862 presented better performance for all variables analyzed compared to the control, therefore, it was submitted to in vitro biochemical tests and selected for the tests with seedlings of *Corymbia*.

2.2.3 Biochemical characterization for plant growth promotion

The rhizobacteria CCMD862 was evaluated for the mechanisms of plant growth promotion. The qualitative tests were performed standardizing the initial concentration of liquid inoculum at 10^8 cfu mL^{-1} in triplicate.

a) Nitrogen fixation: the isolate was sown in nitrogen-free Burk medium, containing (gL^{-1}) 0.52 K_2HPO_4, 0.2 $CaCl_2$, 0.0025 $Na_2MoO_4.2H_2O$, 0.1 $MgSO_4.7H_2O$, 0.005 $FeSO_4.7H_2O$, 0.41 KH_2PO_4, 0.05 Na_2SO_4, 10 glucose and 15 agar (pH 7,3) in 1000ml of distilled water. The plates were incubated at 28ºC for 7 days (Husseiny et al., 2021). The growth was considered positive for nitrogen fixation.

b) Indole-3-Acetic Acid production: the isolate was grown in liquid medium Luria Bertani (LB) under 100 rpm and incubated at 28º C for 78 h. Then, 3 mL of the suspension were centrifuged at 4º C for 10 min at 4.000 rpm, 90 µl of the supernatant and 60 µl of the Salkowski reagent were added to a microtube and incubated in the dark for 30 min to whether a change in the medium color occurred (Gordon & Weber, 1951).

c) Biofilm: biofilm formation capacity was evaluated according to Kavamura & Melo (2014) modified O'Toole & Kolter (1998). The isolate was cultivated in LB medium under 100 rpm agitation and incubated at 28ºC for 24 hours. Then, an aliquot of 100 µl of the culture was incubated again in a microtube with a capacity of 2000 µl, containing 900 µl TSB broth at 1/10 of the force, for 96 hours at 30 ºC. After this period, the bacterial suspensions were manually homogenized with an automatic pipette and evaluated in a spectrophotometer at a wavelength of 600 nm (DO_{600}). Subsequently, the liquid containing the microbial growth was completely discarded and each tube washed three times with sterile distilled water. Clean tubes were added 1000 µl of violet crystal (0.1%), incubated for 15 minutes at room temperature. Then the contents of the tubes were discarded. For the reading, 100 µl of 95% ethanol was added to each tube and the reading of the solution was performed in spectrophotometer under 560nm wavelength (DO_{560}). Since DO_{600} refers to bacterial growth and DO_{560} to biofilm formation, the scale was used in which DO560 ≥ 0.1 confirmed biofilm formation.

d) Phosphate solubilization: The isolate was sown in modified NBRIP culture medium containing (gL^{-1}) 10 of glucose, 2,5 de $Ca_5(OH)(PO_4)_3$, 25 de $MgCl_2.6H_2O$, 0,25 $MgSO_4.7H_2O$, 0,2 KCl e 0,1 $(NH_4)_2SO_4$ (Nautiyal, 1999), pH de 7,0. The plates were incubated for 14 days at 28 C. After this time, the presence of a halo around the colony was considered indicative of phosphate solubilization.

e) Potassium solubilization: The isolate was sown in modified Aleksandrov culture medium containing (gL^{-1}) 5.0 of glucose, 0.5 $MgSO_4$, 0.005 $FeCl_3$, 0.1 $CaCO_3$, 2.0 $Ca_3(PO_4)_2$, 2 of feldspar as source of mineral K, 5 of bromotimol blue and 7,5% of agar. The plates were incubated during 72h to 28 C. After this time, the presence of a yellowish halo around the colony was considered indicative of potassium solubilization (Rajawat et al., 2016).

f) Siderophore production: for the quantification of siderophores by vis/UV spectrophotometry, the CAS solution was used, prepared according to the siderophore detection protocol of Schwyn & Neilands (1987). The isolate was cultivated for five days in LB 2% medium. The absorbance of bacterial growth was evaluated at a λ=600 nm. Then, 0.5 mL of the culture supernatant was suspended in 0.5 mL of the CAS solution, after 30 minutes the absorbance was obtained at the wavelength λ=630 nm. The amount of siderophores was determined by the percentage reduction in the blue color of the solution compared to the control.

2.2.4 Genetic identification of rhizobacteria CCMD862

The rhizobacteria were cultivated in NA culture medium for 24h at 28 ºC. As sample volume, two inoculation loops were added to a microtube containing 1 mL of extraction buffer (Tris-HCl 1x). Then, DNA extraction was performed according to the method described by Mariano and Silveira (2005). The isolates were identified using the 16S rDNA region and the primers 27F (50-AGAGTTTGATCMTGGCTCAG-30) and 1492R (50ACCTTGTTACGACTT-30) (Lane et al., 1985), which amplify 1500 bp. The PCR amplification reaction was composed of 1x Master Mix 2x (Promega) (0.05 U mL 1 Taq DNA polymerase, 4 mM $MgCl_2$ reaction buffer, 0.4 mM of each DNTP (Promega Corporation, Madison, WI, United States), 10 mM of each primer and 20 ng DNA. Amplification of the 16S rDNA region was performed in a thermal cycler (Master Cycler Nexus, Eppendorf, Hamburg, Germany) with the following steps: initial denaturation at 94ºC for 4 minutes; 25 cycles of 94 ºC for 1 min, 55 ºC for 1 min and 72 ºC for 1 min; and a final extension at 72 ºC for 7 min. The reactions were purified using 5 mL of PCR product plus 2 mL Exo-SAP Enzyme (Exonuclease), as recommended by the manufacturer. The sample was purified in thermocycler at 37 ºC for 4 minutes, followed by an incubation period at 80 ºC for 1 minute to inactivate both enzymes irreversibly. After this step, the sample was sent to the company Actgene (https://actgene.com.br).

Analysis of the DNA sequence and assembly of contigs were performed with the aid of the Staden Package (Staden et al., 1998). The isolates were identified based on the comparison of the nucleotide sequences of the 16S rDNA region with the sequences of the isolates type of the genera and species available in the GenBank database (National Center for Biotechnology Information - NCBI), using the BLASTn software (https://www.ncbi.nlm.nih.gov/). Then all sequences were aligned (MEGA7). Bayesian Inference (IB) analysis was performed using Mr. Bayes v. 3.2.6 (Ronquist et al., 2012) implemented in CIPRES (https://www.phylo.org/portal2/home.action) using the best nucleotide substitution model.

The identification of a bacterial isolate using 16S rDNA was used to identify the selected strain with the highest homology strain. Subsequent probabilities were calculated after discarding the first 25% of generations. All trees obtained from individual genes and concatenated by the IB method were visualized through the software Fig Tree 1.4.1 (http://tree.bio.ed.ac.uk/software/figtree).

2.2.5 *Corymbia* seedling growth promotion

The growth promotion of *Corymbia* seedlings was evaluated in greenhouse in September to October 2022, using clonal seedlings of the hybrid *Corymbia torelliana* (F.Muell.) K.D.Hill & L.A.S.Johnson x *Corymbia citriodora* (Hook.) K.D.Hill & L.A.S.Johnson, 40 days old, 24 cm tall and with 6 pairs of leaves in perfect phytosanitary condition, obtained from a commercial nursery. The experimental design was completely randomized with two treatments (control and rhizobacteria *Bacillus* sp. CCMD862) with 5 replications for each treatment.

The substrate used was dystrophic yellow latosol with medium texture (MO: 2,3%; P: 35,0 mg; K: 12,2 cmolc; S:1,0 mg; Ca: 2,2 cmolc; Mg: 0,7 cmolc; B:0,39 mg; Cu: 0,60 mg; Fe: 57 mg; Mn: 9,5 mg; Zn: 0,9 mg; H+Al: 3,0 cmolc; CTC: 5,9 cmolc, all in dm^{-3} / pH 4,8) with pots with a capacity of 1.5 dm^3. The bacterial inoculum was prepared according to the methodology proposed by de Castro et al. (2020), where the bacterial isolate was for 48h at 28ºC, subsequently suspended in sterile distilled water and the concentration adjusted in spectrophotometer to 10^8 cfu mL^{-1}. For inoculation, the seedlings were removed from the tubes and had the apex of the root system sectioned in approximately 1cm, then were inoculated with 10 mL of the bacterial suspension for 30 minutes and immediately transplanted. The control treatment plants received the same volume of sterile distilled water in place of bacterial suspension. The experiment was conducted for 21 days and irrigation was daily and manual to replace losses due to evapotranspiration. The volume of water for irrigation was determined by the second gravimetric method (Klar et al., 1966).

2.2.6 Analysis of gas exchange and photosynthetic parameters

The gas and photosynthetic exchange parameters were estimated in the first or second pair of physiologically mature and fully expanded leaves, from the apex to the base, when the seedlings had 61 days. The photosynthetic rate (*A*), stomatal conductance to water vapor (*Gs*), intercellular concentration of CO_2 ($_{Ci}$) and leaf transpiration rate (*E*) were measured simultaneously to the fluorescence parameters of chlorophyll *a*, using an open system infrared gas analyzer (IRGA - Infrared Gas Analyzer), model LI 6400XT (LI-COR, Lincoln, NE, USA), with a Fluorometer (LI-6400-40, LI-COR Inc.) in the range between 10 and 12h, defined in preliminary analyses under the same experimental conditions. The environmental conditions in the IRGA chamber were maintained at 25 ºC and the vapor pressure deficit between 1.2 and 1.8 kPa, the reference CO_2 concentration of 400 μmol $mol^{-1}$of air and active radiation (PAR) of 1000 μmol of photons $m^{-2}s^{-1}$. The amount of blue light applied was 10% of photosynthetic flux density to maximize stomatal aperture. The environmental conditions inside the greenhouse during the measurements were the air temperature of 34 ± 2 ºC, relative humidity of 53 ± 2%, incident radiation of 680 ± 100 μmol m^{-2} s^{-1} and vapor pressure deficit of 2.1 ± 0.14 KPa. The water use efficiency (*WUE*) was determined by the ratio between total dry mass and water consumption throughout the experiment.

For the analysis of chlorophyll fluorescence the leaves were adapted to dark for 20 minutes and then illuminated with a weak light and modulated light pulse (0.03 µmol m^{-2} s^{-1}) to obtain the initial fluorescence value (*Fo*). A saturating white light pulse of 8000 µmol m^{-2} $s^{-1}$was applied for 0.8 s to ensure maximum fluorescence emission (*Fm*). The sampled leaves were then saturated for 300s with actinic light of 250 µmol m^{-2} s^{-1} to achieve steady state fluorescence (*Fs*). Subsequently, white light saturation pulses were applied to achieve maximum fluorescence (*Fm'*). Actinic light was then turned off, and a far red illumination (2 µmol m^{-2} s^{-1}) was applied to estimate the initial fluorescence adapted to light (*Fo*). From these measurements, the following parameters were calculated: potential activity of *PSII* (Fv/Fo = (Fm Fo)/Fo), electron transport rate (*ETR* = φPSII.PPFD. 0.42), photochemical dissipation coefficients (*Qp* = (Fm Fs)/(Fm Fo)), non-photochemical (*Qn*= 1-Qp) and alternative non-photochemical ((*NPQ*= (Fm - Fm`)/Fm`) (MAXWELL; JOHNSON, 2000).

2.2.7 Chlorophylls contents

The chlorophyll content was quantified with the same leaf used in the gas exchange readings. The collection was performed using foil envelopes and instant freezing in N-liquid and packaged in freezer -20 ºC until the moment of analysis. The samples were macerated in mortar with N-liquid, 5mg of fresh mass were suspended in 250uL of 95% ethanol, stirred in vortex quickly, incubated at 80 ºC for 20 minutes, centrifuged at 13000 rpm, at 4 ºC for 5 minutes, then the supernatant was carefully collected and packed in a new microtube. This procedure was repeated twice more, with 80% ethanol and 50% ethanol (Fernie et al., 2001). Soon after extraction, 50uL of the ethanolic extract was diluted in 120uL of 95% ethanol. Absorbance readings were performed at 645 and 665nm and concentrations of *Chla*, *Chlb* and *Chla + b* were estimated according to Porra et al. (1989), using the UV-VIS spectrophotometer.

2.2.8 Biometrics and Biomass

Growth promotion was evaluated by biometric measurements and biomass accumulation at 21 days after the first biostimulation of *Corymbia* seedlings. Plant height (H) and root length were measured with graduated metal ruler, while the diameter of the stem (DC) was obtained with digital caliper (accuracy 0.02 mm). The number of leaves was evaluated by direct counting of the leaves emitted. The leaf area was estimated with leaf area meter (LICOR-3100) with accuracy of 0.1mm^2. The root volume was measured by the gravimetric method of the beaker (Rondina et al., 2020). The seedlings were sectioned in root and shoot and subjected to drying in an oven at 50 ºC, until constant weight and then weighed for determination of root dry biomass (RDB), shoot (SDB) and total dry biomass (TDB). The Dickson quality index (DQI) was determined by Dickson et al. (1960).

The growth promotion rate (GP) promoted by the biostimulant was estimated through the formula GP = ([TDBbio] - [TDBc])/[TDBc], where: TDBbio: total dry mass of the biostimulated plants; TDBc: total dry mass of the control plants.

2.2.9 Root architecture

The root system architecture of the seedlings was evaluated in the software WinRHIZO Pro 2007a (Régent Instrum. Quebec, Canada), coupled to a professional Epson XL 10000 scanner equipped with additional light unit (TPU). A definition of 600 dpi was used for the measurements. The roots were transferred from the ethanolic solution 30% to an acrylic tray 30 cm wide and 40 cm long containing water. It was possible to obtain images in grayscale, based on a method of skeletonization in three dimensions (width, height and depth) being avoided the maximum overlap of the roots. The system was used to obtain the following variables of the root system: total length, total surface area, volume, average diameter, number of tips as well as length, surface area and volume in four diameter classes (0-1, 1.1-2, 2.1-3, >3.0 mm).

2.2.10 Foliar nutrient content and nutrient use efficiency

After drying, the shoots of the plants were ground (< 5 mm) in a Wiley mill and sent to the laboratory to determine the content of macro and micronutrients (Malavolta; Vitti; Oliveira, 1997). The proportions of nutrients obtained were converted into nutrient content as a function of the dry biomass of the shoot. Nutrient use efficiency (*NUE*) was determined according to Siddiqi & Glass

(1981) Nutrient use efficiency (NUE) was determined according to Siddiqi & Glass (1981) where NUE= (total dry mass produced)^2/(shoot content); in g^2mg^{-1} for macronutrients and kg^2g^{-1} for micronutrientes.

2.2.11 Statistical analysis

The data were submitted to analysis of normality and homogeneity of variances, and when significant, the means of treatments were compared by unpaired Student's t-test ($P < 0.05$) using the software RStudio version 2.4.2.

2.3 Results

2.3.1 Obtaining isolates and screening for growth promotion

Thirty-six isolates phenotypically distinct from the rhizosphere of *R. communis* were purified, being 21 from the rhizosphere soil and 15 endorrizospheres. Gram-positive bacteria were predominant in both investigated regions, representing 80% of the rhizosphere isolates and 66% of the endorrizosphere. Regarding morphology, 52% of coconuts and 80% of rods were predominant in the rhizosphere and endorrizosphere, respectively.

The rice plants used as a model for rhizobacteria selection showed that of the 36 isolates evaluated, the isolate CCMD862 presented the best performance for the variables of length and biomass compared to the other treatments (Table 1) increasing by 55% the plant height, by 70% the root length, by 76% the root dry biomass and 67% the total dry biomass of the plant compared to the control not inoculated, being selected to evaluate the growth promotion of *Corymbia* seedlings.

Table 1. Rice growth promotion with bacteria isolated from the rhizosphere and castor bean endorrizosphere. Height (h), root length (CR), root dry biomass and total biomass at 21 days after inoculation

Test 1*					
Isolated	Groups	Growth (cm)		Dry Biomass (mg)	
		h	CR	Root	Total
CCMD862, 878, 867	4	11.74 A	69.3 B	442 A	583 A
875, 863, 866, 868, 880, 881	3	9.77 B	91.3 A	320 B	605 A
883, 884, 885, 886, 876, 877	2	9.23 B	77.4 B	190 C	427 B
Control + 21 isolateds	1	7.83 C	52.3 C	100 C	319 C
Test 2*					
		Growth (cm)		Dry Biomass (mg)	
Isolated		h	CR	Root	Total
CCMD862		11.82A	82.42A	553A	712A
CCMD878		10.09 B	53.46 C	460B	359B
CCMD867		9.87 B	61.97 B	351C	491 B
Control		7.63 C	48.65C	313C	424B

* Test 1: the isolates were grouped according to the Euclidean similarity matrix and the means compared by the Scott-Knott test (P<0.05). ** Test 2: means compared by the Tukey test (P<0.05). CCMD: acronym for the Maria Duarte Culture Collection, followed by the isolate number. Means with different letters in the column differ statistically according to the tests used.

The morphological characterization revealed that the isolate CCMD862 is a Gram positive rod, central or subterminal endospore-forming. The colonies cultivated in NA at 28ºC for 48h have circular shape, with smooth edge, convex, bright appearance and coloration ranging from orange to pink, with an average diameter of 1.2 mm (Figure 1).

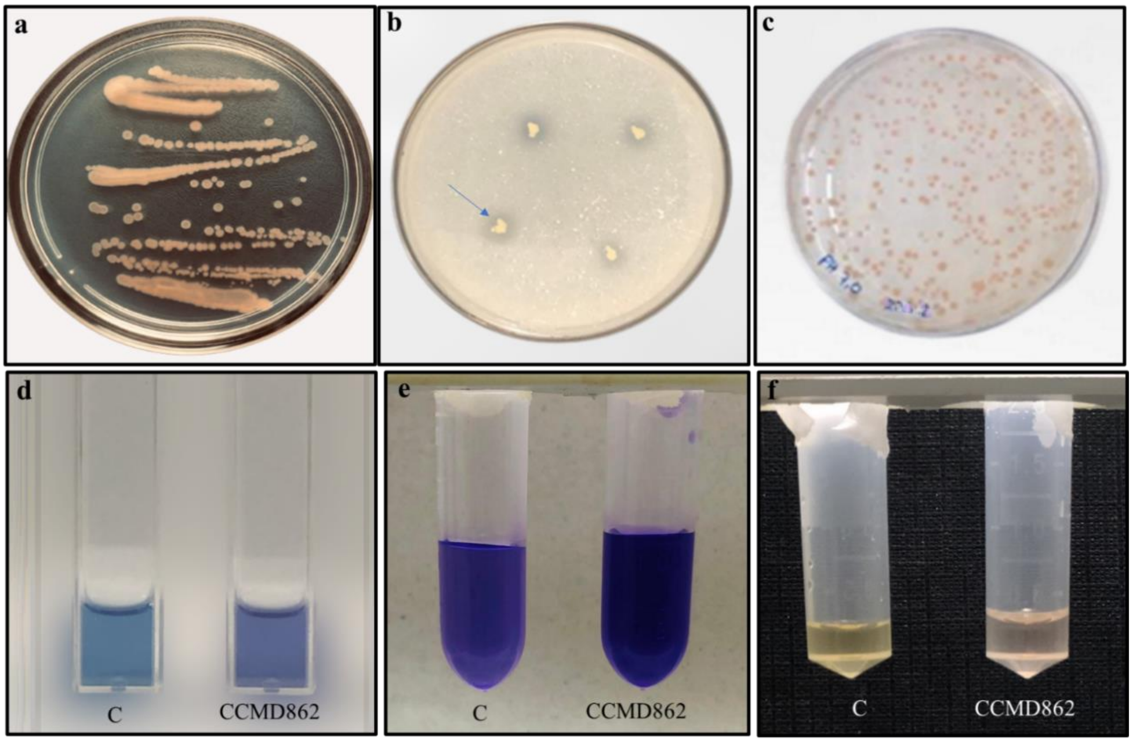


Figure 1. In vitro biochemical tests of the isolate CCMD862. (a) Pure cultivation of the isolate CCMD862 in Petri dish, (b) detection of phosphate solubilization, (c) nitrogen fixation, (d) siderophore production, (e) biofilm production and (f) production of indole acetic acid (AIA).

The rhizobacteria CCMD862 produces AIA, fixes nitrogen, solubilizes phosphate, produces biofilm and synthesizes siderophores (Figure 1). The production of AIA was confirmed by the change in the medium color after addition of the Salkowski reagent. BNF was confirmed by the growth of colonies in Burk medium, free of N. Phosphate solubilization was confirmed by the formation of halo (light zone) around the colonies in NBRIP medium. No solubilization halos were observed around the colonies in Aleksandrov medium using feldspar as mineral K, which indicates that the rhizobacteria have no potential to solubilize potassium in the experimental conditions tested. The production of siderophores was confirmed by the change of the shade of blue after the reaction of the bacterial culture with the CAS solution. Biofilm production was confirmed by the proportional relationship between the bacterial film formed under inert surface with the crystal violet dye.

The CCMD862 isolate sequence was compared in GenBank using the BLASTn tool. The isolate showed 100% identity with the genus *Bacillus* (ATCC14579T). Based on the construction of the phylogenetic tree of 29 accessions, it was possible to identify the isolate as *B. haikouensis* (Figure 2). The sequence was deposited in GenBank as *Bacillus* sp. CCMD862, with access code OQ506330.1 (https://www.ncbi.nlm.nih.gov/nuccore/OQ506330.1).

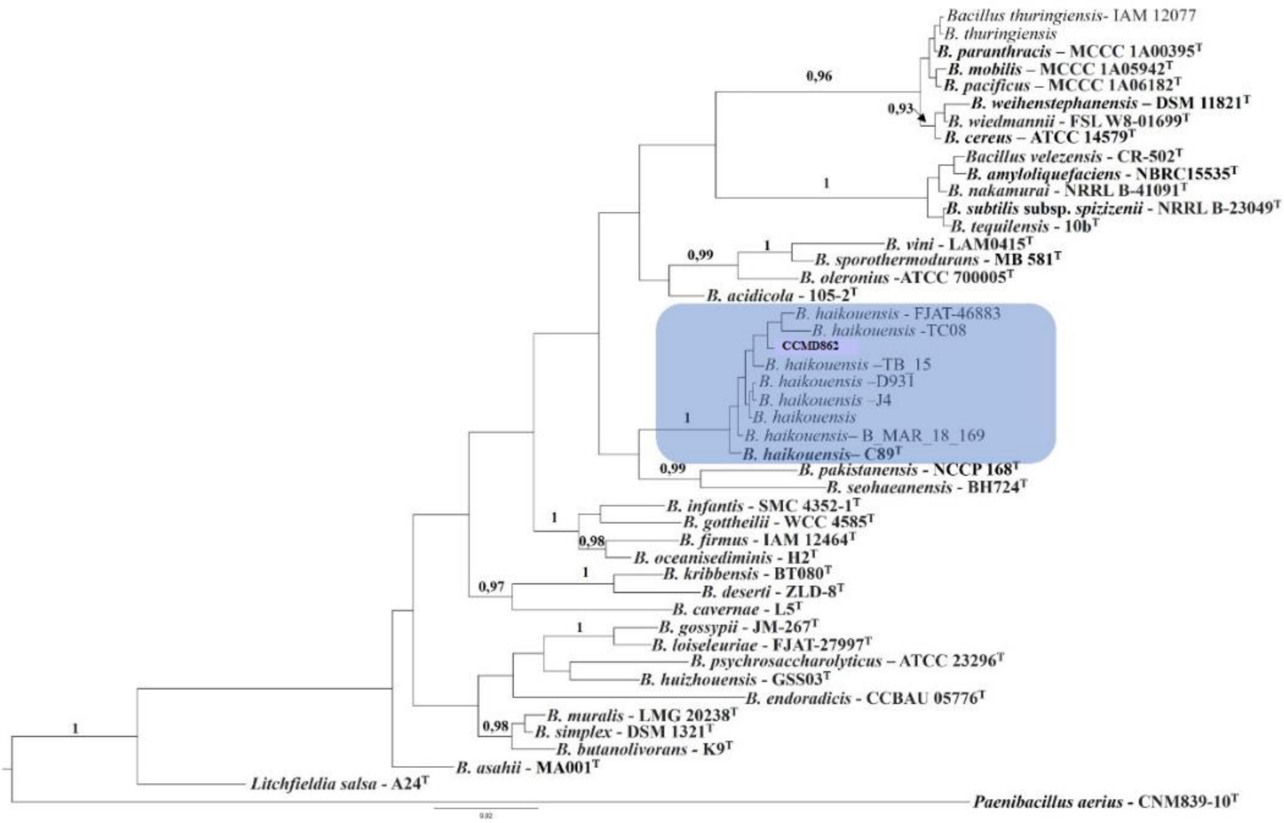


Figure 2. Phylogenetic analysis of the rhizobacteria Bacillus sp. CCMD862. The clade and groups of Bacillus haikouensis are indicated by comparing CCMD862 with the reference strains.

2.3.2 Gas exchange

Inoculation induced increases in gas exchange variables compared to control. The increases were 17% for *A*, 18% for *Gs*, 20% for *E*, 22% for *A/Ci* and 28% for *WUE*. The rhizobacteria did not change *Ci* (Figure 3).

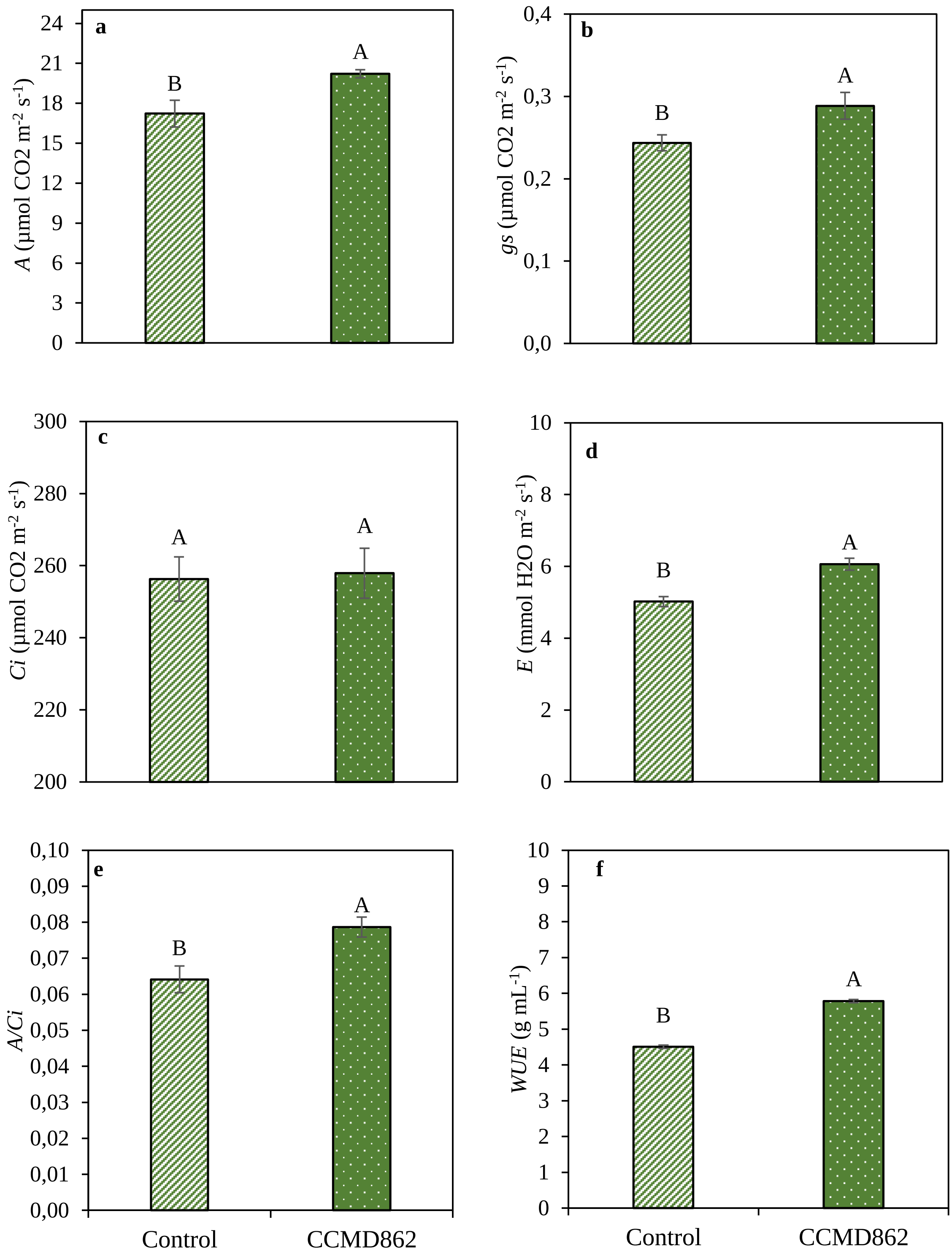


Figure 3. a: Net assimilation of CO_2 (*A*), b: Stomatal conductance to steam d`water (*Gs*), c: Intercellular concentration of CO_2 (*Ci*), d: Transpiration (*E*), Carboxylation efficiency (*A/Ci*) and f: Water use efficiency (*WUE*) in *Corymbia* seedlings inoculated with CCMD862. Means with equal letters do not differ from each other according to the unpaired Student's t-test ($P < 0.05$). Control: treatment without inoculation; CCMD862: treatment inoculated with *Bacillus* sp. CCMD862.

2.3.3 Fluorescence of chlorophyll a

The rhizobacteria caused mean reductions of 34% for *Fo* and 6% for *Qn* in relation to control. For *Fm* there was no significant effect of inoculation. On the other hand, all other variables increased. The increase was 85% for *Fv/Fo*, 20% for *ETR* and 12% for *Qp* (Figure 4).

2.3.4 Chlorophylls contents

The rhizobacteria promoted an increase in chlorophyll content compared to the control. For *Chl a* the increase was 38%, for *Chla+b* in 36% and for *Chla/Chlb* in 34%. The trend of increase observed for *Chl b* did not differ from the control (Figure 5).

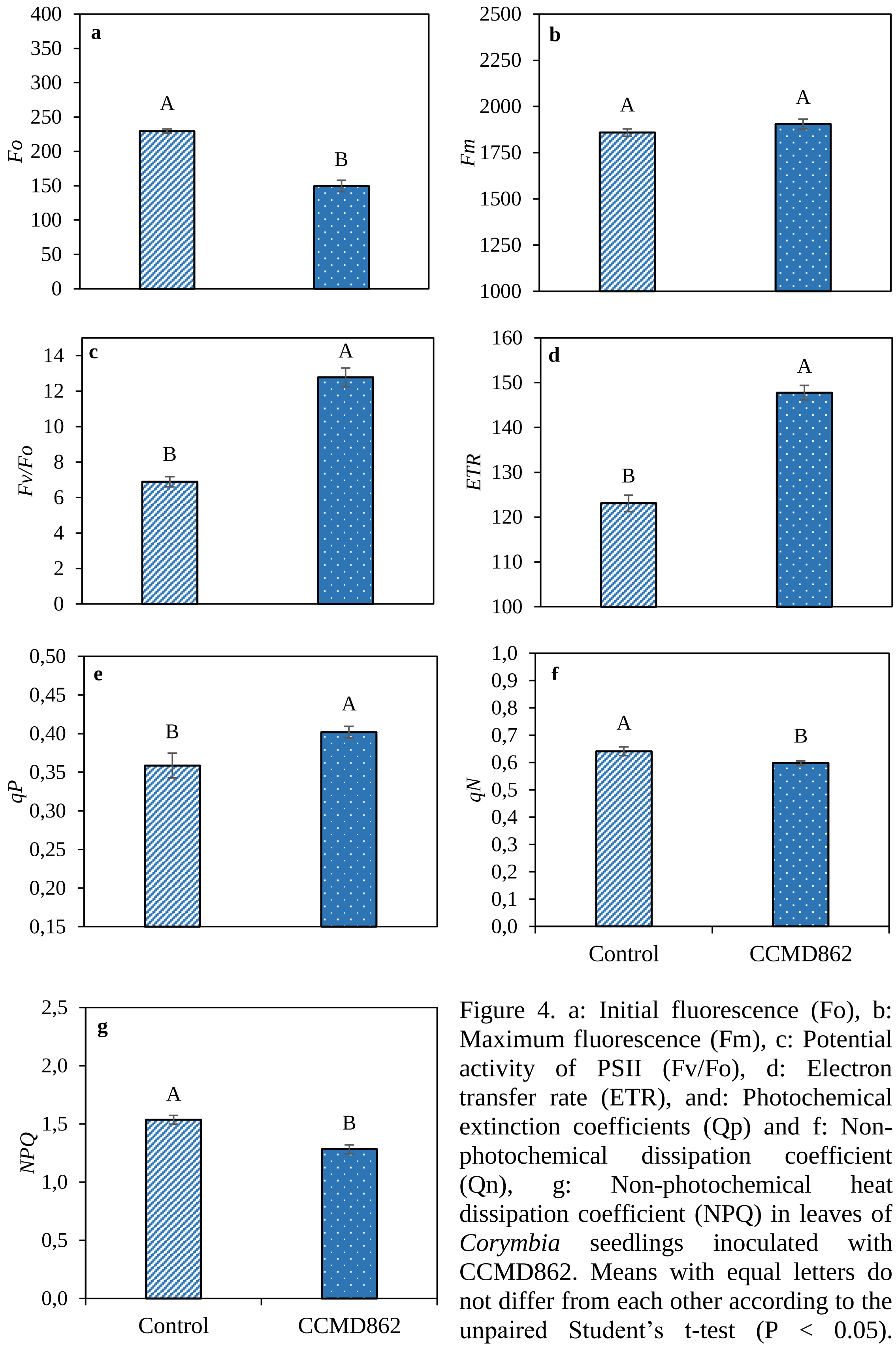


Figure 4. a: Initial fluorescence (Fo), b: Maximum fluorescence (Fm), c: Potential activity of PSII (Fv/Fo), d: Electron transfer rate (ETR), and: Photochemical extinction coefficients (Qp) and f: Non-photochemical dissipation coefficient (Qn), g: Non-photochemical heat dissipation coefficient (NPQ) in leaves of *Corymbia* seedlings inoculated with CCMD862. Means with equal letters do not differ from each other according to the unpaired Student's t-test ($P < 0.05$). Control: treatment without inoculation; CCMD862: treatment inoculated with *Bacillus* sp. CCMD862.

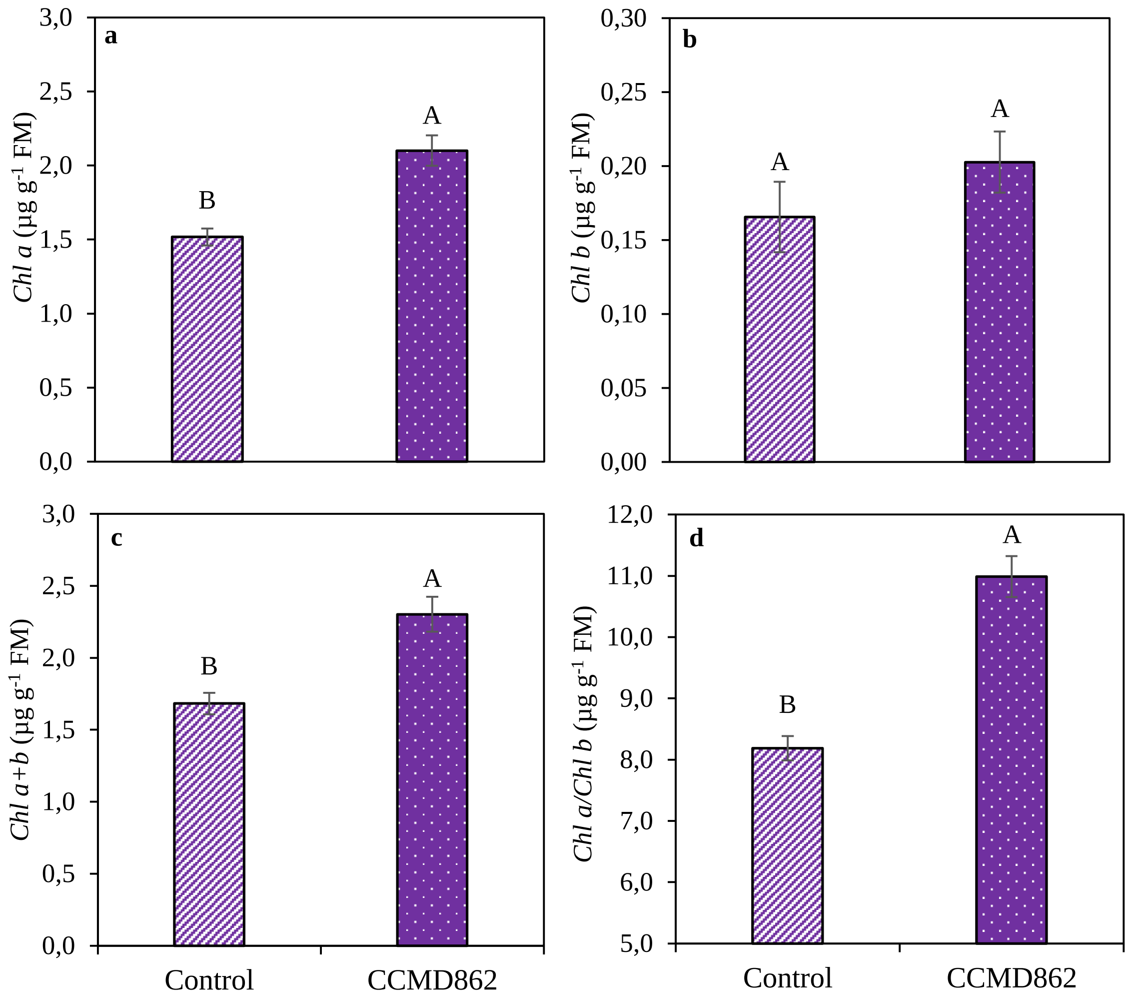


Figure 5. a: Chlorophyll-a (*Chla*), b: Chlorophyll-b (*Chlb*), c: total Chlorophyll (*Chla + b*) and d: Chla and Chlb ratio (*Chla/Chlb*) in *Corymbia* leaves inoculated with CCMD862. Means with equal letters do not differ from each other according to the unpaired Student's t test (P < 0.05). Control: treatment without inoculation; CCMD862: treatment inoculated with *Bacillus* sp. CCMD862. FM: fresh mass.

2.3.5 Biometrics and biomass

Inoculation promoted the growth of seedlings of *Corymbia*. Only the variables number of leaves, dry stem biomass and root/shoot ratio were not affected by the rhizobacteria *Bacillus* sp. CCMD862 (Figure 6).

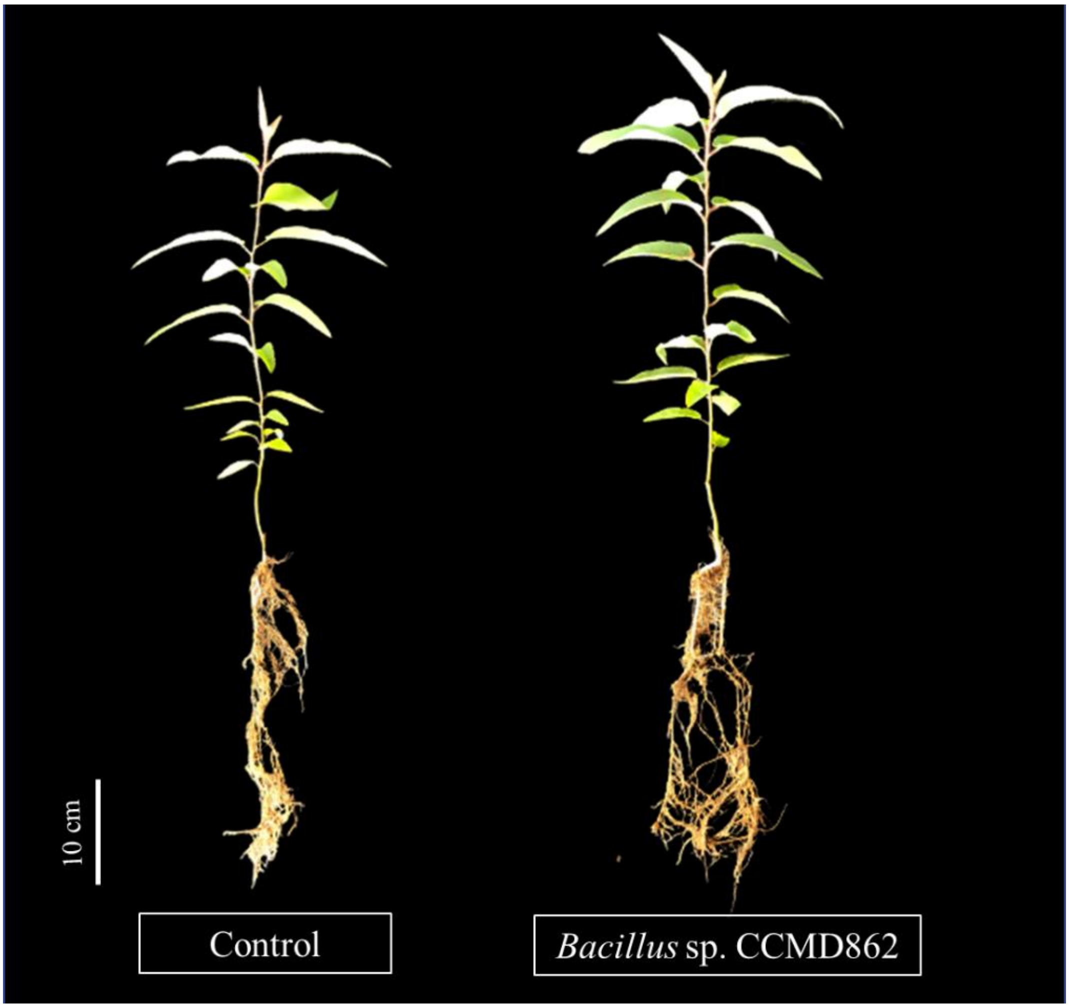


Figure 6. Growth promotion of seedlings of *C. torelliana x C. citriodora* 21 days after inoculation with the rhizobacteria *Bacillus* sp. CCMD862.

For growth variables inoculation resulted in an increase of 10% for plant height, 10% for stem diameter, 20% for leaf area. For the variables of dry biomass the increases were 56% in root, 11% in leaves and 29% total. The DQI increased by 23% and the growth promotion rate was 29% higher than the control plants (Table 2).

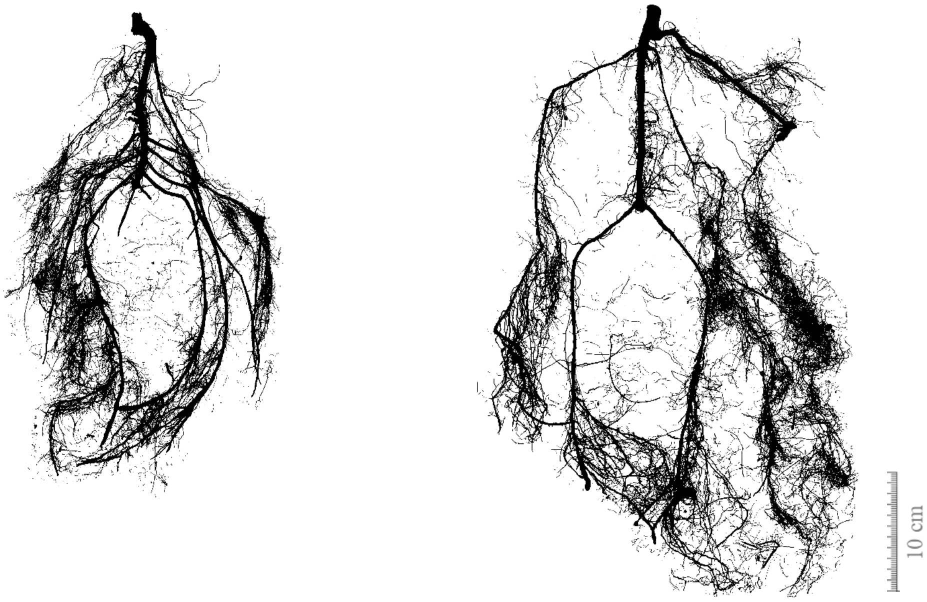
Table 2. Growth variables (biometry and biomass) of *Corymbia* seedlings inoculated with the rhizobacteria *Bacillus* sp. CCMD862.

	Control	*Bacillus* sp. CCMD862	c.v. (%)
Biometry			
Height (cm)	31.7 B	34.9 A	2
Collar diameter (mm)	3.42 B	3.74 A	5
Leaves	20 A	20 A	4
Leaf area (cm^2)	239 B	288 A	5
Dry biomass			
Root (g)	0.877 B	1.369 A	11
Stem (g)	0.543 A	0.658 A	9
Leaves (g)	1.471 B	1.636 A	6
Total (g)	2.840 B	3.670 A	4
Ratio root/part aerial	0.485 A	0.590 A	9
DIQ	0,29 B	0,35 A	4
Rate of promotion of growth (GP)		29%	

Note: Averages with equal letters in the line do not differ significantly according to the unpaired Student's t-test ($P < 0.05$). c.v.: coefficient of variation.

2.3.6 Root architecture

The rhizobacteria positively altered the root architecture of the plants (Figure 7) with an increase of 117% in total length, 43% in total surface area, 74% in root volume, 42% in density and 19% in number of tips, not being significant the increase of 13% in the average diameter of the roots (Table 3).



Control *Bacillus* sp. CCMD862

Figure 7. Aspects of the root architecture of *Corymbia* seedlings not inoculated (Control) and inoculated with the rhizobacteria *Bacillus* sp. CCMD862.

Table 3. Architecture of the root system of seedlings of *Corymbia* inoculated with the rhizobacteria *Bacillus* sp. CCMD862. Values are presented as mean ± standard error.

Variables	Control	*Bacillus* sp. CCMD862
Total length of roots (m)	24.87 ± 4.06 b	54.02 ± 2.36 a
Total surface area (cm^2)	208.7 ± 37.5 b	496.8 ± 44.0 a
Total volume (cm^3)	1.39 ± 0.27 b	4.07 ± 0.24 a
Density (cm m^{-3})	2487 ± 406 b	5402 ± 236 a
Average diameter (mm)	0.26 ± 0.004 a	0.34 ± 0.015 a
Number of tips	16.754 ± 1.088 b	25.353 ± 1.940 a

Note: Averages followed by the same letter do not differ by the unpaired Student's t-test ($P < 0.05$). Control: plants not inoculated. CCMD862: inoculated plants with rhizobacteria *Bacillus* sp. CCMD862.

In the variables of total length, total surface area and total volume, the inoculated seedlings presented more roots with diameter > 3.0 mm, compared to the non-inoculated seedlings (Figure 8).

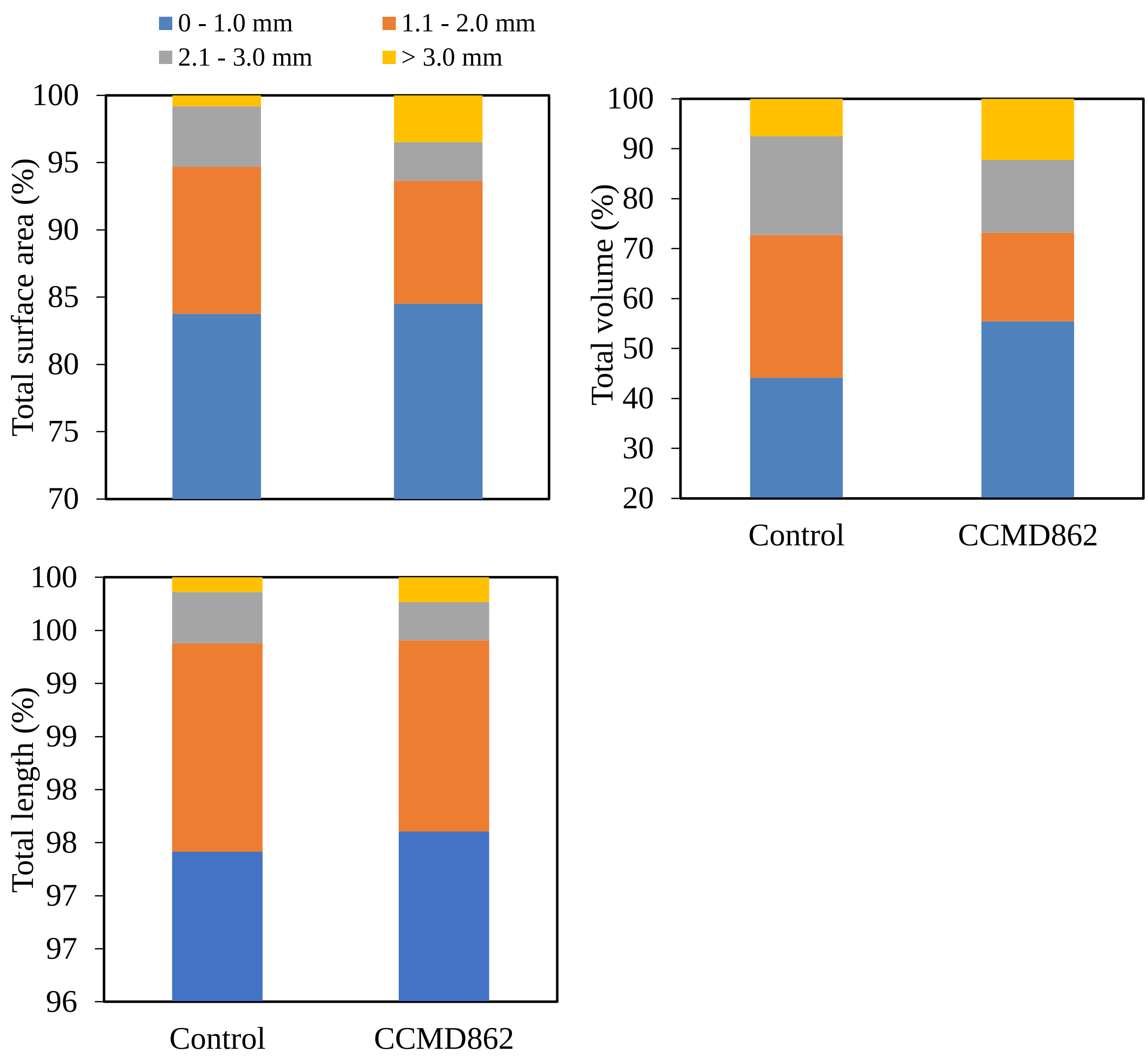


Figure 8. Proportion (%) of root morphology parameters in all diameter classes evaluated in seedlings of *Corymbia* not inoculated (control) and inoculated with rhizobacteria *Bacillus* sp. (CCMD862).

In total length the rhizobacteria increased by 244% the amount of roots with diameters > 3.0mm. There were 47% increases in class 2.1-3.0 mm and 50% in classes 0-1.0 mm and 1.1-2.0 mm, compared to control. In the total surface area, the increment in roots with 0-1.0 mm and 1.1-2.0 mm was 91% and 62% in inoculated plants. These plants also presented about 8x more contact area with roots of diameter > 3.0mm. Root volume was 190% higher in class > 3.0mm in seedlings inoculated with *Bacillus* sp. CCMD862. Similar increment was observed in class 0-1.0mm with an increase of 133%. In the intermediate classes (1.1-2.0 and 2.1-3.0mm) the increases were 15% compared to the non-inoculated seedlings.

2.3.7 Foliar nutrient content and nutrient use efficiency

Inoculation with *Bacillus* sp. CCMD862 induced increases in nutrient content in the leaves of *Corymbia* seedlings, except for N (nitrogen), P (phosphorus), Mg (magnesium) and S (sulfur). Compared to the control, seedlings inoculated with rhizobacteria increased potassium content (K) by 26% in 30% of calcium (Ca). For micronutrients, increases were 12% in boron (B), 24% in copper (Cu), 21% in iron (Fe), 97% in manganese (Mn) and 38% in iron (Fe) (Table 4). The inoculation of the rhizobacteria *Bacillus* sp. CCMD862 promoted improvements in the efficiency of nutrient use compared to non-inoculated plants. The use efficiency was higher in 25% for N, 40% for P, 31% for Ca, 26% for Mg and 40% for S, but there was no difference for the use efficiency of K. For micronutrients, inoculation promoted improvements in the use of B in 40%, 30% for Cu, 37% for Zn. The *NUE* did not differ for Fe and was lower for Mn in 16% compared to the control treatment.

Table 4. Leaf content and use efficiency of nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sulfur (S), boron (B), copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn) in seedling leaves of *Corymbia* without (control) and with inoculation (*Bacillus* sp. CCMD862)

Treatment	N	P	K	Ca	Mg	S	B	Cu	Fe	Mn	Zn
	------------------------------ $g\ kg^{-1}$ ------------------------------						-----------------------$mg\ kg^{-1}$ -------------------------				
Control	0.026 a	0.004 a	0.026 b	0.017 b	0.006 a	0.003 a	0.110 b	0.010 b	0.156 b	0.588 b	0.081 b
Bacillus sp. CCDM862	0.028 a	0.004 a	0.033 a	0.022 a	0.006 a	0.003 a	0.123 a	0.013 a	0.189 a	1.157 a	0.112 a
c.v. (%)	9	10	3	4	7	10	4	3	7	2	6
	NUE ($g^2\ mg^{-1}$)										
Control	0.28 b	1.77 b	0.28 a	0.34 b	1.07 b	1.93b	48.99 b	399 b	4.16 a	13.10 a	61.32 b
Bacillus sp. CCDM862	0.35 a	2.48 a	0.34 a	0.45 a	1.35 a	2.71 a	68.59 a	519 a	3.75 a	10.97 b	84.59 a
c.v. (%)	7	14	8	5	5	5	7	11	14	2	12

Note: Values are averages of 3 repetitions. Equal letters in the column indicate no significant differences unpaired Student's t-test ($P < 0.05$). c.v.: coefficient of variation. NUE: nutrient use efficiency

2.4. Discussion

The inoculation of the hybrid clones of *C. torelliana x C. citriodora* with the rhizobacteria *Bacillus* sp. CCMD862 provided growth improvements, with greater accumulation of root and shoot biomass, with increases in nutritional efficiency and photosynthetic performance.

Bacillus is a genus of gram-positive rods, capable of forming endospores, which tolerate adverse conditions for a long period of time (Vejan et al., 2016), tolerating a wide range of temperatures and pH, as well as efficiently resisting pesticides, fertilizers and heavy metals (Li et al., 2022). The sporulation capacity and ease of cultivation are favorable aspects for its application as an inoculant (Toyota, 2015). This group is recognized for having several characteristics that promote plant growth (Nephali et al., 2022; Melo, 2015).

Whereas *Bacillus* sp. CCMD862 was isolated from the rhizosphere of *R. communis* in a deposit of steel waste rich in heavy metals, it is possible that the rhizobacteria helped the plant to absorb nutrients from the soil while protecting from environmental stress caused by contaminants, expanding the possibilities of use of this rhizobacteria (Sandilya et al., 2022), such as in programs of recovery of degraded areas and the expansion of plantations to areas with the presence of pollutants.

The involvement in the physiological processes of the plant, such as leaf transpiration and photosynthesis, was recently considered as one of the main ways of improving plant growth under the supply of PGPR (Wang et al., 2022). In this research, the analysis of gas exchange showed that the rhizobacteria *Bacillus* sp. CCMD862 increased stomatal conductance, accompanied by higher carboxylation efficiency of the Rubisco enzyme and higher net photosynthesis rate. The larger stomatal opening allows greater CO_2 uptake although this results, almost always, in a higher rate of leaf transpiration. These results suggest a physiological adjustment by the plant to compensate for the higher energy demand in response to biostimulation.

The higher photosynthetic performance observed in inoculated plants was associated with improved energy uptake and transfer in the PSII. This result is well related to the absolute content of chlorophyll, especially chlorophyll *a* the main responsible for the capture, absorption and transfer of light energy (Win et al., 2018). The photochemical and non-photochemical extinction coefficients obtained with the inoculation of *Bacillus* sp. CCMD862 indicate the optimization of the operation of the photosynthetic apparatus, with the protection of the components of the photosystem and less energy loss during the photochemical stage (Campostrini, 2001; Liu et al., 2019). The effects of increases in the rate of net photosynthesis promoted by PGPR can also be observed in the greater contribution of carbon to plant metabolism subsidizing increase in glucose and sucrose production (Gupta et al., 2015), stimulating the synthesis of auxins (González-Hernández et al., 2020), which will sustain the highest root meristematic activity by stimulating growth and greater formation of lateral roots, an important result when considering that the vegetative propagation of Corymbia species is difficult to perform, with low rooting levels (Reis et al, 2014).

Another aspect that influences the success of propagation by cutting refers to the nutritional status of the propagules that will result in seedlings. Nutrition interferes with rhizogenesis, since it regulates the amount of carbohydrates, auxins and other essential compounds for this process (Pereira & Bandeira Peres, 2016). *Bacillus* sp. CCMD862 promoted improvement in the efficiency of use of macro and micronutrients, on average, 2 times higher than in non-inoculated plants.

The multicompetent isolate *Bacillus* sp. CCMD862 was efficient in promoting plant growth through nutritional facilitation resulting from the ability to fix N, phosphate solubilization, production of siderophores and biofilm presented by this strain. *Bacillus* species can contribute 30% to 50% of fixed N in some agricultural crops (Rosenblueth et al., 2018) and minimize N losses by optimizing chemical fertilization (Lima et al., 2021). As for phosphorus, *Bacillus* can increase the solubility of the inorganic P fraction linked to Fe, Al and/or Ca (Fageria & Stone, 2006), through the release of organic acids that can desorb phosphate through the exchange of ligands and thus release Pi for use by the plant (Yu et al., 2014). The greater amount of soluble macro- and micronutrients near the soil-root interface actually has a positive effect on plant nutrition with improvements in growth (Meena et al., 2017).

The nutritional gains presented in this study can also be explained by the changes observed on the root system. The architecture of the root system describes the shape and spatial configuration of the roots in the soil and is the result of the combination of the production of new apical meristem and the initiation and extension of lateral roots (Gregory & Kirkegaard, 2017). Improvements in the structure and dimension of roots promoted by rhizobacteria are recognized, demonstrating the potential that they have to modify the root system after inoculation (Lima et al., 2021).

The synthesis of auxins by rhizobacteria associated with plants is probably one of the main causes of increased plant growth (Liu et al., 2014). Several cellular processes are regulated by AIA, involving changes in the pattern of protein synthesis, cell division, differentiation and elongation (Batista et al., 2021), and depending on the concentrations of exogenous AIA there may be distinct where lower concentrations promote the elongation of the primary root while high levels of IAA decrease its length, but increase the formation of root hairs and lateral roots, as well as the formation of adventitious roots in the stem (Taiz; Zeiger, 2017).

The more intense branching of the roots favors better rooting after planting, as a result the plants are able to explore greater soil volume, accessing more nutrients and water (Vacheron et al., 2013; Verma et al., 2021; Cardoso et al., 2021) and is considered a potential measure to improve plant development (Yu et al., 2014), an effect proven by biostimulation in this study.

Auxin production generally affects cell division, extension and differentiation, increases xylem and root development rate, and controls vegetative growth processes (Glick, 2012). The significant increase in surface area and volume of roots with diameters above 3 mm in inoculated plants may be related to endogenous and exogenous concentrations of IAA.

Although the height and diameter of the stem are the characteristics commonly evaluated to infer about the quality of the seedlings, the morphology of the root system can provide a more accurate indication of the seedling potential. Large root volume, high root fibrosity and an increased number of first-order lateral roots (larger than 1mm in diameter) are related to higher seedling quality and better field performance (Davis & Jacobs, 2005).

The increments in biometric and biomass parameters in plants inoculated with *Bacillus* sp. CCMD862 resulted in higher DQI, with gains in robustness and improvements in biomass partition between shoot and root. In addition to this index, the increase in volume of roots with larger diameters added to the higher rate of estimated overall growth promotion, allows to conclude that there was acceleration of plant growth and elevation of the final quality standard of the same.

Obtaining multifunctional bacterial isolates such as *Bacillus* sp. CCMD862 for seedling production of *Corymbia* is a viable ecological strategy to minimize the effects of recalcitrance to rooting, leading to greater efficiency in the conversion of nutrients into biomass and photosynthetic performance. Such improvements will contribute to the achievement of reducing the age of seedling dispatch and greater survival in the field and yield of planted forests.

2.5 Conclusion

Bacillus sp. CCMD862 is able to biostimulate seedlings of *C. torelliana* x *C. citriodora* favoring the development of the root system, gains in the photosynthetic apparatus, increases in absorption and efficient use of nutrients that resulted in robust seedlings.

The development of an inoculant in the future may reduce the limitations of rooting in *Corymbia*, contributing to the reduction of field mortality and obtaining more homogeneous commercial plantations.

REFERENCES

ABIRI, R. *et al.* The Prospect of Physiological Events Associated with the Micropropagation of Eucalyptus sp. **Forests**, *[S. l.]*, v. 11, n. 11, p. 1211, 2020. Disponível em: https://doi.org/10.3390/f11111211

AFZAL, M. J. *et al.* Combined application of Bacillus sp. MN-54 and phosphorus improved growth and reduced lead uptake by maize in the lead-contaminated soil. **Environmental Science and Pollution Research**, *[S. l.]*, v. 27, n. 35, p. 44528–44539, 2020. Disponível em: https://doi.org/10.1007/S11356-020-10372-4/FIGURES/7. Acesso em: 10 dez. 2022.

AGARWAL, P. *et al.* PGPR-induced OsASR6 improves plant growth and yield by altering root auxin sensitivity and the xylem structure in transgenic Arabidopsis thaliana. **Journal of Plant Physiology**, *[S. l.]*, v. 240, p. 153010, 2019. Disponível em: https://doi.org/10.1016/j.jplph.2019.153010

AHEMAD, M.; KIBRET, M. Mechanisms and applications of plant growth promoting rhizobacteria: Current perspective. **Journal of King Saud University - Science**, *[S. l.]*, v. 26, n. 1, p. 1–20, 2014. Disponível em: https://doi.org/10.1016/j.jksus.2013.05.001

AHKAMI, A. H. *et al.* **Rhizosphere engineering: Enhancing sustainable plant ecosystem**

productivity. *[S. l.]*: Elsevier B.V., 2017. Disponível em: https://doi.org/10.1016/j.rhisph.2017.04.012

ALEKSANDROV, VG; BLAGODYR, RN; ILEV, I. Liberation of phosphoric acid from apatite by silicate bacteria. **Mikrobiol Z**, *[S. l.]*, 1967.

ALI, M. A.; OH, J. H.; KIM, P. J. Evaluation of silicate iron slag amendment on reducing methane emission from flood water rice farming. **Agriculture, Ecosystems & Environment**, *[S. l.]*, v. 128, n. 1–2, p. 21–26, 2008. Disponível em: https://doi.org/10.1016/j.agee.2008.04.014

ALMEIDA, R. A. M. de *et al.* Slag Foaming Fundamentals - A Critical Assessment. **Materials Research**, *[S. l.]*, v. 20, n. 2, p. 474–480, 2017. Disponível em: https://doi.org/10.1590/1980-5373-mr-2016-0059

ANGULO, V. C. *et al.* Caracterización de rizobacterias promotoras de crecimiento en plántulas de Eucalyptus nitens. **Revista Argentina de Microbiología**, *[S. l.]*, v. 46, n. 4, p. 338–347, 2014 a. Disponível em: https://doi.org/10.1016/S0325-7541(14)70093-8

ANGULO, V. C. *et al.* Caracterización de rizobacterias promotoras de crecimiento en plántulas de Eucalyptus nitens. **Revista Argentina de Microbiología**, *[S. l.]*, v. 46, n. 4, p. 338–347, 2014 b. Disponível em: https://doi.org/10.1016/S0325-7541(14)70093-8

ASSIS, T. F. de. Melhoramento genético de Eucalyptus: desafios e perspectivas. **Anais do SIMPÓSIO BRASILEIRO DE SILVICULTURA**, *[S. l.]*, v. 1, n. 1, p. 127–148, 2014.

ASSIS, T. F. de; ABAD, J. I. M.; AGUIAR, A. M. Melhoramento Genético do Eucalipto. *In*: MAURO VALDIR SCHUMACHER E MÁRCIO VIERA (ORG.) (org.). **Silvicultura do Eucalipto no Brasil**. 1. ed. Santa Maria: Editora UFSM, 2015. p. 225–247. *E-book*.

AUSTRALASIAN (IRON AND STEEL) SLAG ASSOCIATION INC. **Membership Annual Survey Results, 2019 Slag Sales and Production Survey, Australia.** *[S. l.: s. n.]* Disponível em: https://www.asa-inc.org.au/files/download/d787f72a84b5512

AVELAR, M. L. M. *et al.* In vitro ESTABLISHMENT OF Eucalyptus AND Corymbia SPECIES FROM EPICORMIC SHOOTS. **Revista Árvore**, *[S. l.]*, v. 44, 2020. Disponível em: https://doi.org/10.1590/1806-908820200000027

BELLINI, C.; PACURAR, D. I.; PERRONE, I. Adventitious Roots and Lateral Roots: Similarities and Differences. **Annual Review of Plant Biology**, *[S. l.]*, v. 65, n. 1, p. 639–666, 2014. Disponível em: https://doi.org/10.1146/annurev-arplant-050213-035645

BERBEL, A. *et al.* Changes in root morphological traits in soybean co-inoculated with Bradyrhizobium spp. and Azospirillum brasilense or treated with A. brasilense exudates. **Biology and Fertility of Soils**, *[S. l.]*, v. 56, p. 537–549, 2020. Disponível em: https://doi.org/10.1007/s00374-020-01453-0. Acesso em: 19 fev. 2023.

BRANCA, T. A. *et al.* Reuse and Recycling of By-Products in the Steel Sector: Recent Achievements Paving the Way to Circular Economy and Industrial Symbiosis in Europe. **Metals**, *[S. l.]*, v. 10, n. 3, p. 345, 2020 a. Disponível em: https://doi.org/10.3390/met10030345

BRANCA, T. A. *et al.* Reuse and Recycling of By-Products in the Steel Sector: Recent Achievements Paving the Way to Circular Economy and Industrial Symbiosis in Europe. **Metals**, *[S. l.]*, v. 10, n. 3, p. 345, 2020 b. Disponível em: https://doi.org/10.3390/met10030345

BRUNO MARCOS NUNES COSMO *et al.* Influência dos metais na fotossíntese. **Revista Agronomia Brasileira**, *[S. l.]*, v. 7, 2023. Disponível em: https://doi.org/10.29372/rab202347

CAI, S. *et al.* Fertilizer Efficiency and Risk Assessment of the Utilization of AOD Slag as a Mineral Fertilizer for Alfalfa (Medicago sativa L.) and Perennial Ryegrass (Lolium perenne L.) Planting. **Sustainability 2022, Vol. 14, Page 1575**, *[S. l.]*, v. 14, n. 3, p. 1575, 2022 a. Disponível em: https://doi.org/10.3390/SU14031575. Acesso em: 14 nov. 2022.

CAI, S. *et al.* Biochemical Analysis and Toxicity Assessment of Utilization of Argon Oxygen Decarbonization Slag as a Mineral Fertilizer for Tall Fescue (Festuca arundinacea Schreb) Planting. **Sustainability**, *[S. l.]*, v. 14, n. 15, p. 9286, 2022 b. Disponível em: https://doi.org/10.3390/su14159286

CALDWELL, M. M.; RICHARDS, J. H. Hydraulic lift: water efflux from upper roots improves effectiveness of water uptake by deep roots. **Oecologia**, *[S. l.]*, n. 79, p. 1–5, 1989.

CALVO, P. *et al.* Plant growth-promoting rhizobacteria induce changes in Arabidopsis thaliana gene expression of nitrate and ammonium uptake genes. **Journal of Plant Interactions**, *[S. l.]*, v. 14, n. 1, p. 224–231, 2019. Disponível em: https://doi.org/10.1080/17429145.2019.1602887

CALVO, P.; NELSON, L.; KLOEPPER, J. W. Agricultural uses of plant biostimulants. **Plant and Soil**, *[S. l.]*, n. 383, p. 3+41, 2014. Disponível em: https://doi.org/10.1007/s11104-014-2131-8

CARDOSO, P. H. S. *et al.* Fertilization management with sewage sludge sustains Eucalyptus productivity in Cerrado infertile soil. **Forest Ecology and Management**, *[S. l.]*, v. 519, p.

120338, 2022. Disponível em: https://doi.org/10.1016/j.foreco.2022.120338

CASTRO, G. S. A.; CRUSCIOL, C. A. C. Effects of superficial liming and silicate application on soil fertility and crop yield under rotation. **Geoderma**, *[S. l.]*, v. 195–196, p. 234–242, 2013. Disponível em: https://doi.org/10.1016/J.GEODERMA.2012.12.006

CHAND, S.; PAUL, B.; KUMAR, M. An Overview of Use of Linz-Donawitz (LD) Steel Slag in Agriculture. **Current World Environment**, *[S. l.]*, v. 10, n. 3, p. 975–984, 2015. Disponível em: https://doi.org/10.12944/CWE.10.3.29

CHAVES, M. M. How Plants Cope with Water Stress in the Field? Photosynthesis and Growth. **Annals of Botany**, *[S. l.]*, v. 89, n. 7, p. 907–916, 2002. Disponível em: https://doi.org/10.1093/aob/mcf105

CHEN, G. *et al.* Competitive mechanism and influencing factors for the simultaneous removal of Cr(III) and Zn(II) in acidic aqueous solutions using steel slag: Batch and column experiments. **Journal of Cleaner Production**, *[S. l.]*, v. 230, p. 69–79, 2019. Disponível em: https://doi.org/10.1016/j.jclepro.2019.04.402

CHIEN-YING YANG, INKA REIJONEN, HAI YU, RAJARATHNAM DHARMARAJAN, BALAJI SESHADRI, N. S. B. Back to Basic Slags as a Phosphorus Source and Liming Material. *In*: **Soil Amendments for Sustainability**. 1. ed. *[S. l.]*: CRC Press, 2018. p. 14. *E-book*.

CHOUDHARY, A. *et al.* Role of plant growth-promoting Rhizobacterium in adventitious root formation. *In*: **Environmental, Physiological and Chemical Controls of Adventitious Rooting in Cuttings**. *[S. l.]*: Elsevier, 2022. p. 159–181. *E-book*. Disponível em: https://doi.org/10.1016/B978-0-323-90636-4.00007-6

CORRÊA, J. C. *et al.* Disponibilidade de metais pesados em Latossolo com aplicação superficial de escória, lama cal, lodos de esgoto e calcário. **Pesquisa Agropecuaria Brasileira**, *[S. l.]*, v. 43, n. 3, p. 411–419, 2008. Disponível em: https://doi.org/10.1590/S0100-204X2008000300017. Acesso em: 28 abr. 2021.

CUNHA, T. Q. G. da *et al.* Eucalyptus expansion in Brazil: Energy yield in new forest frontiers. **Biomass and Bioenergy**, *[S. l.]*, v. 144, p. 105900, 2021. Disponível em: https://doi.org/10.1016/j.biombioe.2020.105900

DAS, S. *et al.* Taxonomic and functional responses of soil microbial communities to slag-based fertilizer amendment in rice cropping systems. **Environment International**, *[S. l.]*, v. 127, p.

531–539, 2019. Disponível em: https://doi.org/10.1016/J.ENVINT.2019.04.012

DAS, S. *et al.* Recycling of ferrous slag in agriculture: Potentials and challenges. **Critical Reviews in Environmental Science and Technology**, *[S. l.]*, p. 1–35, 2020. Disponível em: https://doi.org/10.1080/10643389.2020.1853458. Acesso em: 20 abr. 2021.

DE ARAUJO, M. J. *et al.* Initial productivity and genetic parameters of three corymbia species in brazil: Designing a breeding strategy. **Canadian Journal of Forest Research**, *[S. l.]*, v. 51, n. 1, p. 25–30, 2021. Disponível em: https://doi.org/10.1139/CJFR-2019-0438/ASSET/IMAGES/CJFR-2019-0438IEQ2.GIF. Acesso em: 25 abr. 2023.

DE CASTRO, G. L. S. *et al.* Açaí palm seedling growth promotion by rhizobacteria inoculation. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 51, n. 1, p. 205–216, 2020. Disponível em: https://doi.org/10.1007/s42770-019-00159-2

DEUS, A. C. F. *et al.* Effects of lime and steel slag application on soil fertility and soybean yield under a no till-system. **Soil and Tillage Research**, *[S. l.]*, v. 196, 2020. Disponível em: https://doi.org/10.1016/j.still.2019.104422

DHALI, S. *et al.* Synergistic effect of Bacillus and Rhizobium on cytological and photosynthetic performance of Macrotyloma uniflorum (Lam.) Verdc. Grown in Cr (VI) contaminated soil. **Plant Physiology and Biochemistry**, *[S. l.]*, v. 190, p. 62–69, 2022. Disponível em: https://doi.org/10.1016/J.PLAPHY.2022.08.027

DÍAZ-PILONETA, M. *et al.* Application of Steel Slag for Degraded Land Remediation. **Land**, *[S. l.]*, v. 11, n. 2, p. 224, 2022. Disponível em: https://doi.org/10.3390/LAND11020224. Acesso em: 15 fev. 2022.

DOTANIYA, M. L.; MEENA, V. D. Rhizosphere Effect on Nutrient Availability in Soil and Its Uptake by Plants: A Review. **Proc. Natl. Acad. Sci., India, Sect. B Biol. Sci.**, *[S. l.]*, n. 1, p. 1–15, 2015. Disponível em: https://doi.org/10.1007/s40011-013-0297-0

DUCA, D. *et al.* Indole-3-acetic acid in plant–microbe interactions. **Antonie van Leeuwenhoek**, *[S. l.]*, v. 106, n. 1, p. 85–125, 2014. Disponível em: https://doi.org/10.1007/s10482-013-0095-y

EGAMBERDIEVA, D. *et al.* Phytohormones and beneficial microbes: Essential components for plants to balance stress and fitness. **Frontiers in Microbiology**, *[S. l.]*, v. 8, n. OCT, p. 2104, 2017. Disponível em: https://doi.org/10.3389/FMICB.2017.02104/BIBTEX

EL-MEIHY, R. M. *et al.* Efficiency of heavy metals-tolerant plant growth promoting bacteria for alleviating heavy metals toxicity on sorghum. **Environmental and Experimental Botany**, *[S. l.]*, v. 162, 2019. Disponível em: https://doi.org/10.1016/j.envexpbot.2019.03.005

ELSAYED, A. *et al.* Synergistic effects of Azospirillum brasilense and Bacillus cereus on plant growth, biochemical attributes and molecular genetic regulation of steviol glycosides biosynthetic genes in Stevia rebaudiana. **Plant Physiology and Biochemistry**, *[S. l.]*, v. 189, p. 24–34, 2022. Disponível em: https://doi.org/10.1016/J.PLAPHY.2022.08.016

ENEBAK, S. A.; WEI, G.; KLOEPPER, J. W. Effects of Plant Growth-Promoting Rhizobacteria on Loblolly and Slash Pine Seedlings. **Forest Science**, *[S. l.]*, v. 44, n. 1, p. 139–144, 1998. Disponível em: https://doi.org/https://doi.org/10.1093/forestscience/44.1.139. Acesso em: 20 fev. 2023.

ETESAMI, H. Plant–microbe interactions in plants and stress tolerance. **Plant Life Under Changing Environment**, *[S. l.]*, p. 355–396, 2020. Disponível em: https://doi.org/10.1016/B978-0-12-818204-8.00018-7

EUROPEAN COMMISSION, D.-G. for R. and I.; KUHN, M., SPIEGEL, H., LOPEZ, A. F., REX, M., ERDMANN, R. **Sustainable agriculture using blast furnace and steel slags as liming agents**. Luxembourg: Publications Office, 2006.

FILIPPI, M. C. C. *et al.* Leaf blast (Magnaporthe oryzae) suppression and growth promotion by rhizobacteria on aerobic rice in Brazil. **Biological Control**, *[S. l.]*, v. 58, n. 2, p. 160–166, 2011. Disponível em: https://doi.org/10.1016/J.BIOCONTROL.2011.04.016

GE, Y. *et al.* Biofilm-overproducing Bacillus subtilis B12ΔYwcc decreases Cd uptake in Chinese cabbage through increasing Cd-immobilizing related gene abundance and root surface colonization. **Journal of Environmental Sciences**, *[S. l.]*, v. 120, p. 84–93, 2022. Disponível em: https://doi.org/10.1016/J.JES.2022.01.016

GONÇALVES MAFIA, R. *et al.* **Root colonization and interaction among growth … R. Árvore, Viçosa-MG, v.33, n.1**. *[S. l.: s. n.]*.

GONZÁLEZ, P. *et al.* Rhizobacteria strains as promoters of rooting in hybrids of Eucalyptus nitens × Eucalyptus globulus. **Chilean journal of agricultural research**, *[S. l.]*, v. 78, n. 1, p. 3–12, 2018. Disponível em: https://doi.org/10.4067/S0718-58392018000100003

GORDON, S. A.; WEBER, R. P. Colorimetric Estimation of indoleacetic Acid. **Plant Physiology**, *[S. l.]*, v. 26, n. 1, p. 192–195, 1951. Disponível em:

https://doi.org/10.1104/PP.26.1.192. Acesso em: 19 abr. 2022.

GOSWAMI, M.; DEKA, S. Isolation of a novel rhizobacteria having multiple plant growth promoting traits and antifungal activity against certain phytopathogens. **Microbiological Research**, *[S. l.]*, v. 240, p. 126516, 2020. Disponível em: https://doi.org/10.1016/j.micres.2020.126516

GROBELAK, A. *et al.* Bacterial ACC deaminase activity in promoting plant growth on areas contaminated with heavy metals. **Journal of Ecological Engineering**, *[S. l.]*, v. 19, n. 5, p. 150–157, 2018. Disponível em: https://doi.org/10.12911/22998993/89818

GUERRINI, I. A. *et al.* Composted sewage sludge and steel mill slag as potential amendments for urban soils involved in afforestation programs. **Urban Forestry & Urban Greening**, *[S. l.]*, v. 22, p. 93–104, 2017. Disponível em: https://doi.org/10.1016/J.UFUG.2017.01.015

GUO, J.; BAO, Y.; WANG, M. **Steel slag in China: Treatment, recycling, and management**. *[S. l.]*: Elsevier Ltd, 2018. Disponível em: https://doi.org/10.1016/j.wasman.2018.04.045

GWON, H. S. *et al.* Environmental risk assessment of steel-making slags and the potential use of LD slag in mitigating methane emissions and the grain arsenic level in rice (Oryza sativa L.). **Journal of Hazardous Materials**, *[S. l.]*, v. 353, 2018. Disponível em: https://doi.org/10.1016/j.jhazmat.2018.04.023

HALING, R. E. *et al.* Root hairs improve root penetration, root–soil contact, and phosphorus acquisition in soils of different strength. **Journal of Experimental Botany**, *[S. l.]*, v. 64, n. 12, p. 3711–3721, 2013. Disponível em: https://doi.org/10.1093/jxb/ert200

HAYNES, R. J.; ZHOU, Y. F. Effect of pH and added slag on the extractability of Si in two Si-deficient sugarcane soils. **Chemosphere**, *[S. l.]*, v. 193, p. 431–437, 2018. Disponível em: https://doi.org/10.1016/J.CHEMOSPHERE.2017.10.175

HE, H. *et al.* Effects of alkaline and bioorganic amendments on cadmium, lead, zinc, and nutrient accumulation in brown rice and grain yield in acidic paddy fields contaminated with a mixture of heavy metals. **Environmental Science and Pollution Research**, *[S. l.]*, v. 23, n. 23, p. 23551–23560, 2016. Disponível em: https://doi.org/10.1007/S11356-016-7593-1/TABLES/6. Acesso em: 1 dez. 2022.

HILTUNEN, R., AND HILTUNEN, A. "Environmental aspects of the utilization of steel industry slags". *In*: THE SOUTH AFRICAN INSTITUTE OF MINING AND METALLURGY (org.). **Proceedings of VII intenational conference on molten slags, fluxes and salts**. Cape Town, South Africa: *[s. n.]*, 2004. *E-book*.

HOSSEINI, S. *et al.* Environmental Technology Reviews Production and applications of electric-arc-furnace slag as solid waste in environmental technologies: a review. *[S. l.]*, 2016. Disponível em: https://doi.org/10.1080/21622515.2016.1147615

HU, Z. H. *et al.* Combined application of arbuscular mycorrhizal fungi and steel slag improves plant growth and reduces Cd, Pb accumulation in Zea mays. **International Journal of Phytoremediation**, *[S. l.]*, v. 21, n. 9, p. 857–865, 2019. Disponível em: https://doi.org/10.1080/15226514.2019.1577355

HUSSEINY, S. *et al.* Characterization of growth promoting bacterial endophytes isolated from Artemisia annua L. **South African Journal of Botany**, *[S. l.]*, v. 143, p. 238–247, 2021. Disponível em: https://doi.org/10.1016/J.SAJB.2021.07.042. Acesso em: 19 abr. 2022.

IBGE. **PEVS 2021: Produção da Extração Vegetal e da Silvicultura**. 36. ed. Rio de Janeiro: IBGE, 2022. *E-book*.

INDÚSTRIA BRASILEIRA DE ÁRVORES. **Relatório Anual 2021**. São Paulo: Fundação Getulio Vargas (FGV), 2022. Disponível em: https://www.iba.org/datafiles/publicacoes/relatorios/relatorio-anual-iba2022-compactado.pdf

INSTITUTO AÇO BRASIL. **Relatório de Sustentabilidade 2020**2021. p. 62. Disponível em: https://www.acobrasil.org.br/relatoriodesustentabilidade/

INSTITUTO AÇO BRASIL. **Estatísticas da siderurgia**. *[s. l.]*, 2023. Disponível em: https://acobrasil.org.br/site/.

ISLAM, F. *et al.* Influence of Pseudomonas aeruginosa as PGPR on oxidative stress tolerance in wheat under Zn stress. **Ecotoxicology and Environmental Safety**, *[S. l.]*, v. 104, p. 285–293, 2014. Disponível em: https://doi.org/10.1016/j.ecoenv.2014.03.008

ISLAM, Z. *et al.* Effect of Steel Slag on Soil Fertility and Plant Growth. **Journal of Agricultural Chemistry and Environment**, *[S. l.]*, 2022. Disponível em: https://doi.org/10.4236/jacen.2022.113014. Acesso em: 9 out. 2022.

ITO, K. Steelmaking Slag for Fertilizer Usage. **NIPPON STEEL & SUMITOMO METAL TECHNICAL REPORT**, *[S. l.]*, n. 109, p. 130–136, 2015. Disponível em: https://doi.org/UDC 669 . 184 . 28 : 631 . 82 /. 86

JI, S. H.; GURURANI, M. A.; CHUN, S. C. Isolation and characterization of plant growth promoting endophytic diazotrophic bacteria from Korean rice cultivars. **Microbiological Research**, *[S. l.]*, v. 169, n. 1, p. 83–98, 2014. Disponível em: https://doi.org/10.1016/J.MICRES.2013.06.003

JING, Y.; HE, Z.; YANG, X. Role of soil rhizobacteria in phytoremediation of heavy metal contaminated soils. **Journal of Zhejiang University SCIENCE B**, *[S. l.]*, v. 8, n. 3, p. 192–207, 2007. Disponível em: https://doi.org/10.1631/jzus.2007.B0192

JOÃO LUCAS BOLDT, EDUARDO LUCAS TERRA PEIXOTO, GUSTAVO FERREIRA DE OLIVEIRA, A. H. de M. INOCULATION OF TREE SPECIES WITH MYCORRHIZAL FUNGI FERTILIZED WITH ORGANIC FERTILIZER AND STEEL SLAG IN A LEGAL RESERVE AREA. **Agroecossistemas**, *[S. l.]*, v. 13, n. 1, p. 31–45, 2021.

KAVAMURA, V. N.; MELO, I. S. de. Effects of different osmolarities on bacterial biofilm formation. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 45, n. 2, p. 627–631, 2014. Disponível em: https://doi.org/10.1590/S1517-83822014000200034

KHODAVERDILOO, H. *et al.* Potentially toxic element contamination of arid and semi-arid soils and its phytoremediation. **Arid Land Research and Management**, *[S. l.]*, v. 34, n. 4, p. 361–391, 2020. Disponível em: https://doi.org/10.1080/15324982.2020.1746707. Acesso em: 20 mar. 2021.

KUMAR, P.; SHUKLA, S. Utilization of steel slag waste as construction material: A review. **Materials Today: Proceedings**, *[S. l.]*, v. 78, p. 145–152, 2023. Disponível em: https://doi.org/10.1016/j.matpr.2023.01.015

KURNIATI, E. O.; PEDERSON, F.; KIM, H.-J. Application of steel slags, ferronickel slags, and copper mining waste as construction materials: A review. **Resources, Conservation and Recycling**, *[S. l.]*, v. 198, p. 107175, 2023. Disponível em: https://doi.org/10.1016/j.resconrec.2023.107175

LARRABURU, E. E.; APÓSTOLO, N. M.; LLORENTE, B. E. Anatomy and morphology of photinia (Photinia × fraseri Dress) in vitro plants inoculated with rhizobacteria. **Trees**, *[S. l.]*, v. 24, n. 4, p. 635–642, 2010. Disponível em: https://doi.org/10.1007/s00468-010-0433-x

LEE, D. J. Achievements in forest tree genetic improvement in Australia and New Zealand 2: Development of Corymbia species and hybrids for plantations in eastern Australia.

http://dx.doi.org/10.1080/00049158.2007.10676256, *[S. l.]*, v. 70, n. 1, p. 11–16, 2013. Disponível em: https://doi.org/10.1080/00049158.2007.10676256. Acesso em: 25 abr. 2023.

LI, Y. *et al.* Toxic effects of cadmium on the physiological and biochemical attributes of plants, and phytoremediation strategies: A review. **Environmental Pollution**, *[S. l.]*, v. 325, p. 121433, 2023. Disponível em: https://doi.org/10.1016/j.envpol.2023.121433

LIMA, J. V. *et al.* Rhizobacteria modify root architecture and improve nutrient uptake in oil palm seedlings despite reduced fertilizer. **Rhizosphere**, *[S. l.]*, p. 100420, 2021. Disponível em: https://doi.org/10.1016/j.rhisph.2021.100420. Acesso em: 12 ago. 2021.

LINCOLN TAIZ; EDUARDO ZEIGER; IAN MAX MØLLER; ANGUS MURPHY. **Fisiologia e desenvolvimento vegetal**. 6. ed. Porto Alegre: Artmed, 2017. *E-book*.

LÓPEZ-BUCIO, J. *et al.* Bacillus megaterium Rhizobacteria Promote Growth and Alter Root-System Architecture Through an Auxin- and Ethylene-Independent Signaling Mechanism in Arabidopsis thaliana. **Molecular Plant-Microbe Interactions®**, *[S. l.]*, v. 20, n. 2, p. 207–217, 2007. Disponível em: https://doi.org/10.1094/MPMI-20-2-0207

MA, Y. *et al.* Plant growth promoting rhizobacteria and endophytes accelerate phytoremediation of metalliferous soils. **Biotechnology Advances**, *[S. l.]*, v. 29, n. 2, p. 248–258, 2011. Disponível em: https://doi.org/10.1016/J.BIOTECHADV.2010.12.001

MA, Y. *et al.* **Beneficial role of bacterial endophytes in heavy metal phytoremediation**. *[S. l.: s. n.]* Disponível em: https://doi.org/10.1016/j.jenvman.2016.02.047

MADHAIYAN, M.; POONGUZHALI, S.; SA, T. Metal tolerating methylotrophic bacteria reduces nickel and cadmium toxicity and promotes plant growth of tomato (Lycopersicon esculentum L.). **Chemosphere**, *[S. l.]*, v. 69, n. 2, p. 220–228, 2007. Disponível em: https://doi.org/10.1016/j.chemosphere.2007.04.017

MAËLA PERON GOMIDE. **ANATOMIA DE RAÍZES DE Eucalyptus grandis Hill ex Maiden EM DIFERENTES PROFUNDIDADES DO SOLO SOB UM CONTEXTO DE REDUÇÃO DE CHUVAS**. 2016. - Universidade Estadual Paulista "Júlio de Mesquita Filho", *[s. l.]*, 2016.

MAFIA, R. G. *et al.* Rooting induction and growth of eucalyptus by rhizobacteria: Effect of addition of nutrient source and growing substrate composition. **Revista Arvore**, *[S. l.]*, v. 31, n. 4, p. 589–597, 2007 a. Disponível em: https://doi.org/10.1590/s0100-67622007000400003.

Acesso em: 22 abr. 2021.

MAFIA, R. G. *et al.* Compatibility among rhizobacteria isolates and effect of mixture of isolates in inducing rooting and growth of eucalyptus clones. **Revista Arvore**, *[S. l.]*, v. 31, n. 4, p. 635–643, 2007 b. Disponível em: https://doi.org/10.1590/S0100-67622007000400008

MAFIA, R. G. *et al.* Indução do enraizamento e crescimento do eucalipto por rizobactérias: efeito da adição de fonte alimentar e da composição do substrato de enraizamento. **Revista Árvore**, *[S. l.]*, v. 31, n. 4, p. 589–597, 2007 c. Disponível em: https://doi.org/10.1590/S0100-67622007000400003

MAFIA, R. G. *et al.* Microbiolização e interação entre rizobactérias promotoras do crescimento e clones de eucalipto. **Revista Árvore**, *[S. l.]*, v. 33, n. 5, p. 789–797, 2009. Disponível em: https://doi.org/10.1590/S0100-67622009000500002

MAGALHÃES, M. O. L. *et al.* Potencial de duas espécies de eucalipto na fitoestabilização de solo contaminado com zinco. **Revista Ciência Agronômica**, *[S. l.]*, v. 42, n. 3, p. 805–812, 2011. Disponível em: https://doi.org/10.1590/S1806-66902011000300029

MARTÍNEZ-VIVEROS, O. *et al.* MECHANISMS AND PRACTICAL CONSIDERATIONS INVOLVED IN PLANT GROWTH PROMOTION BY RHIZOBACTERIA. **Journal of soil science and plant nutrition**, *[S. l.]*, v. 10, n. 3, 2010. Disponível em: https://doi.org/10.4067/S0718-95162010000100006

MARTINS, M. S. **INOVAÇÕES TECNOLÓGICAS DA INDÚSTRIA 4.0: APLICAÇÕES E IMPLICAÇÕES PARA A SIDERURGIA BRASILEIRA**. 2019. - Universidade Federal de Uberlândia, *[s. l.]*, 2019.

MATSUMOTO, S. *et al.* Inhibition of arsenic accumulation in Japanese rice by the application of iron and silicate materials. **CATENA**, *[S. l.]*, v. 135, p. 328–335, 2015. Disponível em: https://doi.org/10.1016/J.CATENA.2015.07.004

MEENA, K. K. *et al.* Abiotic Stress Responses and Microbe-Mediated Mitigation in Plants: The Omics Strategies. **Frontiers in Plant Science**, *[S. l.]*, v. 8, 2017 a. Disponível em: https://doi.org/10.3389/fpls.2017.00172

MEENA, V. S. *et al.* Plant beneficial rhizospheric microorganism (PBRM) strategies to improve nutrients use efficiency: A review. **Ecological Engineering**, *[S. l.]*, v. 107, p. 8–32, 2017 b. Disponível em: https://doi.org/10.1016/j.ecoleng.2017.06.058

MOURÃO, M. **Introdução à siderurgia**. *[S. l.: s. n.]*. *E-book*.

NAUTIYAL, C. S. An efficient microbiological growth medium for screening phosphate solubilizing microorganisms. **FEMS Microbiology Letters**, *[S. l.]*, v. 170, n. 1, p. 265–270, 1999. Disponível em: https://doi.org/10.1111/J.1574-6968.1999.TB13383.X. Acesso em: 19 abr. 2022.

NING, D. *et al.* Impacts of steel-slag-based silicate fertilizer on soil acidity and silicon availability and metals-immobilization in a paddy soil. **PLoS ONE**, *[S. l.]*, v. 11, n. 12, 2016. Disponível em: https://doi.org/10.1371/journal.pone.0168163

NJOKU, K. L.; AKINYEDE, O. R.; OBIDI, O. F. Microbial Remediation of Heavy Metals Contaminated Media by Bacillus megaterium and Rhizopus stolonifer. **Scientific African**, *[S. l.]*, v. 10, p. e00545, 2020. Disponível em: https://doi.org/10.1016/j.sciaf.2020.e00545

NWIGWE, C.; FOSSEY, A.; DE SMIDT, O. Effects of Plant Growth Promoting Rhizospheric Bacteria (PGPR) on Survival, Growth and Rooting Architecture of Eucalyptus Hybrid Clones. **Forests**, *[S. l.]*, v. 14, n. 9, p. 1848, 2023. Disponível em: https://doi.org/10.3390/f14091848

O'CONNOR, J. *et al.* Production, characterisation, utilisation, and beneficial soil application of steel slag: A review. **Journal of Hazardous Materials**, *[S. l.]*, v. 419, p. 126478, 2021. Disponível em: https://doi.org/10.1016/J.JHAZMAT.2021.126478

OLEŃSKA, E. *et al.* Beneficial features of plant growth-promoting rhizobacteria for improving plant growth and health in challenging conditions: A methodical review. **Science of The Total Environment**, *[S. l.]*, v. 743, p. 140682, 2020. Disponível em: https://doi.org/10.1016/J.SCITOTENV.2020.140682

OLIVEIRA, A. M. de; CENÁRIO. **CENÁRIO BRASILEIRO DE INOCULANTES E APLICAÇÕES DE BACTÉRIAS ENDOFÍTICAS EM Corymbia**. 2023. - UNIVERSIDADE FEDERAL DOS VALES DO JEQUITINHONHA E MUCURI, *[s. l.]*, 2023.

OZA, E. F. *et al.* Aproveitamento de escória de siderurgia em substratos alternativos para produção de mudas de pimenteira Dedo-de-moça. **Revista Ceres**, *[S. l.]*, v. 65, n. 1, p. 104–109, 2018. Disponível em: https://doi.org/10.1590/0034-737x201865010014

PAN, X. *et al.* Exploring root system architecture and anatomical variability in alfalfa (Medicago sativa L.) seedlings. **BMC Plant Biology**, *[S. l.]*, v. 23, n. 1, p. 449, 2023. Disponível em: https://doi.org/10.1186/s12870-023-04469-4

PANDIT, A. *et al.* Microbial biofilms in nature: unlocking their potential for agricultural applications. **Journal of applied microbiology**, *[S. l.]*, v. 129, n. 2, p. 199–211, 2020. Disponível em: https://doi.org/10.1111/JAM.14609. Acesso em: 12 dez. 2022.

PAZ, I. C. P. *et al.* Eucalyptus growth promotion by endophytic Bacillus spp. **Genetics and Molecular Research**, *[S. l.]*, v. 11, n. 4, p. 3711–3720, 2012. Disponível em: https://doi.org/10.4238/2012.August.17.9

PERALTA, K. D. *et al.* Production of phytohormones, siderophores and population fluctuation of two root-promoting rhizobacteria in Eucalyptus globulus cuttings. **World Journal of Microbiology and Biotechnology**, *[S. l.]*, v. 28, n. 5, p. 2003–2014, 2012. Disponível em: https://doi.org/10.1007/s11274-012-1003-8. Acesso em: 22 abr. 2021.

PIATAK, N. M.; PARSONS, M. B.; SEAL, R. R. Characteristics and environmental aspects of slag: A review. **Applied Geochemistry**, *[S. l.]*, v. 57, p. 236–266, 2015. Disponível em: https://doi.org/10.1016/j.apgeochem.2014.04.009

PII, Y. *et al.* **Microbial interactions in the rhizosphere: beneficial influences of plant growth-promoting rhizobacteria on nutrient acquisition process. A review**. *[S. l.: s. n.]* Disponível em: https://doi.org/10.1007/s00374-015-0996-1

PISHCHIK, V. N. *et al.* **Mechanisms of plant and microbial adaptation to heavy metals in plant–microbial systems**. *[S. l.]*: Maik Nauka Publishing / Springer SBM, 2016. Disponível em: https://doi.org/10.1134/S0026261716030097. Acesso em: 1 abr. 2020.

PRADO, R. de M.; FERNANDES, F. M. Efeito da escória de siderurgia e calcário na disponibilidade de fósforo de um Latossolo Vermelho-Amarelo cultivado com cana-de-açúcar. **Pesquisa Agropecuária Brasileira**, *[S. l.]*, v. 36, n. 9, p. 1199–1204, 2001. Disponível em: https://doi.org/10.1590/S0100-204X2001000900014

RAASCH, L. D.; BONALDO, S. M.; OLIVEIRA, A. A. F. de. Bacillus subtilis: ENRAIZAMENTO E CRESCIMENTO DE MINIESTACAS DE EUCALIPTO EM SINOP, NORTE DE MATO GROSSO, BRASIL. **Biosci. J.**, *[S. l.]*, v. 29, n. Supplement 1, p. 1446–1457, 2013.

RADIĆ, S. *et al.* Effect of electric arc furnace slag on growth and physiology of maize (Zea mays L.). **Acta Biologica Hungarica**, *[S. l.]*, v. 64, n. 4, p. 490–499, 2013. Disponível em: https://doi.org/10.1556/ABiol.64.2013.4.8

RAJKUMAR, M.; FREITAS, H. Influence of metal resistant-plant growth-promoting bacteria on the growth of Ricinus communis in soil contaminated with heavy metals. **Chemosphere**, *[S. l.]*, v. 71, n. 5, p. 834–842, 2008. Disponível em: https://doi.org/10.1016/j.chemosphere.2007.11.038. Acesso em: 13 nov. 2022.

RAJKUMAR, M.; MA, Y.; FREITAS, H. Improvement of Ni phytostabilization by inoculation of Ni resistant Bacillus megaterium SR28C. **Journal of Environmental Management**, *[S. l.]*, v. 128, p. 973–980, 2013. Disponível em: https://doi.org/10.1016/J.JENVMAN.2013.07.001

RÊGO, M. C. F. *et al.* Morphoanatomical and Biochemical Changes in the Roots of Rice Plants Induced by Plant Growth-Promoting Microorganisms. **Journal of Botany**, *[S. l.]*, v. 2014, p. 1–10, 2014. Disponível em: https://doi.org/10.1155/2014/818797

RÊGO, M. C. F. *et al.* The role of rhizobacteria in rice plants: Growth and mitigation of toxicity. **Journal of Integrative Agriculture**, *[S. l.]*, v. 17, n. 12, p. 2636–2647, 2018. Disponível em: https://doi.org/10.1016/S2095-3119(18)62039-8

REIS, C. A. F. *et al.* **Corymbia citriodora: estado da arte de pesquisas no Brasil**. *[s. l.]*, 2013. Disponível em: https://ainfo.cnptia.embrapa.br/digital/bitstream/item/94547/1/Doc.-255-Reis-Ainfo1.pdf. Acesso em: 10 out. 2023.

REIS, C. A. F. *et al.* **Corymbia torelliana: estado da arte de pesquisas no Brasil**. Colombo: Embrapa Florestas, 2014.

RODRÍGUEZ-DÍAZ, M., RODELAS-GONZALÉS, B., POZO-CLEMENTE, C, MARTÍNEZ-TOLEDO, MV, GONZÁLEZ-LÓPEZ, J. Uma revisão sobre a taxonomia e possíveis características de triagem de rizobactérias promotoras de crescimento de plantas. *In*: AHMAD, J. PICHTEL, J., S. H. (org.). **Interações Planta-Bactérias: Estratégias e Técnicas para Promover o Crescimento das Plantas**. Weinheim, Alemanh: Wiley-VCH Verlag GmbH & Co., 2008. p. 55–80. *E-book*.

ROZEFELDS, A. C. Eucalyptus phylogeny and history: A Brief Summary. **Tasforests**, *[S. l.]*, v. 8, p. 11, 1996.

SAMANIEGO-GÁMEZ, B. Y. *et al.* Bacillus spp. inoculation improves photosystem II efficiency and enhances photosynthesis in pepper plants. **Chilean journal of agricultural research**, *[S. l.]*, v. 76, n. 4, p. 409–416, 2016. Disponível em: https://doi.org/10.4067/S0718-58392016000400003. Acesso em: 18 out. 2022.

SANTOS, L. F. dos; OLIVARES, F. L. Microbial inoculants in agriculture and its effects on plant microbiome. *In*: **Rhizosphere Engineering**. *[S. l.]*: Elsevier, 2022. p. 151–169. *E-book*. Disponível em: https://doi.org/10.1016/B978-0-323-89973-4.00016-8

SCHWYN, B.; NEILANDS, J. B. Universal chemical assay for the detection and determination of siderophores. **Analytical Biochemistry**, *[S. l.]*, v. 160, n. 1, p. 47–56, 1987. Disponível em: https://doi.org/10.1016/0003-2697(87)90612-9

SHARMA, P. Role and significance of biofilm-forming microbes in phytoremediation -A review. **Environmental Technology & Innovation**, *[S. l.]*, v. 25, p. 102182, 2022. Disponível em: https://doi.org/10.1016/J.ETI.2021.102182

SHENG, X.-F. *et al.* Characterization of heavy metal-resistant endophytic bacteria from rape (Brassica napus) roots and their potential in promoting the growth and lead accumulation of rape. **Environmental Pollution**, *[S. l.]*, v. 156, n. 3, p. 1164–1170, 2008. Disponível em: https://doi.org/10.1016/j.envpol.2008.04.007

SHEPHERD, M. *et al.* Mapping species differences for adventitious rooting in a Corymbia torelliana × Corymbia citriodora subspecies variegata hybrid. **Tree Genetics & Genomes**, *[S. l.]*, v. 4, n. 4, p. 715–725, 2008. Disponível em: https://doi.org/10.1007/s11295-008-0145-1

SHI, R. yong *et al.* Effects of biomass ash, bone meal, and alkaline slag applied alone and combined on soil acidity and wheat growth. **Journal of Soils and Sediments**, *[S. l.]*, v. 17, n. 8, p. 2116–2126, 2017. Disponível em: https://doi.org/10.1007/S11368-017-1673-9/FIGURES/6. Acesso em: 3 nov. 2022.

SHIN, M.-N. *et al.* Characterization of lead resistant endophytic Bacillus sp. MN3-4 and its potential for promoting lead accumulation in metal hyperaccumulator Alnus firma. **Journal of Hazardous Materials**, *[S. l.]*, v. 199–200, p. 314–320, 2012. Disponível em: https://doi.org/10.1016/j.jhazmat.2011.11.010

SHU, K.; SASAKI, K. Occurrence of steel converter slag and its high value-added conversion for environmental restoration in China: A review. **Journal of Cleaner Production**, *[S. l.]*, v. 373, p. 133876, 2022. Disponível em: https://doi.org/10.1016/J.JCLEPRO.2022.133876

SILVA, A. V. da *et al.* Physiological, biochemical and nutritional changes in soybean in response to application of steel slag. **Australian Journal of Crop Science**, *[S. l.]*, v. 15, n. 15(12):2021, p. 1399–1405, 2021. Disponível em: https://doi.org/10.21475/ajcs.21.15.12.p3269.

Acesso em: 27 ago. 2022.

SINGH, V. *et al.* Plant Growth Promoting Rhizobacteria (PGPR): Current and Future Prospects for Development of Sustainable Agriculture. **J Microb Biochem Technol**, *[S. l.]*, v. 7, n. 2, p. 96–102, 2015. Disponível em: https://doi.org/10.4172/1948-5948.1000188

SOBRAL, M. F. *et al.* Basic slag and its effects on the concentration of nutrients and heavy metals in sugarcane. **Revista Brasileira de Engenharia Agricola e Ambiental**, *[S. l.]*, v. 15, n. 8, p. 867–872, 2011. Disponível em: https://doi.org/10.1590/S1415-43662011000800015

TEIXEIRA, D. A. *et al.* RHIZOBACTERIAL PROMOTION OF EUCALYPT ROOTING AND GROWTH. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 38, p. 118–123, 2007 a. Disponível em: www.cme.msu.edu/RDP/

TEIXEIRA, D. A. *et al.* Rhizobacterial promotion of eucalypt rooting and growth. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 38, n. 1, p. 118–123, 2007 b. Disponível em: https://doi.org/10.1590/S1517-83822007000100025

TIWARI, M.; BAJPAI, D. S.; DEWANGAN, D. U. Steel Slag Utilization — Overview in Indian Perspective. **International Journal of Advanced Research**, *[S. l.]*, v. 4, n. 8, p. 2232–2246, 2016. Disponível em: https://doi.org/10.21474/IJAR01/1442

TRUEMAN, S. J. Cytokinin and auxin effects on survival and rooting of Eucalyptus pellita and E. grandis × E. pellita cuttings. **Rhizosphere**, *[S. l.]*, v. 6, p. 74–76, 2018. Disponível em: https://doi.org/10.1016/J.RHISPH.2018.04.001

TRUEMAN, S. J.; RICHARDSON, D. M. Relationships between Indole-3-Butyric Acid, Photoinhibition and Adventitious Rooting of Corymbia torelliana, C. citriodora and F 1 Hybrid Cuttings. **Tree and Forestry Science and Biotechnology**, *[S. l.]*, v. 2, n. 1, p. 26–33, 2008.

TU, X. *et al.* Advantages and risks of using steel slag in preparing composts from raw organic waste. **Journal of Environmental Science and Health - Part B Pesticides, Food Contaminants, and Agricultural Wastes**, *[S. l.]*, v. 52, n. 1, p. 30–36, 2017. Disponível em: https://doi.org/10.1080/03601234.2016.1224701

VALENTE LIMA, J. *et al.* Rhizobacteria modify root architecture and improve nutrient uptake in oil palm seedlings despite reduced fertilizer. **Rhizosphere**, *[S. l.]*, v. 19, p. 100420, 2021. Disponível em: https://doi.org/10.1016/J.RHISPH.2021.100420

VEJAN, P. *et al.* Role of Plant Growth Promoting Rhizobacteria in Agricultural Sustainability—

A Review. **Molecules 2016, Vol. 21, Page 573**, *[S. l.]*, v. 21, n. 5, p. 573, 2016. Disponível em: https://doi.org/10.3390/MOLECULES21050573. Acesso em: 13 fev. 2023.

VERMA, S.; KUILA, A. Bioremediation of heavy metals by microbial process. **Environmental Technology & Innovation**, *[S. l.]*, v. 14, p. 100369, 2019. Disponível em: https://doi.org/10.1016/j.eti.2019.100369

VOCCIANTE, M. *et al.* The Role of Plant Growth-Promoting Rhizobacteria (PGPR) in Mitigating Plant's Environmental Stresses. **Applied Sciences**, *[S. l.]*, v. 12, n. 3, p. 1231, 2022. Disponível em: https://doi.org/10.3390/app12031231

WANG, G. *et al.* The combined use of a plant growth promoting Bacillus sp. strain and GABA promotes the growth of rice under salt stress by regulating antioxidant enzyme system, enhancing photosynthesis and improving soil enzyme activities. **Microbiological Research**, *[S. l.]*, v. 266, p. 127225, 2023. Disponível em: https://doi.org/10.1016/J.MICRES.2022.127225

WANG, J. *et al.* Bacillus velezensis SX13 promoted cucumber growth and production by accelerating the absorption of nutrients and increasing plant photosynthetic metabolism. **Scientia Horticulturae**, *[S. l.]*, v. 301, p. 111151, 2022. Disponível em: https://doi.org/10.1016/J.SCIENTA.2022.111151

WANG, X. *et al.* Isolation of As-tolerant bacteria and their potentials of reducing As and Cd accumulation of edible tissues of vegetables in metal(loid)-contaminated soils. **Science of the Total Environment**, *[S. l.]*, v. 579, 2017. Disponível em: https://doi.org/10.1016/j.scitotenv.2016.10.239

WANG, X. *et al.* Environmental risks for application of iron and steel slags in soils in China: A review. **Pedosphere**, *[S. l.]*, v. 31, n. 1, p. 28–42, 2021. Disponível em: https://doi.org/10.1016/S1002-0160(20)60058-3

WANI, P. A.; KHAN, M. S. Bacillus species enhance growth parameters of chickpea (Cicer arietinum L.) in chromium stressed soils. **Food and Chemical Toxicology**, *[S. l.]*, v. 48, n. 11, p. 3262–3267, 2010. Disponível em: https://doi.org/10.1016/j.fct.2010.08.035

WHITE, B. *et al.* Effect of silicate slag application on wheat grown under two nitrogen rates. **Plants**, *[S. l.]*, v. 6, n. 4, 2017. Disponível em: https://doi.org/10.3390/plants6040047

WORLD STEEL ASSOCIATION. **Sustainable steel: Sustainability Indicators 2021**. Beijing: China: World Steel, 2022. Disponível em:

https://worldsteel.org/publications/bookshop/sust2021/

YI, H. *et al.* An Overview of Utilization of Steel Slag. **Procedia Environmental Sciences**, *[S. l.]*, v. 16, p. 791–801, 2012. Disponível em: https://doi.org/10.1016/j.proenv.2012.10.108

YILDIRIM, I. Z.; PREZZI, M. Chemical, Mineralogical, and Morphological Properties of Steel Slag. **Advances in Civil Engineering**, *[S. l.]*, v. 2011, p. 1–13, 2011. Disponível em: https://doi.org/10.1155/2011/463638

ZANONI DO PRADO, D. *et al.* Aspergillus flavipes as a novel biostimulant for rooting-enhancement of Eucalyptus. **Journal of Cleaner Production**, *[S. l.]*, v. 234, p. 681–689, 2019. Disponível em: https://doi.org/10.1016/J.JCLEPRO.2019.06.211

3. Bacillus sp. ENHANCES NUTRIENT USE EFFICIENCY AND GROWTH OF *Corymbia* SEEDLINGS FERTILIZED WITH STEELMAKING RESIDUE, ALTERING PHOTOSYNTHETIC AND BIOCHEMICAL RESPONSES

ABSTRACT

Steel slag is a byproduct of primary steel refining, rich in Ca, Mg, Fe, Mn, and Zn, providing potential for agronomic use as a soil amendment and fertilizer. However, it may contain toxic metallic contaminants such as Cr and Pb, posing an environmental management challenge. Plant growth-promoting microorganisms can accumulate, transform, or detoxify heavy metals, contributing to plant growth by improving nutrient solubilization and absorption. This study aimed to evaluate, for the first time, the effects of the combination of electric arc furnace steel slag (EAE) and *Bacillus* sp. on photosynthetic, biochemical, nutritional, and growth parameters of *Corymbia torelliana* x *Corymbia citriodora* hybrid seedlings. The experimental design was completely randomized in a factorial scheme with two factors: Inoculation (non-inoculated and *Bacillus* sp.) and EAE (0% and 1%), resulting in 4 treatments with 8 replicates each. The *Bacillus* sp. + EAE combination promoted increases in root length, root volume, root dry biomass, and root-to-shoot ratio. Enhanced growth was associated with improved potential activity of photosystem II (*Fv/Fo*), optimizing electron transfer rate (*ETR*), and reducing the non-photochemical quenching coefficient (*qN*), resulting in higher net photosynthesis. Starch accumulation and lower soluble sugar content indicated increased respiration rate and energy production required for the proven growth promotion. No phytotoxicity was observed, and the translocation of Pb, Cr, and Cd was reduced in inoculated plants. The application of EAE in *Corymbia* nutrition gains an additional advantage when associated with *Bacillus* sp. biostimulation, offering a potential strategy to accelerate seedling growth, reduce production costs, and prevent environmental damage. The incorporation of EAE into the *Corymbia* production chain represents a potential route for the disposal of this residue that still accumulates in the country's steel mills.

Keywords: Steel slag. Heavy metals. Photosynthesis. Rhizobacteria. *Corymbia torelliana*. *Corymbia citriodora*.

3.1 Introduction

Brazil is the 9th largest steel producer globally and the 1st in Latin America, having produced 36 million tons in 2021, impacting the trade balance with $4.4 billion in exports (WORLD STEEL ASSOCIATION, 2022). Steel can be produced by reducing iron ore to pig iron in a blast furnace and subsequent refining, or from refined steel scrap in electric arc furnaces (BRANCA et al., 2020). These processes generate by-products or residues such as dust, fines, sludge, and steel aggregates (slag), at a rate of 622 kg per ton of crude steel produced, with 155 kg being steel slag (INSTITUTO AÇO BRASIL, 2021).

Electric arc furnace slag (EAE) produced in arc furnaces is a by-product generated during the steel refining stage at high temperatures, characterized as a complex of oxides, primarily calcium (Ca), magnesium (Mg), iron (Fe), and manganese (Mn). Depending on the raw materials used, furnace type, and additives, EAE may contain toxic metal contaminants such as chromium (Cr), cadmium (Cd), lead (Pb), among others, posing potential environmental pollutants (ITO, 2015).

Once in the soil, potentially toxic elements (PTEs) are non-biodegradable and, at elevated concentrations, can be absorbed and metabolized, impacting plant metabolism and compromising growth and development (JING; HE; YANG, 2007; RAJKUMAR; FREITAS, 2008). However, literature highlights the potential of steel residues in agronomic use, both in soil acidity correction due to the presence of highly soluble basic silicates that elevate pH values and as fertilizers due to the presence of macro and micronutrients (DAS et al., 2020; GWON et al., 2018; NING et al., 2016).

Plant growth-promoting microorganisms (PGPR), including free-living bacteria in the rhizosphere, can benefit plants, contributing to increased growth and yield through direct and/or indirect mechanisms such as biopesticides, biofertilizers, phyto-stimulants, and rhizoremediators (SINGH et al., 2015; WANG et al., 2023). These microorganisms can stimulate nutrient absorption, fix, mineralize, and solubilize crucial elements such as N, P, K, and modify the bioavailability of non-essential elements like some metals by releasing chelators such as organic acids and siderophores (MA et al., 2016; VOCCIANTE et al., 2022). PGPR can also accumulate, transform, or detoxify heavy metals in the soil, originating from EAES, facilitating plant growth (MISHRA; SINGH; ARORA, 2017).

In Brazil, 36% of steel slag remains as a passive stock in steel mills, with only 3% being incorporated into agricultural production (INSTITUTO AÇO BRASIL, 2021). Increasing the utilization of EAE depends on developing strategies to ensure the safety of this by-product in agriculture, mitigating potential risks. One strategy could be the combined use of EAE as a nutritional source and plant growth-promoting rhizobacteria to recycle steel industry waste, contributing to the sustainability of the steel sector.

The objective was to assess the effects of the combination of electric arc furnace steel slag (EAE) and *Bacillus* sp. on chlorophyll *a* fluorescence and gas exchange, chlorophyll and soluble sugar and starch content, nutrient use efficiency, heavy metal absorption, and the growth of seedlings of the hybrid *Corymbia torelliana x Corymbia citriodora* in a greenhouse.

3.2. Materials and Methods

3.2.1 Obtaining PGPR and in vitro EAE tolerance test

The PGPR *Bacillus* sp. isolate, deposited in the Maria de Lourdes Reis Duarte Culture Collection at the Plant Protection Laboratory of the Federal Rural University of the Amazon/UFRA under the code CCMD862, was sourced from the rhizosphere of castor bean (*Ricinus communis* L.) in an area with waste deposits from a steel mill located at 5°24'35.3"S 49°04'42.6"W, in Marabá-PA. Table 1 details morphological characteristics, colony aspects, Gram reaction, genetic identification using the 16S rRNA region, and biochemical mechanisms of plant growth promotion by the rhizobacterium, including nitrogen fixation, biofilm production, indole-3-acetic acid (IAA) synthesis, siderophores, and phosphate solubilization.

To assess the growth of *Bacillus* sp. isolate in the presence of electric arc furnace steel slag (EAE) residue, an in vitro test was conducted on nutrient agar (NA). The rhizobacterium was cultured in nutrient broth (composition: peptone 5.0; meat extract 3.0, in g L^{-1}, pH 6.8 ± 0.2) for 24 hours at 28ºC, with agitation, and the final suspension concentration adjusted to 10^8 CFU mL^{-1}. Subsequently, the suspension was serially diluted to 10^{-2} to 10^{-6}, and 100 µL of dilutions 10^{-2} to 10^{-6} were spread on NA with a sterile Drigalsky loop. EAE concentrations evaluated were 0% (control), 1%, 2%, 4%, 6%, 8%, and 10% at pH 7.0. Plates were incubated at 28ºC for 48h, followed by counting the number of CFU mL^{-1}. The test was conducted with three repetitions for each treatment (EAE concentration), and mean CFU numbers were compared using Tukey's test ($P < 0.05$).

Table 1 Origin, sample, colony morphology and appearance, Gram reaction, taxonomic identification and biochemical characterization of the plant growth promotion mechanisms exhibited by rhizobacterium CCMD862

Origin	Sample	Colony	Gram	Biochemistry*						Taxonomic identification
				BNF[a]	AIA[b]	Biofilm[c]	SP[d]	IS[e]	SK[f]	
PA/ Brazil	Rhizospheric soil	Bright pink	+	+	+	+	+	+	-	*Bacillus* sp.

*[a]Biological nitrogen fixation (HUSSEINY *et al.*, 2021); [b]Indoleacetic acid production (GORDON; WEBER, 1951); [c]Biofilm (KAVAMURA; MELO, 2014); [d]Phosphate solubilization (NAUTIYAL, 1999); [e]Production of siderophores (SCHWYN; NEILANDS, 1987), [f]Potassium solubilization (ALEKSANDROV, VG; BLAGODYR, RN; ILEV, 1967).

3.2.2 Stages and experimental design

Two preliminary trials were conducted to assess the potential of *Bacillus* sp. in promoting plant growth under different concentrations of EAE. The first trial involved rice (*Oryza sativa* cultivar Primavera) as suggested by Cardoso et al. (2021), cultivated under concentrations of 0%, 1%, 2%, and 4% EAE. The second trial used eucalyptus (*Eucalyptus* sp.) cultivated at concentrations of 0%, 1%, and 2% EAE (DATA NOT SHOWN). These trials determined the suitable EAE concentration for eucalyptus growth, and the experiment was repeated.

The EAE residue was generously provided by Siderúrgica Norte Brasil S.A, located in Marabá, Pará. After milling and sieving (fraction ≤ 2mm), 10 g of EAE were incorporated per kg of substrate to compose the 1% EAE treatment. The substrates were placed in plastic bags and incubated for 20 days, homogenized every 2 days, with humidity maintained at 50% of the water retention capacity. After this period, the substrates were transferred to pots with a capacity of 1.5 dm^3, standardized at 1.5 kg. The characteristics of EAE are presented in Table 2.

Table 2. Characterization of steel slag (EAE) used in the work.

Item	*Description*											
Production process	Steel refining - Electric Arc Furnace											
Sample origin	PA/Brazil											
Classification*	Class IIA (non-hazardous / non-inert / non-corrosive / non-reactive)											
Appearance	Solid material, rough and irregular surface texture, gray coloration											
Granulometry	≤ 2.0 mm											
pH	11.6											
Composition**	P	K	Ca	Mg	B	Cu	Fe	Mn	Zn	Cr	Pb	Cd
(g kg^{-1})	31	15	27	11	0.05	1.6	400	14	180	1.7	10.3	0.08

*(ABNT NBR 10004a/10005b/10006c); **Fertility according to MAPA (2017).

The experiment was conducted between September and October 2022 in a greenhouse. The soil used was dystrophic Yellow Latosol with medium texture (P 60.5; K 38.0; Na 6.4 in mg dm^{-3}; Al 0.06; Ca 3.11; Ca+Mg 3.85 in cmolc dm^{-3}; Fe 85.5; Zn 2.5; Cu 2.2; Mn 20.2 in mg kg^{-1}, pH 5.7). No additional fertilization was performed, except for the incorporated slag. The "torelliodora" hybrid seedlings were obtained from the cross between the parental clones *Corymbia torelliana* (F.Muell.) K.D.Hill & L.A.S.Johnson x *Corymbia citriodora* (Hook.) K.D.Hill & L.A.S.Johnson. The seedlings, acquired from a commercial nursery, were 40 days old, approximately 24 cm in height, and had 13 leaves in excellent phytosanitary condition. This hybrid was chosen for its prominence in eucalyptus-producing areas in the state of Pará, destined for charcoal production.

The experimental design was completely randomized in a factorial scheme with two factors: a) Inoculation (non-inoculated and *Bacillus* sp.) and b) EAE concentrations of 0% and 1%, resulting in 4 treatments with 8 repetitions, totaling 32 experimental units.

The bacterial inoculum was prepared following the methodology proposed by de Castro et al. (2020), cultivating the rhizobacteria in nutrient broth for 48 hours at 28 ºC, then suspending it in sterile distilled water with the concentration adjusted to 10^8 CFU mL^{-1} using a spectrophotometer.

For inoculation, the seedlings had their root system apex cut by approximately 1 cm after being removed from the tubes. They were then inoculated with 10 mL of the bacterial suspension for 30 minutes and immediately transplanted. After 15 days, a new inoculation was performed with 10 mL of the bacterial suspension via irrigation at the plant base. Plants in the control treatment received the same volume of sterile distilled water.

The experiment lasted for 21 days, with daily manual watering to compensate for evapotranspiration losses. The watering volume was determined gravimetrically according to Klar et al. (1966), totaling 30 mL per day.

The evaluation of *Corymbia* growth involved the analysis of gas exchange and photosynthetic parameters. Gas exchange and photosynthetic parameters were estimated on the first or second pair of physiologically mature and fully expanded leaves, from apex to base, when the seedlings were 61 days old. Photosynthetic rate (*A*), stomatal conductance to water vapor (*gs*), intercellular CO_2 concentration (*Ci*), and transpiration rate (*E*) were measured simultaneously with chlorophyll *a* fluorescence parameter using an open gas exchange system with an Infrared Gas Analyzer (IRGA – LI 6400XT, LI-COR, Lincoln, NE, USA) coupled with a fluorometer (LI-6400-40, LI-COR Inc.). Measurements were taken between 10 and 12 hours under specified experimental conditions. The IRGA chamber maintained environmental conditions at 25 °C, vapor pressure deficit between 1.2 and 1.8 kPa, reference CO_2 concentration of 400 μmol mol^{-1}, and photosynthetically active radiation (PAR) of 1000 μmol photons $m^{-2}s^{-1}$. Blue light was applied at 10% of the photosynthetic photon flux density to maximize stomatal opening. Greenhouse conditions during measurements were air temperature of 34 ± 2 °C, relative humidity of 53 ± 2%, incident radiation of 680 ± 100 μmol $m^{-2}s^{-1}$, and air vapor pressure deficit of 2.1 ± 0.14 KPa. Water use efficiency (*WUE*) was determined by the ratio of total dry mass to water consumption throughout the experiment.

For chlorophyll *a* fluorescence analysis, leaves were dark-adapted for 20 minutes, illuminated with low light, and modulated light pulse (0.03 µmol $m^{-2}s^{-1}$) to obtain the initial fluorescence value (*Fo*). A saturating white light pulse of 8000 µmol $m^{-2}s^{-1}$ was applied for 0.8 s to ensure maximum fluorescence emission (*Fm*). Sampled leaves were then illuminated for 300s with actinic light of 250 µmol $m^{-2}s^{-1}$ to reach steady-state fluorescence (*Fs*). Subsequently, saturating white light pulses were applied to obtain maximum fluorescence (*Fm'*). Actinic light was turned off, and far-red light (2 µmol $m^{-2}s^{-1}$) was applied to estimate light-adapted initial fluorescence (*Fo'*). From these measurements, the following parameters were calculated: potential PSII activity (Fv/Fo = (Fm − Fo)/Fo), electron transport rate (ETR = φPSII.PPFD.0.42), coefficients of photochemical (qP = (Fm' − Fs)/(Fm' − Fo')), non-photochemical (qN = 1 - qP), and alternative non-photochemical quenching (NPQ = (Fm - Fm')/Fm') (MAXWELL; JOHNSON, 2000).

The determination of chlorophyll *a* and *b* content, soluble sugars, and starch was carried out on the same leaf used for gas readings. Plant material was collected using aluminum foil envelopes and instantly frozen in liquid nitrogen, then stored at -20 °C until analysis. Ethanol extraction followed the method of Fernie et al. (2001). Samples were ground with liquid nitrogen, and 5 mg of fresh mass were suspended in 250 µL of 95% ethanol, vortexed, incubated at 80 °C for 20 minutes, centrifuged, and the supernatant collected. This process was repeated with 80% and 50% ethanol. The pellet formed was used for starch determination, and the supernatant (~750 µL) for chlorophyll *a* and *b*, soluble carbohydrate (e.g., glucose, fructose, and sucrose) determination.

Post-extraction, 50 µL of the ethanolic extract were diluted in 120 µL of 95% ethanol. Absorbance readings at 645 and 665 nm were taken using a UV-VIS spectrophotometer (Thermo Scientific Multiskan GO), and concentrations of Chla, Chlb, and Chla + b were estimated according to Porra et al. (1989).

Glucose, fructose, and sucrose were estimated using a continuous enzymatic substrate assay, as described by Fernie et al. (2001). Kinetics were evaluated by NADPH formation, with the addition of Hexokinase, Phosphoglucose isomerase, and Invertase. Calculations were based on the equation: µmol NADPH = Δ OD/ (2.85 x 6.22), corrected for the initial sample mass, and results expressed in µmol glucose, fructose, and sucrose per g fresh mass.

For starch determination, the pellet from ethanol extraction was solubilized in 400 µL 0.2 M KOH, heated to 95 °C. Subsequently, 70 µL of 1 N glacial acetic acid was added for neutralization. For starch degradation, 40 µL of the homogenate were diluted with 60 µL of degradation mix (25 mL 50 mM sodium acetate buffer, pH 4.9; 60 µL amyloglucosidase (1.3 U/µL) + 1.2 µL alpha-amylase (0.15 U/µL)).

The degradation reaction occurred through incubation at 56°C for 1 hour. After the degradation step and the formation of glucose residues from starch present in the samples, 12µL of the obtained degradation extract were diluted in 160µL of reaction medium (mix 2) [15.5mL HEPES buffer (1M KOH + 30mM MgCl2 at pH 7.0) + 480µL ATP 109 mM (60mg/mL) + 480µL NADP+ 48.4mM (36mg/mL) + 80µL glyceraldehyde-6-phosphate dehydrogenase (G6PDH 700U/mL)], using a 96-well microplate and reading at 340 nm on a spectrophotometer.

The kinetics were assessed by NADPH formation, facilitated by the addition of 2 U of Hexokinase (2 U in 2 µL), allowing the estimation of starch content determined as glucose equivalents (Hendriks et al. 2003), according to the equation: µmol of NADPH = Δ OD / (2.85 x 6.22).

3.2.3 Biometry and biomass

The growth promotion was evaluated by biometric measurements and biomass accumulation 21 days after the first bio-stimulation of *Corymbia* seedlings. Plant height and root length were measured with a graduated metal ruler, while stem diameter was obtained using a digital caliper (precision of 0.02 mm). Leaf count was assessed by direct counting of emitted leaves. Leaf area was estimated with a leaf area meter (LICOR-3100) with a precision of 0.1 mm^2. Root volume was measured using the gravimetric method of the graduated cylinder. Seedlings were sectioned into root and aboveground parts, dried in an oven at 50 °C until constant weight, and then weighed for dry biomass determination.

3.2.4 Leaf Content and heavy metal translocation

Dried samples were ground (< 5 mm) in a Wiley mill for determination of Cr, Pb, and Cd contents (USEPA, 2007) in both aboveground and root parts. Translocation of heavy metals from roots to leaves was measured by calculating TF (TF = C leaves / C roots), where C leaves and C roots represent the heavy metal contents in aboveground (mg kg^{-1}) and roots (mg kg^{-1}), respectively. As recommended by Fayiga and Ma (2006), TF values > 1 indicate effective translocation of heavy metals from roots to leaves.

3.2.5 Nutrient use efficiency

The dried samples were ground (< 5 mm) in a Wiley mill and sent to the laboratory for the determination of macro and micronutrient content (MALAVOLTA, E.; VITTI, G. C.; OLIVEIRA, 1997). Based on the dry matter and nutrient content in the plant, nutrient use efficiency (g total dry matter)^2/mg of nutrient in the plant) was evaluated as proposed by Siddiqui & Glass (1981).

3.2.6 Statistical analysis

The data were subjected to normality and homoscedasticity analyses to meet the assumptions of analysis of variance (ANOVA), and when significant, treatment means were compared by the Student's t-test with Bonferroni correction ($p < 0.05$) using IBM® SPSS® Statistics Version 25 (IBM Corp. 2017).

3.3 Results

3.3.1 *Bacillus* sp. Tolerance Test to EAE

Results from the determination of *Bacillus* sp. isolate tolerance to electric arc furnace dust (EAE) concentrations in vitro demonstrated that the rhizobacteria were tolerant to the steelmaking residue, showing 68% higher growth than the control at a concentration of 1% EAE (10 mg/L) and 97% at a concentration of 2% (20 mg/L). From the concentration of 4% (40 mg/L), there was a progressive inhibition of growth compared to the control (Fig 1).

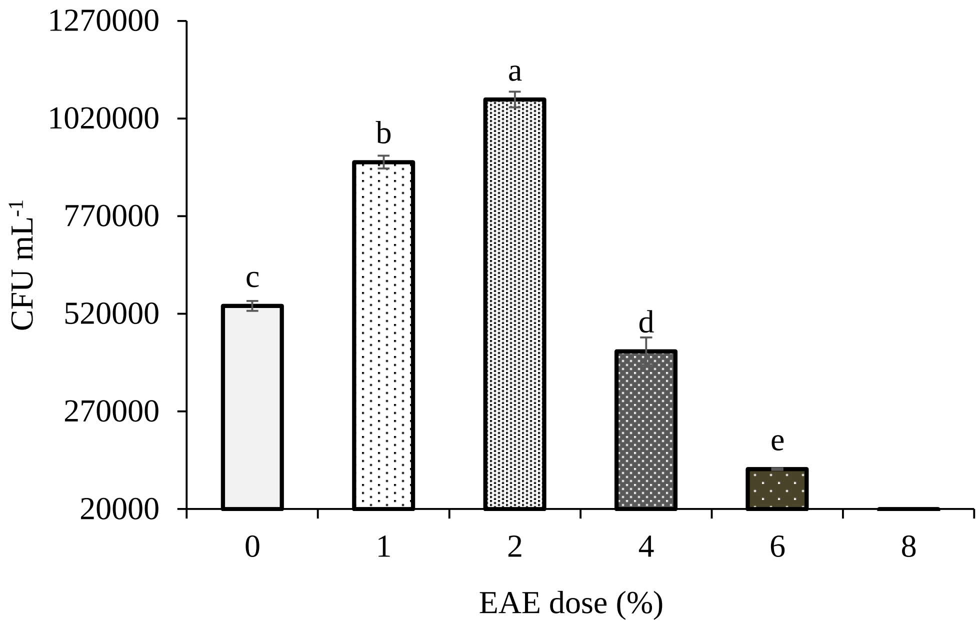


Fig 1 Number of colony-forming units (CFU) of *Bacillus* sp. in culture medium containing doses of electric arc furnace slag (EAE). Means ± standard error with different letters according to Tukey's test ($P < 0.05$).

3.3.2 Photosynthetic parameters

The greatest contribution to chlorophyll *a* fluorescence parameters was attributed to bio-stimulation with rhizobacteria. For *Fv/Fo* and *NPQ*, there was a significant effect of *Bacillus* sp. and EAE individually, as well as their interaction (*Bacillus* sp.+EAE). *Bacillus* sp. and *Bacillus* sp.+ EAE had significant effects on *ETR*, *qP*, and *qN* (Table 3).

In the absence of EAE, *Bacillus* sp. increased *Fv/Fo*, *ETR*, and *qP* while reducing *qN* and *NPQ*. The same response pattern was observed for *Bacillus* sp.+ EAE, with gains of 89.5% in potential PSII activity (*Fv/Fo*), 24% in electron transfer rate (*ETR*), and 18.4% in the photochemical extinction coefficient (*qP*), along with a 10% reduction in the non-photochemical extinction coefficient (*qN*) and a 46% reduction in *NPQ* compared to the uninoculated control without EAE (Fig 2).

Table 3 F-statistics for the effect of rhizobacteria inoculation (*Bacillus* sp.), fertilization with electric arc furnace steel slag (EAE) and the interaction between them (*Bacillus* sp. + EAE) on the biochemical variables of chlorophyll a fluorescence, gas exchange and chlorophyll content in *Corymbia* seedlings.

Variáveis	*Bacillus* sp.	EAE	*Bacillus* sp. + EAE	C.V. (%)
Chlorophyll a fluorescence				
Fv/Fo	75,6*	10,8*	7,4*	13
ETR	334,0*	3,3	20,0*	4
qP	31,1*	1,02	6.75*	11
qN	30,8*	1,03	6,76*	6
NPQ	61.43*	10.44*	9.67*	12
Chlorophyll content				
Chl a	0.41	23.05*	8.86*	7
Chl b	0.21	10.57*	2.95	12
Chl a+b	0,22	21.74*	8.07*	7
Chl a /Chl b	23.14*	12.52*	25.13*	2
Gas exchange				
A	57,19*	0,2	3,8	8
gs	0,8	0,3	5,0*	11
Ci	11,1*	0,06	12,7*	6
E	2,28	1.9	9.61*	10
A/Ci	48,4*	0,02	9,6*	11
WUE	9,6*	222,0*	133*	4

* Significant at $P \leq 0.05$.

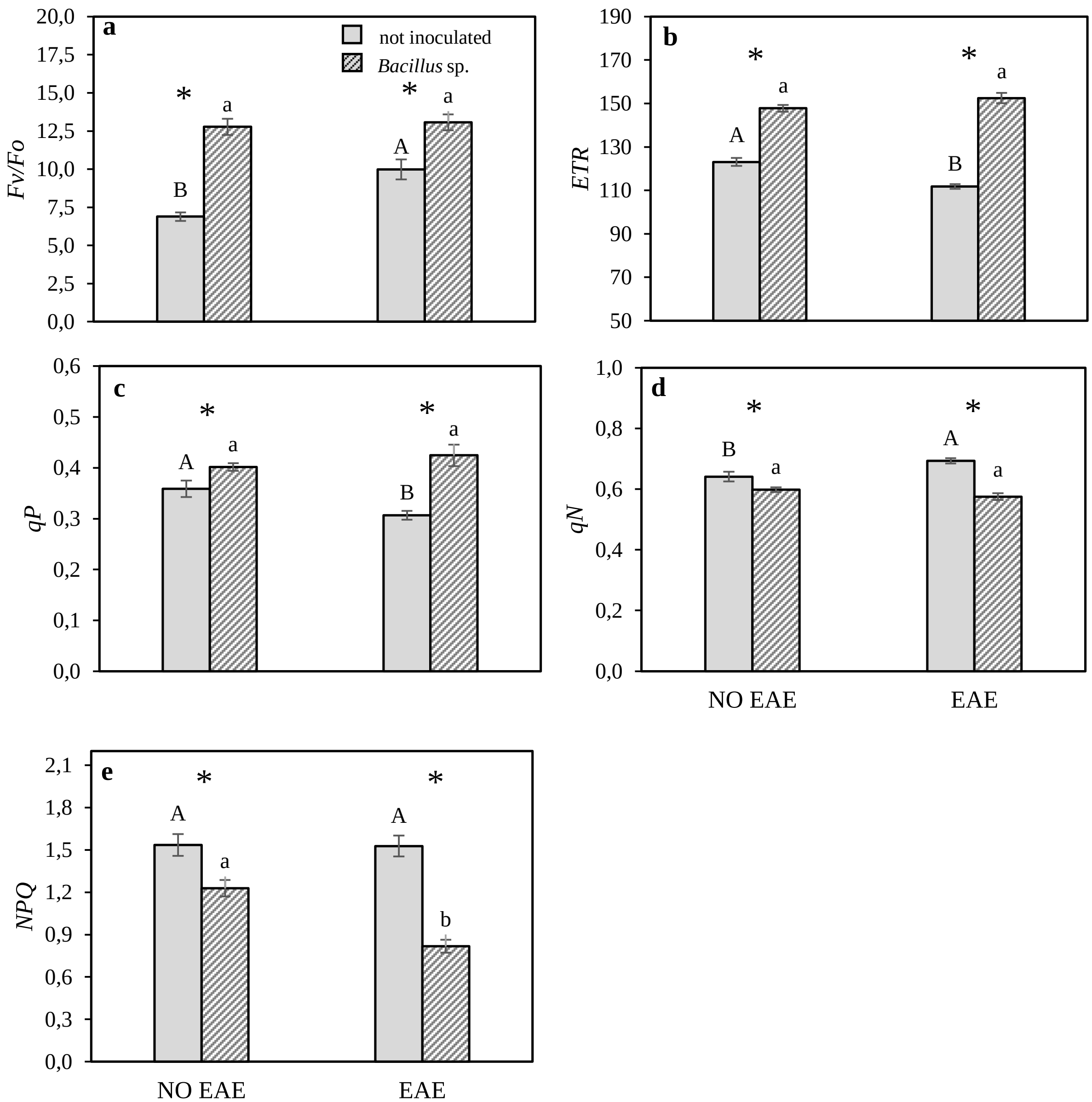


Fig 2 a: potential activity PSII (Fv/Fo), b: electron transfer rate (ETR), c: photochemical extinction coefficient (qP), d: non-photochemical extinction coefficient (qN), e: alternative non-photochemical extinction coefficient (NPQ) in *Corymbia* seedlings treated with *Bacillus* sp., EAE, *Bacillus* sp.+EAE. Uppercase letters compare the effect of the rhizobacteria for each dose of EAE (electric arc furnace slag). Lowercase letters compare the effect of inoculation between doses of EAE. The (*) indicates a difference between treatments within the EAE dose. Averages with equal letters do not differ significantly according to Student's t-test ($P < 0.05$). NO EAE: without the addition of steel waste; EAE: with the addition of steel waste.

The EAE had the greatest effects on chlorophyll contents in eucalyptus seedlings. For both Chl a and Chl a+b, there was an effect of the EAE factor alone and the *Bacillus* sp. + EAE interaction. The Chl b variable was influenced only by EAE. The Chl a/Chl b ratio was influenced by the factors individually and when combined (Table 3).

When evaluated alone, in the absence of EAE, the rhizobacterium promoted increases in Chl a, Chl a+b, and the Chl a/Chl b ratio. The same response pattern was observed with the combination of *Bacillus* sp. + EAE, except for Chl a/Chl b, which showed increases of 16% in Chl a and 76% in Chl a+b compared to the absence of EAE. Gains in Chl b were not statistically significant.

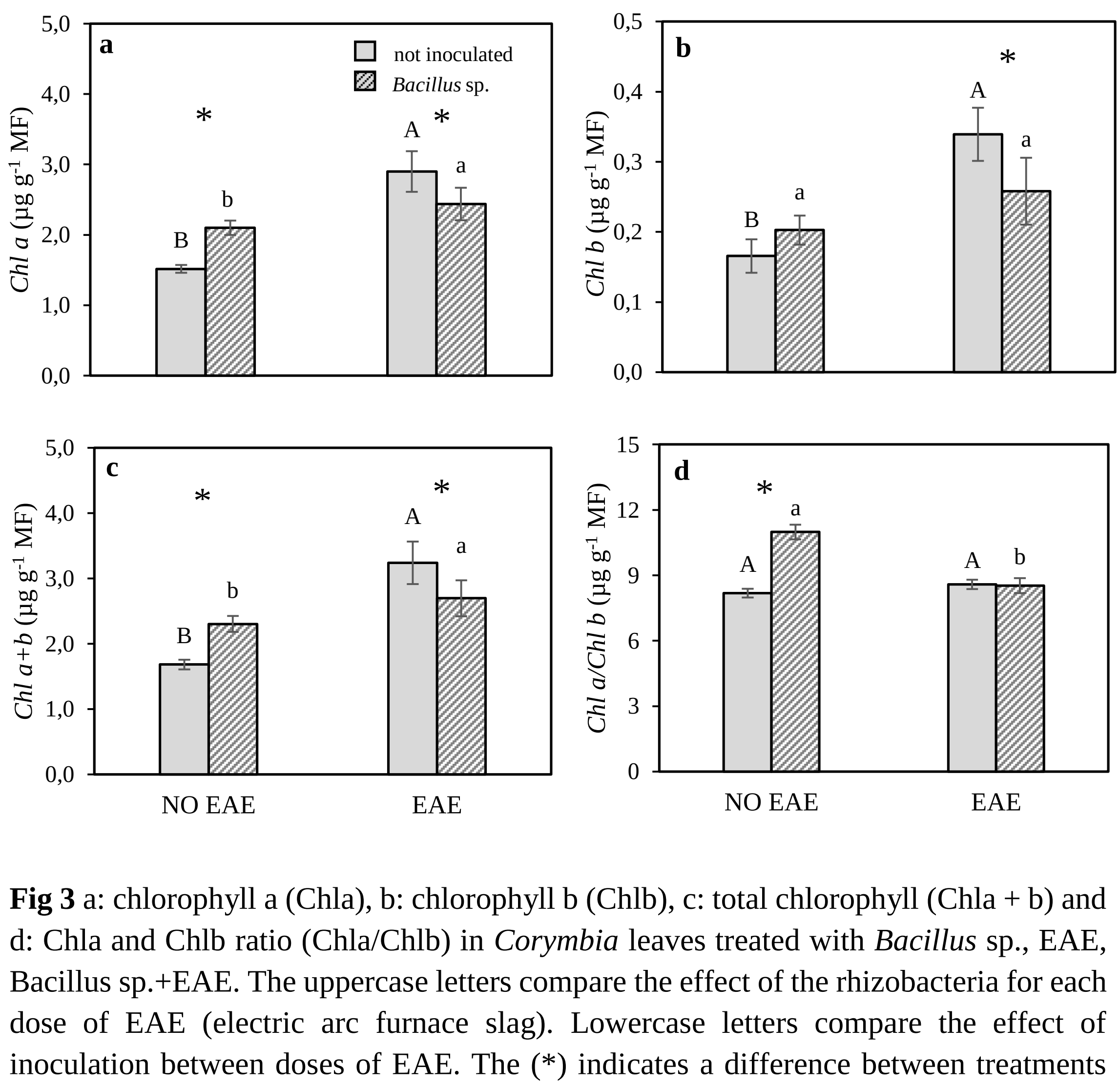


Fig 3 a: chlorophyll a (Chla), b: chlorophyll b (Chlb), c: total chlorophyll (Chla + b) and d: Chla and Chlb ratio (Chla/Chlb) in *Corymbia* leaves treated with *Bacillus* sp., EAE, Bacillus sp.+EAE. The uppercase letters compare the effect of the rhizobacteria for each dose of EAE (electric arc furnace slag). Lowercase letters compare the effect of inoculation between doses of EAE. The (*) indicates a difference between treatments within the EAE dose. Averages with equal letters do not differ significantly according to

Student's t-test ($P < 0.05$). NO EAE: without the addition of steel waste; EAE: with the addition of steel waste.

On the other hand, the independent effect of EAE resulted in the greatest increases in chlorophyll contents, increasing Chl a by 91%, Chl b by 104%, and Chl a+b by 92% compared to plants that did not receive the residue (Fig 3).

3.3.3 Gas exchange

Bio-stimulation with *Bacillus* sp. significantly influenced gas exchange parameters, including *A*, *Ci*, *A/Ci*, and *WUE*. The *Bacillus* sp. + EAE interaction had a significant effect on all analyzed variables except *A*. Only *WUE* was affected by EAE alone (Table 3).

In the absence of EAE, *Bacillus* sp. increased *A* by 21%, *gs* by 12%, *E* by 15%, *A/Ci* by 19%, and *WUE* by 28%. The combination of *Bacillus* sp. + EAE maintained the increase in *A* and *A/Ci* at 27% and 37%, respectively, while reducing *Ci* by 9% and *WUE* by 35%. Comparing the effects of *Bacillus* sp. + EAE to EAE alone, there was an 18% reduction in *Ci*, a 41% increase in *A*, and a 53% increase in *A/Ci*. The combination reduced *WUE* by 11%, but had no effect on *gs* and *E* variables (Fig 4).

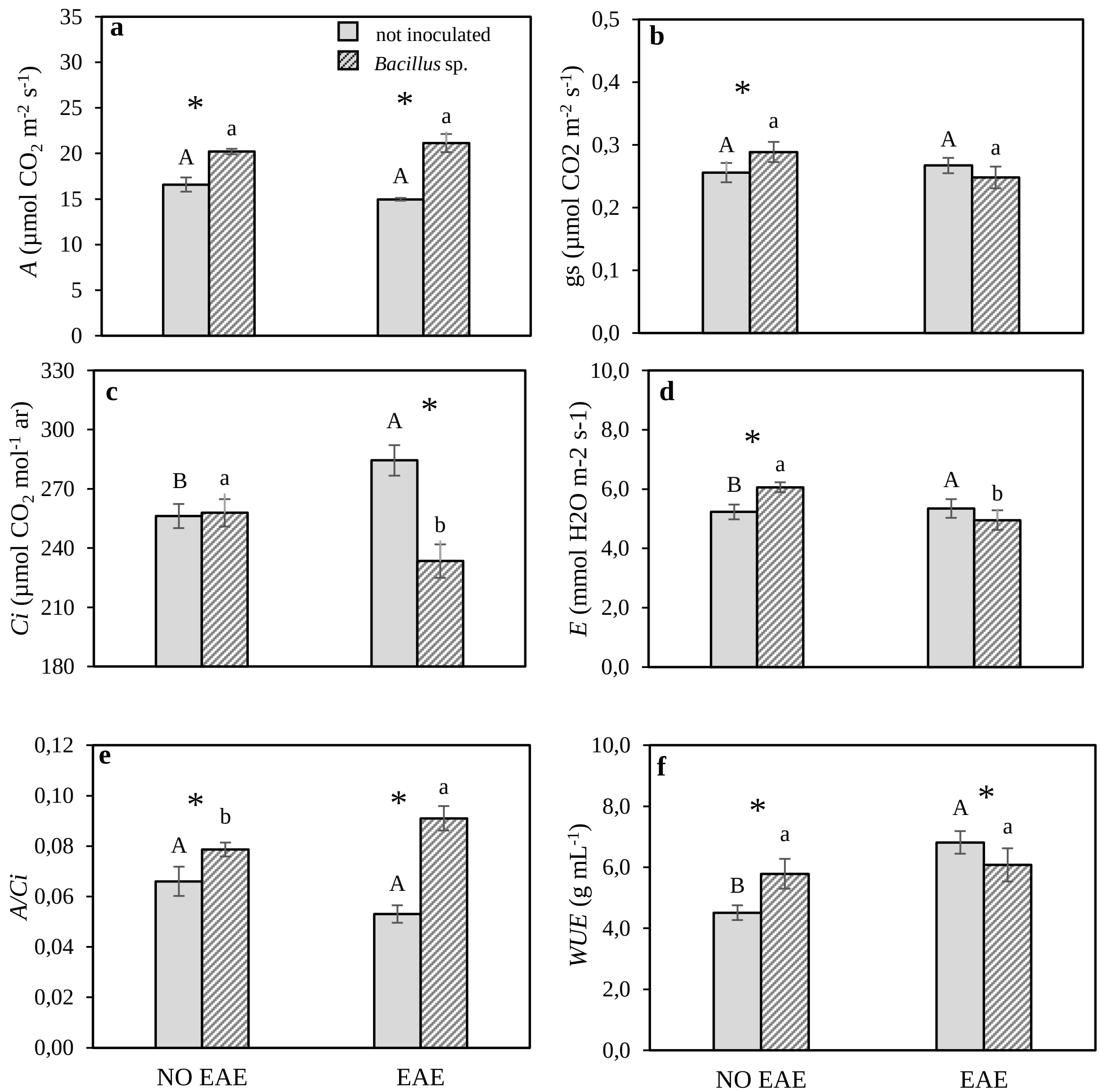


Fig 4 a: Net CO2 assimilation rate (A), b: stomatal conductance to water vapor (gs), c: intercellular CO2 concentration (Ci), d: transpiration (E), e: carboxylation efficiency (A/Ci) and f: water use efficiency (WUE) in *Corymbia* seedlings treated with *Bacillus* sp., EAE, *Bacillus* sp.+ EAE. Uppercase letters compare the effect of the rhizobacteria for each dose of EAE (electric arc furnace slag). Lowercase letters compare the effect of inoculation between doses of EAE. The (*) indicates a difference between treatments within the EAE dose. Averages with equal letters do not differ significantly according to Student's t-test ($P < 0.05$). NO EAE: without the addition of steel waste; EAE: with the addition of steel waste.

3.3.4 Soluble sugars and foliar starch contents

Inoculation had the greatest effect on soluble sugars and starch levels in plant leaves. Glucose, fructose, sucrose, and starch were influenced by *Bacillus* sp. and *Bacillus* sp. + EAE. For fructose and starch, there was also a significant effect of the EAE factor (Table 4).

Table 4. F-statistics for the effect of rhizobacterium inoculation (*Bacillus* sp.), fertilization with electric arc furnace steel slag (EAE) and the interaction between them (*Bacillus* sp. + EAE) on the soluble sugar and leaf starch content of seedlings of *Corymbia.*

Variables	*Bacillus* sp.	EAE	*Bacillus* sp. + EAE	C.V. (%)
Glucose	19.76*	0,09	14.1*	10
Fructose	7,4*	108,0*	4,18*	11
Sucrose	31,8*	0,67	6.84*	10
Starch	133*	19.9*	34.3*	4

* Significant ao $P \leq 0{,}05$.

In the absence of EAE, *Bacillus* sp. reduced sucrose content by 21% and increased starch by 19%, with no changes in glucose and fructose levels. The combination of *Bacillus* sp. + EAE showed a similar response, resulting in a 47% reduction in sucrose and an 18% increase in starch. However, in this comparison, *Bacillus* sp. + EAE reduced glucose by 31% and increased fructose by 41%, compared to the control without inoculation and without EAE.

Comparing the effects of EAE and *Bacillus* sp. + EAE, the steel slag promoted increases of 78% in glucose, 27% in fructose, 114% in sucrose, and reduced starch content by 5%. EAE induced significant increases in fructose (80%) and starch (11%) contents (Fig 5).

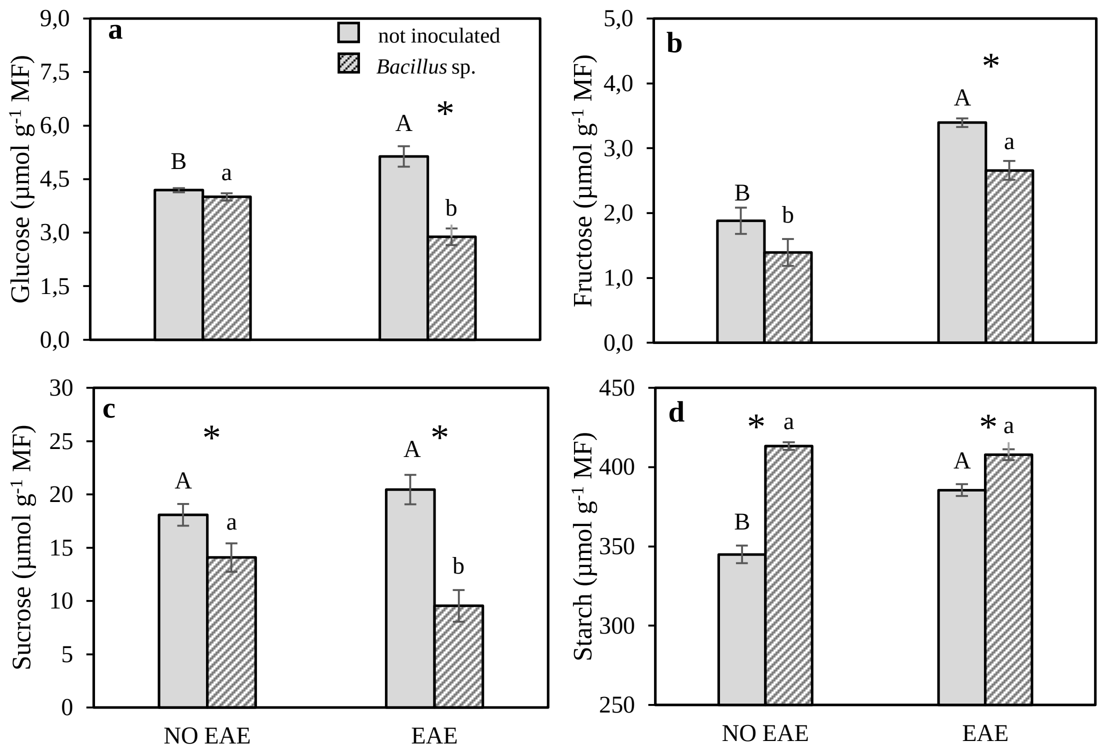


Fig. 5 Contents of a: glucose, b: fructose, c: sucrose and d: starch in *Corymbia* leaves treated with *Bacillus* sp., EAE, *Bacillus* sp.+ EAE. Uppercase letters compare the effect of the rhizobacteria for each dose of EAE (electric arc furnace slag). Lowercase letters compare the effect of inoculation between doses of EAE. The (*) indicates a difference between treatments within the EAE dose. Averages with equal letters do not differ significantly according to Student's t-test ($P < 0.05$). NO EAE: without the addition of steel waste; EAE: with the addition of steel waste.

3.3.5 Heavy metal contents and translocation

The levels and translocation of heavy metals were assessed in *Corymbia* roots under the influence of individual factors (EAE and *Bacillus* sp.) or in combination (*Bacillus* sp. + EAE). EAE had the greatest impact on Cd and Cr levels, while *Bacillus* sp. dominated in influencing Pb levels (Table 5). For foliar metal levels, only Pb showed effects from *Bacillus* sp. and *Bacillus* sp. + EAE. Cd levels were below detection limits in foliar tissues.

Table 5 F-statistics for the effect of rhizobacteria inoculation (*Bacillus* sp.), fertilization with electric arc furnace slag (EAE) and the interaction between them (*Bacillus* sp. + EAE) on the levels (mg kg^{-1}) of chromium (Cr), lead (Pb) and cadmium (Cd) in the roots and leaves of *Corymbia* seedlings.

Variáveis	*Bacillus* sp.	EAE	*Bacillus* sp. + EAE	C.V. (%)
Roots				
Cd	58.07*	416.66*	40.90*	13
Cr	0.30	19.69*	7.69*	10
Pb	8.14*	0.01	1.97	14
Leaves				
Cr	0.12	0.045	0.40	5
Pb	171.52*	0.23	8.47*	7

*Significant at $P \leq 0,05$.

In the absence of EAE, *Bacillus* sp. increased Pb levels in roots by 39% compared to the uninoculated control without EAE. The same trend was observed with *Bacillus* sp. + EAE, though it did not differ from the control. Comparing the effects of *Bacillus* sp. and *Bacillus* sp. + EAE on foliar Pb levels, there was a reduction of 49% and 37%, respectively, compared to the control without EAE and uninoculated. In the presence of EAE, the rhizobacteria resulted in a 32% reduction in foliar Pb compared to EAE applied alone in *Corymbia* seedlings (Fig 6a and 6d). With TF values < 1, there was no effective translocation of Pb from roots to aboveground parts in any evaluated treatments. However, Pb translocation from roots to leaves of *Corymbia* was altered by rhizobacteria inoculation, resulting in a 64% reduction in the absence of EAE (*Bacillus* sp.) and a 51% reduction in the presence of EAE (*Bacillus* sp. + EAE) compared to controls (Table 6).

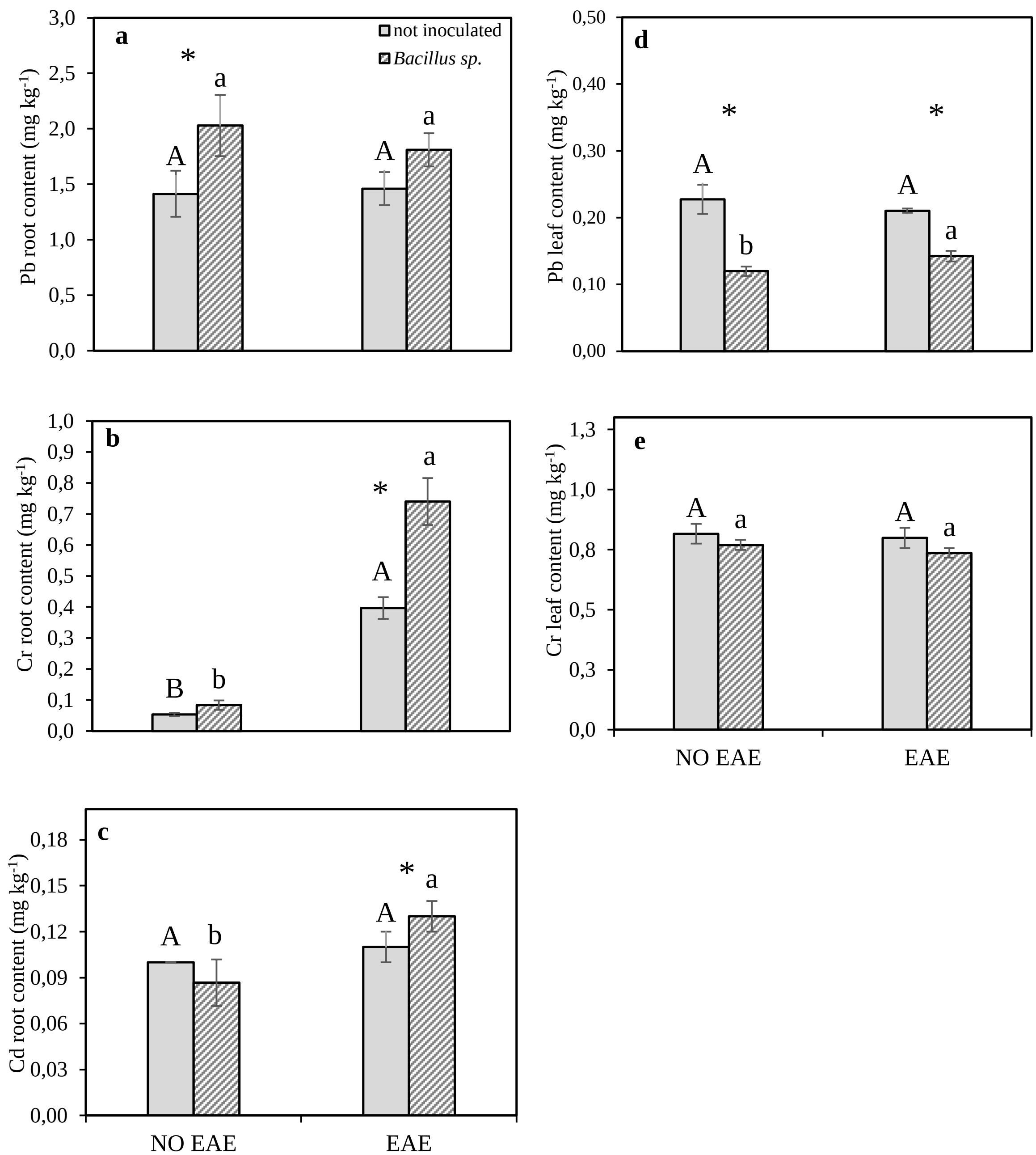


Figure. 6 Root contents of a: lead (Pb), b: chromium (Cr), c: cadmium (Cd). Foliar contents of d: lead (Pb), e: chromium (Cr) in *Corymbia* seedlings treated with *Bacillus* sp., EAE, *Bacillus* sp.+ EAE. Uppercase letters compare the effect of the rhizobacterium for each dose of EAE. Lowercase letters compare the effect of *Bacillus* sp. between the doses of EAE. The (*) indicates a difference between treatments within the EAE dose. Averages with equal letters do not differ significantly according to Student's t-test ($P < 0.05$). NO EAE: without the addition of steel waste; EAE: with the addition of steel waste.

Table 6. Effects of inoculating the rhizobacterium *Bacillus* sp. in the absence and presence of electric arc furnace slag (EAE) on the translocation factor (TF) of chromium (Cr) and lead (Pb) in *Corymbia* seedlings

	NO EAE		EAE		
TF	Not inoculated	*Bacillus* sp.	Not inoculated	*Bacillus* sp.	C.V. (%)
Cr	15.3 Aa	9.43 Ba	2.03 Ab	1.00 Ab	9
Pb	0.16 Aa	0.06 Ba	0.15 Aa	0.08 Ba	6

Note: Uppercase letters compare the effect of *Bacillus* sp. for each dose of EAE. Lowercase letters compare the effect of *Bacillus* sp between the doses of EAE. Averages with equal letters do not differ significantly according to the Sudent`s t-test ($P < 0.05$). NO EAE: without the addition of steel waste; EAE: with the addition of steel waste. c.v. coefficient of variation. TF values > 1 indicate that the element was translocated.

Isolated *Bacillus* sp. inoculation, in the absence of EAE, had no effect on Cr levels in plant root tissues, remaining below 0.1 mg kg^{-1}. EAE had the greatest impact on Cr levels in roots, promoting a 643% increase compared to control plants. The combination of *Bacillus* sp. + EAE increased Cr levels by 1287% compared to the uninoculated control without EAE and by 86% compared to the effect of EAE alone (Fig 6b). Foliar Cr levels did not differ among treatments, reaching an average of 0.8 mg kg^{-1} (Fig 6e). High TF values for Cr demonstrate effective translocation from roots to aboveground parts, with all treatments showing lower TF values compared to the control plants without rhizobacteria and EAE. *Bacillus* sp. inoculation reduced translocation by 40%, while the combination *Bacillus* sp. + EAE reduced TF by 93% compared to control plants without EAE and uninoculated. EAE alone significantly reduced Cr translocation by 86% compared to the control without EAE (Table 6).

In the absence of EAE, there was no effect of *Bacillus* sp. inoculation on Cd content in the roots of *Corymbia* seedlings. The combination of *Bacillus* sp. + EAE resulted in a 30% increase compared to the control without EAE and inoculation, and an 18% increase compared to plants with EAE alone (Fig 6c). Foliar Cd levels remained below the method's quantification limit (< 0.01 mg kg^{-1}).

3.3.6 Foliar Nutrient Content and Nutrient Use Efficiency

Bacillus sp., in the absence of EAE, led to an 8% increase in foliar K, 69% in Mn, and 19% in Zn. On the other hand, it reduced S by 15% and had no effect on the levels of other nutrients compared to non-inoculated control plants. In the presence of EAE, the combination of *Bacillus* sp. + EAE increased P (31%), K (18%), Cu (18%), Mn (13%), and Zn (20%), and reduced Ca (41%) and Fe (13%) compared to plants fertilized with EAE but not inoculated.

In the absence of EAE, *Bacillus* sp. optimized the nutrient use efficiency of P (29%), Ca (21%), Mg (21%), S (33%), B (41%), Cu (41%), and Zn (33%), and reduced Mn by 21%. No changes were observed in the NUE of N, K, Fe compared to non-inoculated control plants. EAE improved the use efficiency of all evaluated nutrients, although the increments observed for N (19%) and Fe (14%) did not significantly differ from the control plants without EAE and inoculation. Improvements were 81% for P, 53% for K, 36% for Mg, 24% for S, 51% for B, 83% for Cu, 55% for Mn, and 102% for Zn (Table 7).

The combination of *Bacillus* sp. + EAE led to improvements in NUE for P (26%), K (16%), S (34%), B (33%), Cu (36%), Mn (12%), and Zn (51%), while reducing the NUE for Fe by 44% compared to control plants without inoculation and EAE. However, when comparing the NUE of plants subjected to *Bacillus* sp. + EAE with plants treated only with the *Bacillus* sp. bacterium, no differences were observed in NUE for N, P, K, S, B, Cu, but there was an increase for Mn (42%) and Zn (13%), and a reduction in Ca (15%), Mg (18%), and Fe (50%) (Table 7).

Table 7. Effects of inoculation of the rhizobacteria *Bacillus* sp. in the absence and presence of EAE (electric arc furnace slag) on leaf contents and nutrient use efficiency in *Corymbia* seedlings.

Nutrient	Leaf contents				Nutrient use efficiency (g^2 mg^{-1})			
	NO EAE		EAE		NO EAE		EAE	
	not inoculated	*Bacillus* sp.	not inoculated	*Bacillus* sp.	not inoculated	*Bacillus* sp.	not inoculated	*Bacillus* sp.
N (g kg^{-1})	13.20Aa	12.49Aa	13.66Aa	12.74Aa	0.15Aa	0.18Aa	0.18Aa	0.18Aa
P (g kg^{-1})	2.08Aa	1.95Aa	1.50Bb	1.97Aa	1.07Bb	1.38Aa	1.94Aa	1.35Ab
K (g kg^{-1})	13.39Ba	14.56Aa	11.77Bb	13.87Aa	0.16Ab	0.18Aa	0.25Aa	0.19Ba
Ca (g kg^{-1})	9.27Ab	9.52Aa	18.07Aa	10.65Ba	0.19Ba	0.23Aa	0.14Bb	0.20Ab
Mg (g kg^{-1})	2.87Aa	2.84Aa	2.91Aa	2.76Aa	0.56Bb	0.68Aa	0.76Aa	0.55Bb
S (g kg^{-1})	1.80Aa	1.52Ba	1.49Ab	1.51Aa	1.06Bb	1.41Aa	1.31Aa	1.43Aa
B (mg kg^{-1})	56.24Aa	54.21Aa	55.12Aa	54.85Aa	0.24Bb	0.34Aa	0.36Aa	0.31Aa
Cu (mg kg^{-1})	5.26Aa	5.65Aa	4.28Bb	5.08Ab	1.73Bb	2.44Aa	3.18Aa	2.36Bb
Fe (mg kg^{-1})	79.51Ab	82.88Aa	97.90Aa	84.85Ba	0.013Aa	0.014Aa	0.015Aa	0.007Bb
Mn (mg kg^{-1})	299.73Ba	445.33Aa	264.81Ba	298.93Ab	0.084Ab	0.006Bb	0.130Aa	0.094Ba
Zn (mg kg^{-1})	41.46Ba	49.40Aa	36.42Bb	43.98Ab	0.33Bb	0.44Ab	0.67Aa	0.50Ba

Note: Uppercase letters compare the effect of inoculation on EAE dose. Lowercase letters compare the effect of inoculation between EAE doses. Means with the same letters do not differ significantly according to Student's t-test with Bonferroni correction ($p < 0.05$). NO EAE: without addition of steel residue; EAE: with addition of steel residue.

3.3.7 Biometry and Biomass

Growth parameters were proportionally influenced by *Bacillus* sp., EAE, and *Bacillus* sp. + EAE (Fig 7). Independently, the rhizobacterium had a significant effect on height, stem diameter, root length, and root volume. The variables height, number of leaves, leaf area, root length, and robustness index were influenced by the EAE factor. The combination *Bacillus* sp. + EAE was significant for height, stem diameter, leaf area, root volume, and robustness index (Table 8).

Table 8. F-statistics for the effect of rhizobacterium inoculation (*Bacillus* sp.), fertilization with electric arc furnace steel slag (EAE) and the interaction between them (*Bacillus* sp. + EAE) on growth variables in *Corymbia* seedlings.

Variables	*Bacillus* sp.	EAE	*Bacillus* sp. + EAE	C.V. (%)
Biometry				
H (cm)	18,1*	234,5*	60,3*	2
DC (mm)	12,6*	2,0	6,7*	4
NF	4,0	28,2*	0,1	4
AF (cm^2)	0,05	47,4*	39,7*	4
CR (cm)	49,5*	46,2*	2,5	5
VR (cm^3)	58,0*	1,3	14,5*	6
IR	3,95	238*	6,97*	6
Dry biomass				
Root (g)	382,2*	242*	119*	3
Stem (g)	0,12	5,8*	9,6*	12
Leaf (g)	5,8*	91,8*	34,2*	6
Total (g)	9,61*	222*	133*	4
Root/shoot ratio	75,2*	0,06	0,05	8

* Significant at $P \leq 0.05$. H: height; CD: collar diameter; NF: number of leaves; AF: leaf area; CR: root length; VR: root volume; IR: robustness index.

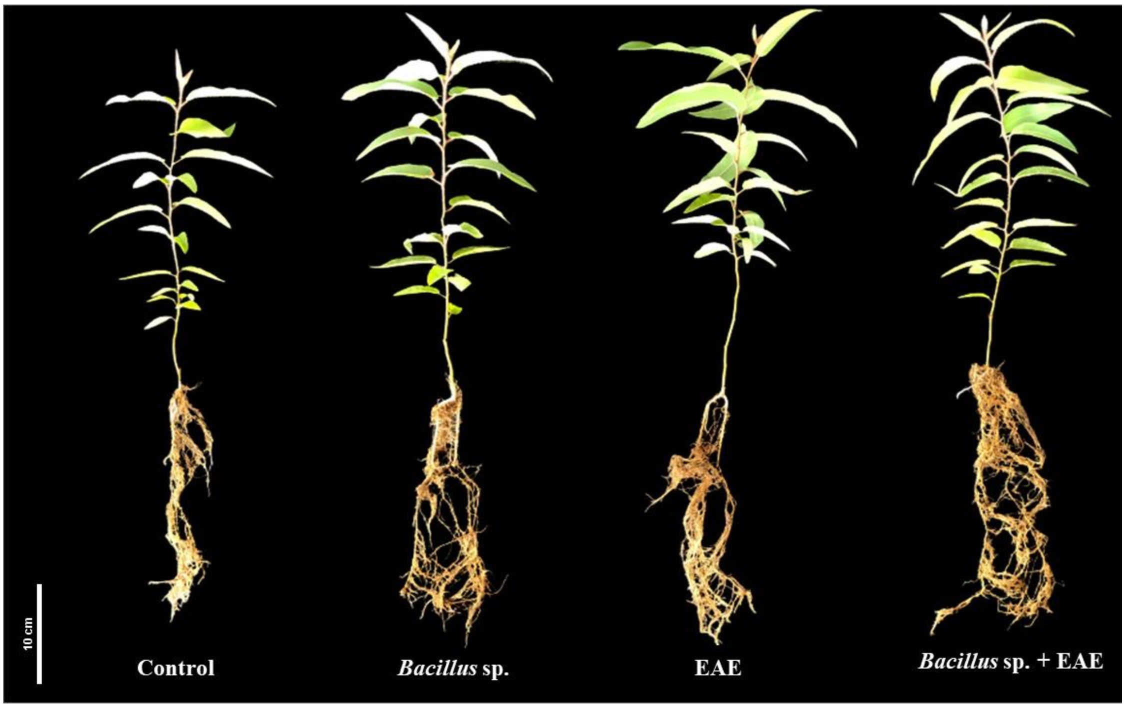


Fig 7 Growth of *C. torelliana* x *C. citriodora* seedlings inoculated with *Bacillus* sp, fertilized with electric arc furnace slag (EAE) and the combination of the two (*Bacillus* sp. + EAE) at the end of the experiment.

In the absence of EAE, *Bacillus* sp. promoted increases in height, stem diameter, leaf area, and root volume. When associated with EAE, the combination *Bacillus* sp. + EAE resulted in increases of 6% in height, 15% in root volume, and 13% in the robustness index but reduced stem diameter by 5% compared to the control without EAE. When comparing the effect of the combination *Bacillus* sp. + EAE with EAE alone, the combination led to a 48% increase in root volume and a reduction of 13% in leaf area and 1.5% in height, while increasing the robustness index by 2% (Table 9).

The accumulation of dry biomass in roots, leaves, and total biomass was influenced by the factors alone and the combination of *Bacillus* sp. + EAE. For stem biomass, there was a significant effect of both *Bacillus* sp. + EAE and EAE alone. For the root/shoot ratio, only the effect of *Bacillus* sp. was significant (Table 8).

Table 9. Effects of inoculating the rhizobacterium *Bacillus* sp. in the absence and presence of EAE on growth variables in *Corymbia* seedlings. The data shown refers to the variables with a significant interaction between the factors evaluated.

	NO EAE		EAE		
	Not inoculated	*Bacillus* sp.	Not inoculated	*Bacillus* sp.	C.V. (%)
Biometry					
H (cm)	31.7Bb	34.9Ab	37.7Aa	37.1Ba	2
DC (mm)	3.42Ba	3.74Aa	3.48Aa	3.53Ab	4
AF (cm^2)	239Bb	288Aa	339Aa	293 Ba	6
VR (cm^3)	10.5Ba	12.0Ab	9.4Ba	13.9Aa	7
IR	9.3Ab	9.8Ab	10.8Ba	11.1Aa	6
Dry biomass					
Root (g)	0.877Bb	1.369Ab	1.305Ba	1.444Aa	3
Stem (g)	0.543Ab	0.658Aa	0.774Aa	0.629Ba	12
Leaf (g)	1.471Bb	1.636Ab	2.214Aa	1.816Ba	6
Total (g)	2.840Bb	3.670Ab	4.320Aa	3.800Ba	4

Note: Uppercase letters compare the effect of Bacillus sp. for each dose of EAE. Lower case letters compare the effect of Bacillus sp. between EAE doses. Averages with equal letters do not differ significantly according to the t-test ($P < 0.05$). NO EAE: without the addition of steel waste; EAE: with the addition of steel waste; H: height; DC: collar diameter; AF: leaf area; VR: root volume; IR: robustness index.

In the absence of EAE, the rhizobacterium increased root dry biomass by 56%, leaf biomass by 11%, and total biomass by 29%. Similar response patterns were observed for *Bacillus* sp. + EAE, with gains of 3.5% in root mass, 11% in leaf mass, and 3.5% in total biomass (Table 9). When comparing the effects of *Bacillus* sp. + EAE and EAE, the combination promoted a 10% increase in root dry biomass; however, EAE yielded better results for stem, leaf, and total dry biomass.

No significant effects were observed on leaf number and root/shoot ratio (Table 8). Root length was reduced by 7% due to EAE, both in the presence and absence of *Bacillus* sp. (Table 10).

Table 10. Effects of inoculating the rhizobacterium *Bacillus* sp. in the absence and presence of EAE on growth variables in *Corymbia* seedlings. The data presented refers to the variables with no significant interaction between the factors evaluated.

	NO EAE		EAE		C.V. (%)
	Not inoculated	*Bacillus* sp.	Not inoculated	*Bacillus* sp.	
Biometry					
NF	20 A		21 A		4
CR (cm)	31.2 A		28,8 B		2
Dry biomass					
Root/shoot ratio	0.525 A		0.519 A		8

Note: Uppercase letters compare the effect of *Bacillus* sp. for each dose of EAE. Lowercase letters compare the effect of *Bacillus* sp between the doses of EAE. Averages with equal letters do not differ significantly according to the Student`s t-test ($P < 0.05$). NO EAE: without the addition of steel waste; EAE: with the addition of steel waste; NF: number of leaves; CR: root length.

3.4 Discussion

The results of this study support the hypothesis that combining the byproduct electric arc furnace slag (EAE) with the rhizobacterium *Bacillus* sp. positively enhances the photosynthetic, biochemical, and nutritional activity, leading to greater growth of seedlings. This represents the first record of such a strategy for optimizing EAE recycling in *Corymbia* seedling production.

To sustain the high rates of Rubisco carboxylation observed in this study, a greater supply of chemical energy in the form of ATP and NADPH from the photochemical stage of photosynthesis is necessary. The production of this reducing power occurs through improved capture, absorption, and transfer of light energy to photosystems I and II. The conversion of light energy into chemical energy can be assessed by chlorophyll a fluorescence parameters such as *Fv/Fo, ETR, qP, qN, NPQ*, among others (CAMPOSTRINI, 2001), considered sensitive indicators of growth promotion, especially in association with growth-promoting microorganisms (KANAGENDRAN et al., 2019).

The present study demonstrated that the application of *Bacillus* sp. + EAE on *Corymbia* seedlings resulted in higher potential activity of photosystem II (*Fv/Fo*) and a greater electron transfer rate (*ETR*). There was an increase in energy dissipation in photochemical reactions (*qP*) and a decrease in non-photochemical dissipation (*qN* and *NPQ*), indicating greater efficiency in using light energy. These results demonstrate the integrity of photosystems I and II, suggesting that the combined use of rhizobacteria and

EAE improves the capture and transfer of energy, ensuring the supply of ATP and NADPH needed in the biochemical stage of photosynthesis (FISHER; KRAMER, 2021).

Similar improvements in the functioning of PSII were also observed in bio-stimulated seedlings of açaí (DE CASTRO et al., 2020) and pepper (SAMANIEGO-GÁMEZ et al., 2016). The enhanced energy capture and transfer observed in this study reflected in higher photosynthetic performance, which is closely related to chlorophyll content, especially chlorophyll *a*—the primary pigment responsible for capturing, absorbing, and transferring light energy. Its synthesis depends on an adequate nutritional state, with a focus on the supply of Mg^{2+} and Fe^{2+} (TANAKA; TANAKA, 2006).

The inoculation of rhizobacteria can lead to increases in Chl a+b, Chl a, Chl b, and photosynthetic rate (TUFAIL et al., 2021) through the activation of the biosynthetic pathway of hormones such as gibberellin and cytokinins, amino acids, and chlorophyll precursors (CHAUHAN et al., 2015). *Bacillus* species can fix atmospheric nitrogen, an essential element and a crucial component of chlorophyll structure and amino acid production (ELSAYED et al., 2022). However, the best pigment content results were positively associated with EAE, possibly due to the iron levels in the *Bacillus* sp. + EAE treatment, considering the overall nutritional state acquired by the plants.

The highest rates of net CO_2 assimilation (*A*) were observed in plants subjected to the *Bacillus* sp. rhizobacteria, particularly in interaction with EAE (*Bacillus* sp. + EAE). The lower intercellular concentration of CO_2 (*Ci*) and the higher carboxylation efficiency (*A/Ci*) obtained with this interaction suggest greater utilization of assimilated CO_2 by Rubisco enzyme activity, responsible for carbon fixation via the Calvin-Benson cycle, as reported in cucumber plants inoculated with the growth promoter *Bacillus velezensis* (WANG et al., 2022).

Stomatal conductance (*gs*) was slightly modified by the inoculation of *Bacillus* sp. in the absence of EAE, indicating no high resistance to CO_2 entry, typically imposed in situations with smaller stomatal openings. The *gs* behavior reflects transpiration rate (*E*), as higher stomatal opening generally results in greater transpiration. *Bacillus* sp. + EAE reduced transpiration and improved water use efficiency (*WUE*), suggesting that plants subjected to this treatment established a balance between stomatal opening and water loss via transpiration, fixing more carbon with less water elimination.

The fixed CO_2 can be allocated for the biosynthesis of organic compounds, such as carbohydrates, necessary to sustain high rates of respiration in rapidly growing plants (TAIZ et al., 2017). Generally, triose-phosphate molecules synthesized in the chloroplast are exported to the cytosol, where they undergo enzymatic interchange into glucose and fructose, prioritizing the supply of ATP and NADPH through cellular respiration. The continuous production of triose-phosphate leads to the accumulation of sucrose and starch (SMIRNOVA et al., 2015). The production, accumulation, and consumption of these compounds depend on the plant's energy requirements, with high metabolic rates typically resulting in reduced cytosolic glucose levels (NELSON and COX, 2022). Plants subjected to *Bacillus* sp. + EAE showed reduced glucose and sucrose levels, suggesting increased glycolytic oxidation for ATP and NADPH production. Interestingly, fructose exhibited slightly higher values compared to the isolated effect of *Bacillus* sp., indicating it may act as a glycolytic intermediate, potentially feeding another glycolysis pathway. Additionally, sucrose production contributes to the reduction of cytoplasmic glucose and fructose.

Accelerated growth leads to increased export of sucrose to sink organs such as roots and meristems, along with greater starch storage for periods without light. Increases in *A* and *A/Ci* may be related to the enhanced allocation of photoassimilates for starch biosynthesis in plants treated with the rhizobacterium, both in the presence (*Bacillus* sp. + EAE) and absence of residue (*Bacillus* sp.). Thus, bio-stimulation may have accelerated the growth rate of *Corymbia* seedlings, increasing energy demand, resulting in greater sucrose export and starch storage, consequently reducing glucose levels, although fructose levels were elevated in these plants. The increase in soluble sugars induced by rhizobacteria is recognized (ABDELAAL et al., 2021), including the *Bacillus genus* (WANG et al., 2022). The reduction in sucrose levels suggests that the rhizobacterium induced its consumption, given its ease of translocation within the plant. This reduction aligns with the observed growth and biomass accumulation gains, particularly in the root system, in plants inoculated with *Bacillus* sp.

The slag used in the study had heavy metals such as Cr and Pb in its composition (Table 2). According to Brazilian legislation defining acceptable levels of heavy metals in mineral fertilizers and soil amendments (BRASIL, 2006), the Cr content in the slag used was within the permitted range, but the Pb content was about ten times above the limit. However, no phytotoxic symptoms caused by the investigated metals were observed, such as a reduction in stomatal opening and photosynthetic rate, water transport and utilization, chlorophyll content, and dry biomass (LI et al., 2023). Despite heavy metals in slag, many studies have shown they can be used without soil contamination and plant phytotoxicity risks (DEUS et al., 2020; GAO et al., 2023; HUAIWEI; XIN, 2011; WANG et al., 2018a), as also evidenced in this study.

Slag is a calcium silicate that acts as a soil acidity corrector, releasing OH- ions that can form hydroxide precipitates with heavy metal ions, reducing their availability in the soil (GAO et al., 2023; YANG et al., 2021). Additionally, slags have adsorption surfaces that can immobilize heavy metals in contaminated soil (O'CONNOR et al., 2021).

The *Bacillus* sp. rhizobacterium reduced the translocation of Pb and Cr in *Corymbia* plants, indicating its potential use in situations with heavy metal contamination in the soil. PGPR can directly assist plants in the presence of heavy metals by increasing the mobilization of these elements, thus alleviating metal toxicity levels in plants (ANDRADES-MORENO et al., 2014), or indirectly by providing nutrients through nitrogen fixation, phosphorus solubilization, and increased production of iron transporters, regulating plant growth hormones, promoting plant growth, and stress tolerance (IKRAM et al., 2018; ZHU et al., 2023). These effects are related to various microbial mechanisms, including the production of phytohormones, siderophores (chelators), biofilms, and phosphate solubilization (JAN et al., 2019; MASLENNIKOVA et al., 2022; WANG et al., 2018b), characteristics present in the *Bacillus* isolate used in this study.

The application of EAE and inoculation with *Bacillus* sp. increased nutrient use efficiency (NUE) for most evaluated nutrients, especially P, Mg, S, B, Cu, and Zn. This result may be related to the growth-promoting capability established by the rhizobacteria in the plant root system, enabling better soil exploration and increased absorption of water and nutrients provided by EAE. Additionally, EAE contains nutrients in its composition, likely raising their concentration in the soil solution, contributing to higher NUE. Furthermore, EAE promoted greater Ca absorption, a crucial nutrient for root system development. Increased root system is a significant mechanism in enhancing plant NUE, as it boosts nutrient absorption rates. Several studies report enhanced nutrient availability and plant biomass production with slag use (DAS et al., 2020; TOZSİN; ÖZTAŞ, 2023; WANG et al., 2018a).

The *Bacillus* sp. strain used in this research is proficient in biofilm production, associated with improved rhizospheric hydration, root system protection under water or chemical stress (VELMOUROUGANE; PRASANNA; SAXENA, 2017), enhancing soil fertility, and promoting plant growth (ADAK et al., 2016; VELMOUROUGANE et al., 2022). This contributes to establishing microbe-root interactions and nutrient uptake.

The combined physiological, biochemical, and nutritional modifications highlight the effectiveness of the *Bacillus* sp. + EAE combination in promoting plant growth, resulting in increased total dry biomass accumulation. The steel slag brought improvements to plant biomass individually or in combination with *Bacillus* sp., showing significant gains in plant height, leaf area, root volume, and nutrient use efficiency.

The observed increases in roots allow for an expanded soil exploration area, enhancing nutrient absorption and resulting in more robust and productive plants (AHKAMI et al., 2017). Root structure and dimension improvements induced by rhizobacteria are acknowledged, demonstrating their potential to modify the root system post-inoculation (LIMA et al., 2021).

These root-level modifications may be related to higher levels of indoleacetic acid (IAA) produced by the rhizobacteria (EGAMBERDIEVA et al., 2017; PERALTA et al., 2012; SILVA et al., 2022). Inoculation with *Bacillus* species producing IAA promotes root gains in eucalyptus clones. Angulo et al. (2014) achieved up to 45% increases in root diameter and 135% in root biomass. González-Díaz et al. (2019) observed enhanced rooting and root biomass in inoculated eucalyptus cuttings. The hormonal changes triggered by the bio-stimulant are closely related to increased root volume, as auxins induce the production of lateral roots and root hairs.

The integrated and balanced growth between roots and aboveground parts depends on the hormonal balance between IAA and cytokinins. These two hormones operate in a positive feedback model, where cytokinins produced in the root apex signal the proliferation of shoot buds, inducing aboveground development, the site of IAA production, which in turn acts on the root system (STROTMANN; STAHL, 2021; ZHANG et al., 2013). However, it is essential to consider both the quantity of IAA produced and available for plant use and the plant tissues' sensitivity to changes in this hormone's concentration, as factors modulating final plant responses (LEVEAU; LINDOW, 2005).

Several benefits to the plant can be listed due to the root modifications promoted by bio-stimulation, such as improved plant anchorage, a larger explored soil area, leading to an expectation of increased water and mineral nutrient absorption (VALENTE LIMA et al., 2020). These gains enhance seedling robustness, contributing to better adaptation to field conditions.

Our results demonstrate that the application of *Bacillus* sp. improved photosynthetic performance, increased growth, and NUE of *Corymbia* while reducing the translocation of heavy metals from roots to aerial parts, as described in other species such as rice, tomato, wheat, spinach, and corn (LIU et al., 2022; SARWAR et al., 2023; HASSAN et al., 2016; THATOI et al., 2014; WANG et al., 2022b).

The advantages shown so far suggest that the application of EAE associated with *Bacillus* sp. bio-stimulation for *Corymbia* nutrition may represent an excellent strategy to accelerate seedling growth, reduce production costs, and prevent environmental damage. The potential for technological development in the biofertilizer field, with cleaner and more sustainable products and processes, will help reduce the environmental impact caused by steel mill residues in plant yards.

3.5 Conclusions

Bio-stimulation of eucalyptus seedlings with the *Bacillus* sp. isolate promoted growth gains in hybrid *Corymbia* clones, and these improvements were amplified in the presence of the EAE steel mill residue. The combination of *Bacillus* sp. + EAE promotes growth through improvements in physiological, biochemical, and nutritional processes.

The better photosynthetic performance in plants subjected to this combination is related to higher efficiency in photochemical processes, as observed in chlorophyll a fluorescence variable. Changes in glucose, fructose, sucrose, and starch levels indicate increased respiration rates related to the energy conversion needed for the enhanced growth promoted by the bacterium alone and combined with EAE. The accumulation of heavy metals in roots and the reduction of translocation to aerial tissues demonstrated that *Bacillus* sp., alone or combined with EAE, has a protective effect against Pb, Cr, and Cd toxicity in *Corymbia* seedlings.

The higher nutrient use efficiency induced by the *Bacillus* sp. + EAE combination may have contributed to modulating physiological and biochemical processes related to the increased growth of *Corymbia* hybrids.

REFERENCES

ABIRI, R. *et al.* The Prospect of Physiological Events Associated with the Micropropagation of Eucalyptus sp. **Forests**, *[S. l.]*, v. 11, n. 11, p. 1211, 2020. Disponível em: https://doi.org/10.3390/f11111211

AFZAL, M. J. *et al.* Combined application of Bacillus sp. MN-54 and phosphorus improved growth and reduced lead uptake by maize in the lead-contaminated soil. **Environmental Science and Pollution Research**, *[S. l.]*, v. 27, n. 35, p. 44528–44539, 2020. Disponível em: https://doi.org/10.1007/S11356-020-10372-4/FIGURES/7. Acesso em: 10 dez. 2022.

AGARWAL, P. *et al.* PGPR-induced OsASR6 improves plant growth and yield by altering root auxin sensitivity and the xylem structure in transgenic Arabidopsis thaliana. **Journal of Plant Physiology**, *[S. l.]*, v. 240, p. 153010, 2019. Disponível em: https://doi.org/10.1016/j.jplph.2019.153010

AHEMAD, M.; KIBRET, M. Mechanisms and applications of plant growth promoting rhizobacteria: Current perspective. **Journal of King Saud University - Science**, *[S. l.]*, v. 26, n. 1, p. 1–20, 2014. Disponível em: https://doi.org/10.1016/j.jksus.2013.05.001

AHKAMI, A. H. *et al.* **Rhizosphere engineering: Enhancing sustainable plant ecosystem productivity**. *[S. l.]*: Elsevier B.V., 2017. Disponível em: https://doi.org/10.1016/j.rhisph.2017.04.012

ALEKSANDROV, VG; BLAGODYR, RN; ILEV, I. Liberation of phosphoric acid from apatite by silicate bacteria. **Mikrobiol Z**, *[S. l.]*, 1967.

ALI, M. A.; OH, J. H.; KIM, P. J. Evaluation of silicate iron slag amendment on reducing methane emission from flood water rice farming. **Agriculture, Ecosystems & Environment**, *[S. l.]*, v. 128, n. 1–2, p. 21–26, 2008. Disponível em: https://doi.org/10.1016/j.agee.2008.04.014

ALMEIDA, R. A. M. de *et al.* Slag Foaming Fundamentals - A Critical Assessment. **Materials Research**, *[S. l.]*, v. 20, n. 2, p. 474–480, 2017. Disponível em: https://doi.org/10.1590/1980-5373-mr-2016-0059

ANGULO, V. C. *et al.* Caracterización de rizobacterias promotoras de crecimiento en plántulas de Eucalyptus nitens. **Revista Argentina de Microbiología**, *[S. l.]*, v. 46, n. 4, p. 338–347, 2014 a. Disponível em: https://doi.org/10.1016/S0325-7541(14)70093-8

ANGULO, V. C. *et al.* Caracterización de rizobacterias promotoras de crecimiento en plántulas de Eucalyptus nitens. **Revista Argentina de Microbiología**, *[S. l.]*, v. 46, n. 4, p. 338–347, 2014 b. Disponível em: https://doi.org/10.1016/S0325-7541(14)70093-8

ASSIS, T. F. de. Melhoramento genético de Eucalyptus: desafios e perspectivas. **Anais do SIMPÓSIO BRASILEIRO DE SILVICULTURA**, *[S. l.]*, v. 1, n. 1, p. 127–148, 2014.

ASSIS, T. F. de; ABAD, J. I. M.; AGUIAR, A. M. Melhoramento Genético do

Eucalipto. *In*: MAURO VALDIR SCHUMACHER E MÁRCIO VIERA (ORG.) (org.). **Silvicultura do Eucalipto no Brasil**. 1. ed. Santa Maria: Editora UFSM, 2015. p. 225–247. *E-book.*

AUSTRALASIAN (IRON AND STEEL) SLAG ASSOCIATION INC. **Membership Annual Survey Results, 2019 Slag Sales and Production Survey, Australia.** *[S. l.: s. n.]* Disponível em: https://www.asa-inc.org.au/files/download/d787f72a84b5512

AVELAR, M. L. M. *et al.* In vitro ESTABLISHMENT OF Eucalyptus AND Corymbia SPECIES FROM EPICORMIC SHOOTS. **Revista Árvore**, *[S. l.]*, v. 44, 2020. Disponível em: https://doi.org/10.1590/1806-908820200000027

BELLINI, C.; PACURAR, D. I.; PERRONE, I. Adventitious Roots and Lateral Roots: Similarities and Differences. **Annual Review of Plant Biology**, *[S. l.]*, v. 65, n. 1, p. 639–666, 2014. Disponível em: https://doi.org/10.1146/annurev-arplant-050213-035645

BERBEL, A. *et al.* Changes in root morphological traits in soybean co-inoculated with Bradyrhizobium spp. and Azospirillum brasilense or treated with A. brasilense exudates. **Biology and Fertility of Soils**, *[S. l.]*, v. 56, p. 537–549, 2020. Disponível em: https://doi.org/10.1007/s00374-020-01453-0. Acesso em: 19 fev. 2023.

BRANCA, T. A. *et al.* Reuse and Recycling of By-Products in the Steel Sector: Recent Achievements Paving the Way to Circular Economy and Industrial Symbiosis in Europe. **Metals**, *[S. l.]*, v. 10, n. 3, p. 345, 2020 a. Disponível em: https://doi.org/10.3390/met10030345

BRANCA, T. A. *et al.* Reuse and Recycling of By-Products in the Steel Sector: Recent Achievements Paving the Way to Circular Economy and Industrial Symbiosis in Europe. **Metals**, *[S. l.]*, v. 10, n. 3, p. 345, 2020 b. Disponível em: https://doi.org/10.3390/met10030345

BRUNO MARCOS NUNES COSMO *et al.* Influência dos metais na fotossíntese. **Revista Agronomia Brasileira**, *[S. l.]*, v. 7, 2023. Disponível em: https://doi.org/10.29372/rab202347

CAI, S. *et al.* Fertilizer Efficiency and Risk Assessment of the Utilization of AOD Slag as a Mineral Fertilizer for Alfalfa (Medicago sativa L.) and Perennial Ryegrass (Lolium perenne L.) Planting. **Sustainability 2022, Vol. 14, Page 1575**, *[S. l.]*, v. 14, n. 3, p. 1575, 2022 a. Disponível em: https://doi.org/10.3390/SU14031575. Acesso em: 14 nov. 2022.

CAI, S. *et al.* Biochemical Analysis and Toxicity Assessment of Utilization of Argon Oxygen Decarbonization Slag as a Mineral Fertilizer for Tall Fescue (Festuca arundinacea Schreb) Planting. **Sustainability**, *[S. l.]*, v. 14, n. 15, p. 9286, 2022 b. Disponível em: https://doi.org/10.3390/su14159286

CALDWELL, M. M.; RICHARDS, J. H. Hydraulic lift: water efflux from upper roots improves effectiveness of water uptake by deep roots. **Oecologia**, *[S. l.]*, n. 79, p. 1–5, 1989.

CALVO, P. *et al.* Plant growth-promoting rhizobacteria induce changes in Arabidopsis thaliana gene expression of nitrate and ammonium uptake genes. **Journal of Plant Interactions**, *[S. l.]*, v. 14, n. 1, p. 224–231, 2019. Disponível em: https://doi.org/10.1080/17429145.2019.1602887

CALVO, P.; NELSON, L.; KLOEPPER, J. W. Agricultural uses of plant biostimulants. **Plant and Soil**, *[S. l.]*, n. 383, p. 3+41, 2014. Disponível em: https://doi.org/10.1007/s11104-014-2131-8

CARDOSO, P. H. S. *et al.* Fertilization management with sewage sludge sustains Eucalyptus productivity in Cerrado infertile soil. **Forest Ecology and Management**, *[S. l.]*, v. 519, p. 120338, 2022. Disponível em: https://doi.org/10.1016/j.foreco.2022.120338

CASTRO, G. S. A.; CRUSCIOL, C. A. C. Effects of superficial liming and silicate application on soil fertility and crop yield under rotation. **Geoderma**, *[S. l.]*, v. 195–196, p. 234–242, 2013. Disponível em: https://doi.org/10.1016/J.GEODERMA.2012.12.006

CHAND, S.; PAUL, B.; KUMAR, M. An Overview of Use of Linz-Donawitz (LD) Steel Slag in Agriculture. **Current World Environment**, *[S. l.]*, v. 10, n. 3, p. 975–984, 2015. Disponível em: https://doi.org/10.12944/CWE.10.3.29

CHAVES, M. M. How Plants Cope with Water Stress in the Field? Photosynthesis and Growth. **Annals of Botany**, *[S. l.]*, v. 89, n. 7, p. 907–916, 2002. Disponível em: https://doi.org/10.1093/aob/mcf105

CHEN, G. *et al.* Competitive mechanism and influencing factors for the simultaneous removal of Cr(III) and Zn(II) in acidic aqueous solutions using steel slag: Batch and column experiments. **Journal of Cleaner Production**, *[S. l.]*, v. 230, p. 69–79, 2019. Disponível em: https://doi.org/10.1016/j.jclepro.2019.04.402

CHIEN-YING YANG, INKA REIJONEN, HAI YU, RAJARATHNAM DHARMARAJAN, BALAJI SESHADRI, N. S. B. Back to Basic Slags as a Phosphorus Source and Liming Material. *In*: **Soil Amendments for Sustainability**. 1. ed. *[S. l.]*: CRC Press, 2018. p. 14. *E-book*.

CHOUDHARY, A. *et al.* Role of plant growth-promoting Rhizobacterium in adventitious root formation. *In*: **Environmental, Physiological and Chemical Controls of Adventitious Rooting in Cuttings**. *[S. l.]*: Elsevier, 2022. p. 159–181. *E-book.* Disponível em: https://doi.org/10.1016/B978-0-323-90636-4.00007-6

CORRÊA, J. C. *et al.* Disponibilidade de metais pesados em Latossolo com aplicação superficial de escória, lama cal, lodos de esgoto e calcário. **Pesquisa Agropecuaria Brasileira**, *[S. l.]*, v. 43, n. 3, p. 411–419, 2008. Disponível em: https://doi.org/10.1590/S0100-204X2008000300017. Acesso em: 28 abr. 2021.

CUNHA, T. Q. G. da *et al.* Eucalyptus expansion in Brazil: Energy yield in new forest frontiers. **Biomass and Bioenergy**, *[S. l.]*, v. 144, p. 105900, 2021. Disponível em: https://doi.org/10.1016/j.biombioe.2020.105900

DAS, S. *et al.* Taxonomic and functional responses of soil microbial communities to slag-based fertilizer amendment in rice cropping systems. **Environment International**, *[S. l.]*, v. 127, p. 531–539, 2019. Disponível em: https://doi.org/10.1016/J.ENVINT.2019.04.012

DAS, S. *et al.* Recycling of ferrous slag in agriculture: Potentials and challenges. **Critical Reviews in Environmental Science and Technology**, *[S. l.]*, p. 1–35, 2020. Disponível em: https://doi.org/10.1080/10643389.2020.1853458. Acesso em: 20 abr. 2021.

DE ARAUJO, M. J. *et al.* Initial productivity and genetic parameters of three corymbia species in brazil: Designing a breeding strategy. **Canadian Journal of Forest Research**, *[S. l.]*, v. 51, n. 1, p. 25–30, 2021. Disponível em: https://doi.org/10.1139/CJFR-2019-0438/ASSET/IMAGES/CJFR-2019-0438IEQ2.GIF. Acesso em: 25 abr. 2023.

DE CASTRO, G. L. S. *et al.* Açaí palm seedling growth promotion by rhizobacteria inoculation. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 51, n. 1, p. 205–216, 2020. Disponível em: https://doi.org/10.1007/s42770-019-00159-2

DEUS, A. C. F. *et al.* Effects of lime and steel slag application on soil fertility and soybean yield under a no till-system. **Soil and Tillage Research**, *[S. l.]*, v. 196, 2020. Disponível em: https://doi.org/10.1016/j.still.2019.104422

DHALI, S. *et al.* Synergistic effect of Bacillus and Rhizobium on cytological and photosynthetic performance of Macrotyloma uniflorum (Lam.) Verdc. Grown in Cr (VI) contaminated soil. **Plant Physiology and Biochemistry**, *[S. l.]*, v. 190, p. 62–69, 2022. Disponível em: https://doi.org/10.1016/J.PLAPHY.2022.08.027

DÍAZ-PILONETA, M. *et al.* Application of Steel Slag for Degraded Land Remediation. **Land**, *[S. l.]*, v. 11, n. 2, p. 224, 2022. Disponível em: https://doi.org/10.3390/LAND11020224. Acesso em: 15 fev. 2022.

DOTANIYA, M. L.; MEENA, V. D. Rhizosphere Effect on Nutrient Availability in Soil and Its Uptake by Plants: A Review. **Proc. Natl. Acad. Sci., India, Sect. B Biol. Sci.**, *[S. l.]*, n. 1, p. 1–15, 2015. Disponível em: https://doi.org/10.1007/s40011-013-0297-0

DUCA, D. *et al.* Indole-3-acetic acid in plant–microbe interactions. **Antonie van Leeuwenhoek**, *[S. l.]*, v. 106, n. 1, p. 85–125, 2014. Disponível em: https://doi.org/10.1007/s10482-013-0095-y

EGAMBERDIEVA, D. *et al.* Phytohormones and beneficial microbes: Essential components for plants to balance stress and fitness. **Frontiers in Microbiology**, *[S. l.]*, v. 8, n. OCT, p. 2104, 2017. Disponível em: https://doi.org/10.3389/FMICB.2017.02104/BIBTEX

EL-MEIHY, R. M. *et al.* Efficiency of heavy metals-tolerant plant growth promoting bacteria for alleviating heavy metals toxicity on sorghum. **Environmental and Experimental Botany**, *[S. l.]*, v. 162, 2019. Disponível em: https://doi.org/10.1016/j.envexpbot.2019.03.005

ELSAYED, A. *et al.* Synergistic effects of Azospirillum brasilense and Bacillus cereus on plant growth, biochemical attributes and molecular genetic regulation of steviol glycosides biosynthetic genes in Stevia rebaudiana. **Plant Physiology and Biochemistry**, *[S. l.]*, v. 189, p. 24–34, 2022. Disponível em: https://doi.org/10.1016/J.PLAPHY.2022.08.016

ENEBAK, S. A.; WEI, G.; KLOEPPER, J. W. Effects of Plant Growth-Promoting Rhizobacteria on Loblolly and Slash Pine Seedlings. **Forest Science**, *[S. l.]*, v. 44, n. 1, p. 139–144, 1998. Disponível em: https://doi.org/https://doi.org/10.1093/forestscience/44.1.139. Acesso em: 20 fev. 2023.

ETESAMI, H. Plant–microbe interactions in plants and stress tolerance. **Plant Life Under Changing Environment**, *[S. l.]*, p. 355–396, 2020. Disponível em: https://doi.org/10.1016/B978-0-12-818204-8.00018-7

EUROPEAN COMMISSION, D.-G. for R. and I.; KUHN, M., SPIEGEL, H., LOPEZ, A. F., REX, M., ERDMANN, R. **Sustainable agriculture using blast furnace and steel slags as liming agents**. Luxembourg: Publications Office, 2006.

FILIPPI, M. C. C. *et al.* Leaf blast (Magnaporthe oryzae) suppression and growth promotion by rhizobacteria on aerobic rice in Brazil. **Biological Control**, *[S. l.]*, v. 58, n. 2, p. 160–166, 2011. Disponível em: https://doi.org/10.1016/J.BIOCONTROL.2011.04.016

GE, Y. *et al.* Biofilm-overproducing Bacillus subtilis B12ΔYwcc decreases Cd uptake in Chinese cabbage through increasing Cd-immobilizing related gene abundance and root surface colonization. **Journal of Environmental Sciences**, *[S. l.]*, v. 120, p. 84–93, 2022. Disponível em: https://doi.org/10.1016/J.JES.2022.01.016

GONÇALVES MAFIA, R. *et al.* **Root colonization and interaction among growth … R. Árvore, Viçosa-MG, v.33, n.1**. *[S. l.: s. n.]*.

GONZÁLEZ, P. *et al.* Rhizobacteria strains as promoters of rooting in hybrids of Eucalyptus nitens × Eucalyptus globulus. **Chilean journal of agricultural research**, *[S. l.]*, v. 78, n. 1, p. 3–12, 2018. Disponível em: https://doi.org/10.4067/S0718-58392018000100003

GORDON, S. A.; WEBER, R. P. Colorimetric Estimation of indoleacetic Acid. **Plant Physiology**, *[S. l.]*, v. 26, n. 1, p. 192–195, 1951. Disponível em: https://doi.org/10.1104/PP.26.1.192. Acesso em: 19 abr. 2022.

GOSWAMI, M.; DEKA, S. Isolation of a novel rhizobacteria having multiple plant growth promoting traits and antifungal activity against certain phytopathogens. **Microbiological Research**, *[S. l.]*, v. 240, p. 126516, 2020. Disponível em: https://doi.org/10.1016/j.micres.2020.126516

GROBELAK, A. *et al.* Bacterial ACC deaminase activity in promoting plant growth on areas contaminated with heavy metals. **Journal of Ecological Engineering**, *[S. l.]*, v. 19, n. 5, p. 150–157, 2018. Disponível em: https://doi.org/10.12911/22998993/89818

GUERRINI, I. A. *et al.* Composted sewage sludge and steel mill slag as potential amendments for urban soils involved in afforestation programs. **Urban Forestry & Urban Greening**, *[S. l.]*, v. 22, p. 93–104, 2017. Disponível em: https://doi.org/10.1016/J.UFUG.2017.01.015

GUO, J.; BAO, Y.; WANG, M. **Steel slag in China: Treatment, recycling, and management**. *[S. l.]*: Elsevier Ltd, 2018. Disponível em: https://doi.org/10.1016/j.wasman.2018.04.045

GWON, H. S. *et al.* Environmental risk assessment of steel-making slags and the potential use of LD slag in mitigating methane emissions and the grain arsenic level in rice (Oryza sativa L.). **Journal of Hazardous Materials**, *[S. l.]*, v. 353, 2018. Disponível em: https://doi.org/10.1016/j.jhazmat.2018.04.023

HALING, R. E. *et al.* Root hairs improve root penetration, root–soil contact, and phosphorus acquisition in soils of different strength. **Journal of Experimental Botany**, *[S. l.]*, v. 64, n. 12, p. 3711–3721, 2013. Disponível em: https://doi.org/10.1093/jxb/ert200

HAYNES, R. J.; ZHOU, Y. F. Effect of pH and added slag on the extractability of Si in two Si-deficient sugarcane soils. **Chemosphere**, *[S. l.]*, v. 193, p. 431–437, 2018. Disponível em: https://doi.org/10.1016/J.CHEMOSPHERE.2017.10.175

HE, H. *et al.* Effects of alkaline and bioorganic amendments on cadmium, lead, zinc, and nutrient accumulation in brown rice and grain yield in acidic paddy fields contaminated with a mixture of heavy metals. **Environmental Science and Pollution Research**, *[S. l.]*, v. 23, n. 23, p. 23551–23560, 2016. Disponível em: https://doi.org/10.1007/S11356-016-7593-1/TABLES/6. Acesso em: 1 dez. 2022.

HILTUNEN, R., AND HILTUNEN, A. "Environmental aspects of the utilization of steel industry slags". *In*: THE SOUTH AFRICAN INSTITUTE OF MINING AND METALLURGY (org.). **Proceedings of VII intenational conference on molten slags, fluxes and salts**. Cape Town, South Africa: *[s. n.]*, 2004. *E-book.*

HOSSEINI, S. *et al.* Environmental Technology Reviews Production and applications of electric-arc-furnace slag as solid waste in environmental technologies: a review. *[S. l.]*, 2016. Disponível em: https://doi.org/10.1080/21622515.2016.1147615

HU, Z. H. *et al.* Combined application of arbuscular mycorrhizal fungi and steel slag improves plant growth and reduces Cd, Pb accumulation in Zea mays. **International Journal of Phytoremediation**, *[S. l.]*, v. 21, n. 9, p. 857–865, 2019. Disponível em: https://doi.org/10.1080/15226514.2019.1577355

HUSSEINY, S. *et al.* Characterization of growth promoting bacterial endophytes isolated from Artemisia annua L. **South African Journal of Botany**, *[S. l.]*, v. 143, p. 238–247, 2021. Disponível em: https://doi.org/10.1016/J.SAJB.2021.07.042. Acesso em: 19 abr. 2022.

IBGE. **PEVS 2021: Produção da Extração Vegetal e da Silvicultura**. 36. ed. Rio de Janeiro: IBGE, 2022. *E-book.*

INDÚSTRIA BRASILEIRA DE ÁRVORES. **Relatório Anual 2021**. São Paulo: Fundação Getulio Vargas (FGV), 2022. Disponível em: https://www.iba.org/datafiles/publicacoes/relatorios/relatorio-anual-iba2022-compactado.pdf

INSTITUTO AÇO BRASIL. **Relatório de Sustentabilidade 2020**2021. p. 62.

Disponível em: https://www.acobrasil.org.br/relatoriodesustentabilidade/

INSTITUTO AÇO BRASIL. **Estatísticas da siderurgia**. *[s. l.]*, 2023. Disponível em: https://acobrasil.org.br/site/.

ISLAM, F. *et al.* Influence of Pseudomonas aeruginosa as PGPR on oxidative stress tolerance in wheat under Zn stress. **Ecotoxicology and Environmental Safety**, *[S. l.]*, v. 104, p. 285–293, 2014. Disponível em: https://doi.org/10.1016/j.ecoenv.2014.03.008

ISLAM, Z. *et al.* Effect of Steel Slag on Soil Fertility and Plant Growth. **Journal of Agricultural Chemistry and Environment**, *[S. l.]*, 2022. Disponível em: https://doi.org/10.4236/jacen.2022.113014. Acesso em: 9 out. 2022.

ITO, K. Steelmaking Slag for Fertilizer Usage. **NIPPON STEEL & SUMITOMO METAL TECHNICAL REPORT**, *[S. l.]*, n. 109, p. 130–136, 2015. Disponível em: https://doi.org/UDC 669 . 184 . 28 : 631 . 82 /. 86

JI, S. H.; GURURANI, M. A.; CHUN, S. C. Isolation and characterization of plant growth promoting endophytic diazotrophic bacteria from Korean rice cultivars. **Microbiological Research**, *[S. l.]*, v. 169, n. 1, p. 83–98, 2014. Disponível em: https://doi.org/10.1016/J.MICRES.2013.06.003

JING, Y.; HE, Z.; YANG, X. Role of soil rhizobacteria in phytoremediation of heavy metal contaminated soils. **Journal of Zhejiang University SCIENCE B**, *[S. l.]*, v. 8, n. 3, p. 192–207, 2007. Disponível em: https://doi.org/10.1631/jzus.2007.B0192

JOÃO LUCAS BOLDT, EDUARDO LUCAS TERRA PEIXOTO, GUSTAVO FERREIRA DE OLIVEIRA, A. H. de M. INOCULATION OF TREE SPECIES WITH MYCORRHIZAL FUNGI FERTILIZED WITH ORGANIC FERTILIZER AND STEEL SLAG IN A LEGAL RESERVE AREA. **Agroecossistemas**, *[S. l.]*, v. 13, n. 1, p. 31–45, 2021.

KAVAMURA, V. N.; MELO, I. S. de. Effects of different osmolarities on bacterial biofilm formation. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 45, n. 2, p. 627–631, 2014. Disponível em: https://doi.org/10.1590/S1517-83822014000200034

KHODAVERDILOO, H. *et al.* Potentially toxic element contamination of arid and semi-arid soils and its phytoremediation. **Arid Land Research and Management**, *[S. l.]*, v. 34, n. 4, p. 361–391, 2020. Disponível em: https://doi.org/10.1080/15324982.2020.1746707. Acesso em: 20 mar. 2021.

KUMAR, P.; SHUKLA, S. Utilization of steel slag waste as construction material: A review. **Materials Today: Proceedings**, *[S. l.]*, v. 78, p. 145–152, 2023. Disponível em: https://doi.org/10.1016/j.matpr.2023.01.015

KURNIATI, E. O.; PEDERSON, F.; KIM, H.-J. Application of steel slags, ferronickel slags, and copper mining waste as construction materials: A review. **Resources, Conservation and Recycling**, *[S. l.]*, v. 198, p. 107175, 2023. Disponível em: https://doi.org/10.1016/j.resconrec.2023.107175

LARRABURU, E. E.; APÓSTOLO, N. M.; LLORENTE, B. E. Anatomy and morphology of photinia (Photinia × fraseri Dress) in vitro plants inoculated with rhizobacteria. **Trees**, *[S. l.]*, v. 24, n. 4, p. 635–642, 2010. Disponível em: https://doi.org/10.1007/s00468-010-0433-x

LEE, D. J. Achievements in forest tree genetic improvement in Australia and New Zealand 2: Development of Corymbia species and hybrids for plantations in eastern Australia. **http://dx.doi.org/10.1080/00049158.2007.10676256**, *[S. l.]*, v. 70, n. 1, p. 11–16, 2013. Disponível em: https://doi.org/10.1080/00049158.2007.10676256. Acesso em: 25 abr. 2023.

LI, Y. *et al.* Toxic effects of cadmium on the physiological and biochemical attributes of plants, and phytoremediation strategies: A review. **Environmental Pollution**, *[S. l.]*, v. 325, p. 121433, 2023. Disponível em: https://doi.org/10.1016/j.envpol.2023.121433

LIMA, J. V. *et al.* Rhizobacteria modify root architecture and improve nutrient uptake in oil palm seedlings despite reduced fertilizer. **Rhizosphere**, *[S. l.]*, p. 100420, 2021. Disponível em: https://doi.org/10.1016/j.rhisph.2021.100420. Acesso em: 12 ago. 2021.

LINCOLN TAIZ; EDUARDO ZEIGER; IAN MAX MØLLER; ANGUS MURPHY. **Fisiologia e desenvolvimento vegetal**. 6. ed. Porto Alegre: Artmed, 2017. *E-book*.

LÓPEZ-BUCIO, J. *et al.* Bacillus megaterium Rhizobacteria Promote Growth and Alter Root-System Architecture Through an Auxin- and Ethylene-Independent Signaling Mechanism in Arabidopsis thaliana. **Molecular Plant-Microbe Interactions®**, *[S. l.]*, v. 20, n. 2, p. 207–217, 2007. Disponível em: https://doi.org/10.1094/MPMI-20-2-0207

MA, Y. *et al.* Plant growth promoting rhizobacteria and endophytes accelerate phytoremediation of metalliferous soils. **Biotechnology Advances**, *[S. l.]*, v. 29, n. 2, p. 248–258, 2011. Disponível em: https://doi.org/10.1016/J.BIOTECHADV.2010.12.001

MA, Y. *et al.* **Beneficial role of bacterial endophytes in heavy metal phytoremediation**. *[S. l.: s. n.]* Disponível em: https://doi.org/10.1016/j.jenvman.2016.02.047

MADHAIYAN, M.; POONGUZHALI, S.; SA, T. Metal tolerating methylotrophic bacteria reduces nickel and cadmium toxicity and promotes plant growth of tomato (Lycopersicon esculentum L.). **Chemosphere**, *[S. l.]*, v. 69, n. 2, p. 220–228, 2007.

Disponível em: https://doi.org/10.1016/j.chemosphere.2007.04.017

MAËLA PERON GOMIDE. **ANATOMIA DE RAÍZES DE Eucalyptus grandis Hill ex Maiden EM DIFERENTES PROFUNDIDADES DO SOLO SOB UM CONTEXTO DE REDUÇÃO DE CHUVAS**. 2016. - Universidade Estadual Paulista “Júlio de Mesquita Filho”, *[s. l.]*, 2016.

MAFIA, R. G. *et al.* Rooting induction and growth of eucalyptus by rhizobacteria: Effect of addition of nutrient source and growing substrate composition. **Revista Arvore**, *[S. l.]*, v. 31, n. 4, p. 589–597, 2007 a. Disponível em: https://doi.org/10.1590/s0100-67622007000400003. Acesso em: 22 abr. 2021.

MAFIA, R. G. *et al.* Compatibility among rhizobacteria isolates and effect of mixture of isolates in inducing rooting and growth of eucalyptus clones. **Revista Arvore**, *[S. l.]*, v. 31, n. 4, p. 635–643, 2007 b. Disponível em: https://doi.org/10.1590/S0100-67622007000400008

MAFIA, R. G. *et al.* Indução do enraizamento e crescimento do eucalipto por rizobactérias: efeito da adição de fonte alimentar e da composição do substrato de enraizamento. **Revista Árvore**, *[S. l.]*, v. 31, n. 4, p. 589–597, 2007 c. Disponível em: https://doi.org/10.1590/S0100-67622007000400003

MAFIA, R. G. *et al.* Microbiolização e interação entre rizobactérias promotoras do crescimento e clones de eucalipto. **Revista Árvore**, *[S. l.]*, v. 33, n. 5, p. 789–797, 2009. Disponível em: https://doi.org/10.1590/S0100-67622009000500002

MAGALHÃES, M. O. L. *et al.* Potencial de duas espécies de eucalipto na fitoestabilização de solo contaminado com zinco. **Revista Ciência Agronômica**, *[S. l.]*, v. 42, n. 3, p. 805–812, 2011. Disponível em: https://doi.org/10.1590/S1806-66902011000300029

MARTÍNEZ-VIVEROS, O. *et al.* MECHANISMS AND PRACTICAL CONSIDERATIONS INVOLVED IN PLANT GROWTH PROMOTION BY RHIZOBACTERIA. **Journal of soil science and plant nutrition**, *[S. l.]*, v. 10, n. 3, 2010. Disponível em: https://doi.org/10.4067/S0718-95162010000100006

MARTINS, M. S. **INOVAÇÕES TECNOLÓGICAS DA INDÚSTRIA 4.0: APLICAÇÕES E IMPLICAÇÕES PARA A SIDERURGIA BRASILEIRA**. 2019. - Universidade Federal de Uberlândia, *[s. l.]*, 2019.

MATSUMOTO, S. *et al.* Inhibition of arsenic accumulation in Japanese rice by the application of iron and silicate materials. **CATENA**, *[S. l.]*, v. 135, p. 328–335, 2015. Disponível em: https://doi.org/10.1016/J.CATENA.2015.07.004

MEENA, K. K. *et al.* Abiotic Stress Responses and Microbe-Mediated Mitigation in Plants: The Omics Strategies. **Frontiers in Plant Science**, *[S. l.]*, v. 8, 2017 a. Disponível em: https://doi.org/10.3389/fpls.2017.00172

MEENA, V. S. *et al.* Plant beneficial rhizospheric microorganism (PBRM) strategies to improve nutrients use efficiency: A review. **Ecological Engineering**, *[S. l.]*, v. 107, p. 8–32, 2017 b. Disponível em: https://doi.org/10.1016/j.ecoleng.2017.06.058

MOURÃO, M. **Introdução à siderurgia**. *[S. l.: s. n.]*. *E-book.*

NAUTIYAL, C. S. An efficient microbiological growth medium for screening phosphate solubilizing microorganisms. **FEMS Microbiology Letters**, *[S. l.]*, v. 170, n. 1, p. 265–270, 1999. Disponível em: https://doi.org/10.1111/J.1574-6968.1999.TB13383.X. Acesso em: 19 abr. 2022.

NING, D. *et al.* Impacts of steel-slag-based silicate fertilizer on soil acidity and silicon availability and metals-immobilization in a paddy soil. **PLoS ONE**, *[S. l.]*, v. 11, n. 12, 2016. Disponível em: https://doi.org/10.1371/journal.pone.0168163

NJOKU, K. L.; AKINYEDE, O. R.; OBIDI, O. F. Microbial Remediation of Heavy Metals Contaminated Media by Bacillus megaterium and Rhizopus stolonifer. **Scientific African**, *[S. l.]*, v. 10, p. e00545, 2020. Disponível em: https://doi.org/10.1016/j.sciaf.2020.e00545

NWIGWE, C.; FOSSEY, A.; DE SMIDT, O. Effects of Plant Growth Promoting Rhizospheric Bacteria (PGPR) on Survival, Growth and Rooting Architecture of Eucalyptus Hybrid Clones. **Forests**, *[S. l.]*, v. 14, n. 9, p. 1848, 2023. Disponível em: https://doi.org/10.3390/f14091848

O'CONNOR, J. *et al.* Production, characterisation, utilisation, and beneficial soil application of steel slag: A review. **Journal of Hazardous Materials**, *[S. l.]*, v. 419, p. 126478, 2021. Disponível em: https://doi.org/10.1016/J.JHAZMAT.2021.126478

OLEŃSKA, E. *et al.* Beneficial features of plant growth-promoting rhizobacteria for improving plant growth and health in challenging conditions: A methodical review. **Science of The Total Environment**, *[S. l.]*, v. 743, p. 140682, 2020. Disponível em: https://doi.org/10.1016/J.SCITOTENV.2020.140682

OLIVEIRA, A. M. de; CENÁRIO. **CENÁRIO BRASILEIRO DE INOCULANTES E APLICAÇÕES DE BACTÉRIAS ENDOFÍTICAS EM Corymbia**. 2023. - UNIVERSIDADE FEDERAL DOS VALES DO JEQUITINHONHA E MUCURI, *[s. l.]*, 2023.

OZA, E. F. *et al.* Aproveitamento de escória de siderurgia em substratos alternativos para produção de mudas de pimenteira Dedo-de-moça. **Revista Ceres**, *[S. l.]*, v. 65, n. 1, p. 104–109, 2018. Disponível em: https://doi.org/10.1590/0034-737x201865010014

PAN, X. *et al.* Exploring root system architecture and anatomical variability in alfalfa (Medicago sativa L.) seedlings. **BMC Plant Biology**, *[S. l.]*, v. 23, n. 1, p. 449, 2023. Disponível em: https://doi.org/10.1186/s12870-023-04469-4

PANDIT, A. *et al.* Microbial biofilms in nature: unlocking their potential for agricultural applications. **Journal of applied microbiology**, *[S. l.]*, v. 129, n. 2, p. 199–211, 2020. Disponível em: https://doi.org/10.1111/JAM.14609. Acesso em: 12 dez. 2022.

PAZ, I. C. P. *et al.* Eucalyptus growth promotion by endophytic Bacillus spp. **Genetics and Molecular Research**, *[S. l.]*, v. 11, n. 4, p. 3711–3720, 2012. Disponível em: https://doi.org/10.4238/2012.August.17.9

PERALTA, K. D. *et al.* Production of phytohormones, siderophores and population fluctuation of two root-promoting rhizobacteria in Eucalyptus globulus cuttings. **World Journal of Microbiology and Biotechnology**, *[S. l.]*, v. 28, n. 5, p. 2003–2014, 2012. Disponível em: https://doi.org/10.1007/s11274-012-1003-8. Acesso em: 22 abr. 2021.

PIATAK, N. M.; PARSONS, M. B.; SEAL, R. R. Characteristics and environmental aspects of slag: A review. **Applied Geochemistry**, *[S. l.]*, v. 57, p. 236–266, 2015. Disponível em: https://doi.org/10.1016/j.apgeochem.2014.04.009

PII, Y. *et al.* **Microbial interactions in the rhizosphere: beneficial influences of plant growth-promoting rhizobacteria on nutrient acquisition process. A review**. *[S. l.: s. n.]* Disponível em: https://doi.org/10.1007/s00374-015-0996-1

PISHCHIK, V. N. *et al.* **Mechanisms of plant and microbial adaptation to heavy metals in plant–microbial systems**. *[S. l.]*: Maik Nauka Publishing / Springer SBM, 2016. Disponível em: https://doi.org/10.1134/S0026261716030097. Acesso em: 1 abr. 2020.

PRADO, R. de M.; FERNANDES, F. M. Efeito da escória de siderurgia e calcário na disponibilidade de fósforo de um Latossolo Vermelho-Amarelo cultivado com cana-de-açúcar. **Pesquisa Agropecuária Brasileira**, *[S. l.]*, v. 36, n. 9, p. 1199–1204, 2001. Disponível em: https://doi.org/10.1590/S0100-204X2001000900014

RAASCH, L. D.; BONALDO, S. M.; OLIVEIRA, A. A. F. de. Bacillus subtilis: ENRAIZAMENTO E CRESCIMENTO DE MINIESTACAS DE EUCALIPTO EM SINOP, NORTE DE MATO GROSSO, BRASIL. **Biosci. J.**, *[S. l.]*, v. 29, n. Supplement 1, p. 1446–1457, 2013.

RADIĆ, S. *et al.* Effect of electric arc furnace slag on growth and physiology of maize (Zea mays L.). **Acta Biologica Hungarica**, *[S. l.]*, v. 64, n. 4, p. 490–499, 2013. Disponível em: https://doi.org/10.1556/ABiol.64.2013.4.8

RAJKUMAR, M.; FREITAS, H. Influence of metal resistant-plant growth-promoting bacteria on the growth of Ricinus communis in soil contaminated with heavy metals. **Chemosphere**, *[S. l.]*, v. 71, n. 5, p. 834–842, 2008. Disponível em: https://doi.org/10.1016/j.chemosphere.2007.11.038. Acesso em: 13 nov. 2022.

RAJKUMAR, M.; MA, Y.; FREITAS, H. Improvement of Ni phytostabilization by inoculation of Ni resistant Bacillus megaterium SR28C. **Journal of Environmental Management**, *[S. l.]*, v. 128, p. 973–980, 2013. Disponível em: https://doi.org/10.1016/J.JENVMAN.2013.07.001

RÊGO, M. C. F. *et al.* Morphoanatomical and Biochemical Changes in the Roots of Rice Plants Induced by Plant Growth-Promoting Microorganisms. **Journal of Botany**, *[S. l.]*, v. 2014, p. 1–10, 2014. Disponível em: https://doi.org/10.1155/2014/818797

RÊGO, M. C. F. *et al.* The role of rhizobacteria in rice plants: Growth and mitigation of toxicity. **Journal of Integrative Agriculture**, *[S. l.]*, v. 17, n. 12, p. 2636–2647, 2018. Disponível em: https://doi.org/10.1016/S2095-3119(18)62039-8

REIS, C. A. F. *et al.* **Corymbia citriodora: estado da arte de pesquisas no Brasil**. *[s. l.]*, 2013. Disponível em: https://ainfo.cnptia.embrapa.br/digital/bitstream/item/94547/1/Doc.-255-Reis-Ainfo1.pdf. Acesso em: 10 out. 2023.

REIS, C. A. F. *et al.* **Corymbia torelliana: estado da arte de pesquisas no Brasil**. Colombo: Embrapa Florestas, 2014.

RODRÍGUEZ-DÍAZ, M., RODELAS-GONZALÉS, B., POZO-CLEMENTE, C, MARTÍNEZ-TOLEDO, MV, GONZÁLEZ-LÓPEZ, J. Uma revisão sobre a taxonomia e possíveis características de triagem de rizobactérias promotoras de crescimento de plantas. *In*: AHMAD, J. PICHTEL, J., S. H. (org.). **Interações Planta-Bactérias: Estratégias e Técnicas para Promover o Crescimento das Plantas**. Weinheim, Alemanh: Wiley-VCH Verlag GmbH & Co., 2008. p. 55–80. *E-book*.

ROZEFELDS, A. C. Eucalyptus phylogeny and history: A Brief Summary. **Tasforests**, *[S. l.]*, v. 8, p. 11, 1996.

SAMANIEGO-GÁMEZ, B. Y. *et al.* Bacillus spp. inoculation improves photosystem II efficiency and enhances photosynthesis in pepper plants. **Chilean journal of agricultural research**, *[S. l.]*, v. 76, n. 4, p. 409–416, 2016. Disponível em:

https://doi.org/10.4067/S0718-58392016000400003. Acesso em: 18 out. 2022.

SANTOS, L. F. dos; OLIVARES, F. L. Microbial inoculants in agriculture and its effects on plant microbiome. *In*: **Rhizosphere Engineering**. *[S. l.]*: Elsevier, 2022. p. 151–169. *E-book*. Disponível em: https://doi.org/10.1016/B978-0-323-89973-4.00016-8

SCHWYN, B.; NEILANDS, J. B. Universal chemical assay for the detection and determination of siderophores. **Analytical Biochemistry**, *[S. l.]*, v. 160, n. 1, p. 47–56, 1987. Disponível em: https://doi.org/10.1016/0003-2697(87)90612-9

SHARMA, P. Role and significance of biofilm-forming microbes in phytoremediation - A review. **Environmental Technology & Innovation**, *[S. l.]*, v. 25, p. 102182, 2022. Disponível em: https://doi.org/10.1016/J.ETI.2021.102182

SHENG, X.-F. *et al.* Characterization of heavy metal-resistant endophytic bacteria from rape (Brassica napus) roots and their potential in promoting the growth and lead accumulation of rape. **Environmental Pollution**, *[S. l.]*, v. 156, n. 3, p. 1164–1170, 2008. Disponível em: https://doi.org/10.1016/j.envpol.2008.04.007

SHEPHERD, M. *et al.* Mapping species differences for adventitious rooting in a Corymbia torelliana × Corymbia citriodora subspecies variegata hybrid. **Tree Genetics & Genomes**, *[S. l.]*, v. 4, n. 4, p. 715–725, 2008. Disponível em: https://doi.org/10.1007/s11295-008-0145-1

SHI, R. yong *et al.* Effects of biomass ash, bone meal, and alkaline slag applied alone and combined on soil acidity and wheat growth. **Journal of Soils and Sediments**, *[S. l.]*, v. 17, n. 8, p. 2116–2126, 2017. Disponível em: https://doi.org/10.1007/S11368-017-1673-9/FIGURES/6. Acesso em: 3 nov. 2022.

SHIN, M.-N. *et al.* Characterization of lead resistant endophytic Bacillus sp. MN3-4 and its potential for promoting lead accumulation in metal hyperaccumulator Alnus firma. **Journal of Hazardous Materials**, *[S. l.]*, v. 199–200, p. 314–320, 2012. Disponível em: https://doi.org/10.1016/j.jhazmat.2011.11.010

SHU, K.; SASAKI, K. Occurrence of steel converter slag and its high value-added conversion for environmental restoration in China: A review. **Journal of Cleaner Production**, *[S. l.]*, v. 373, p. 133876, 2022. Disponível em: https://doi.org/10.1016/J.JCLEPRO.2022.133876

SILVA, A. V. da *et al.* Physiological, biochemical and nutritional changes in soybean in response to application of steel slag. **Australian Journal of Crop Science**, *[S. l.]*, v. 15, n. 15(12):2021, p. 1399–1405, 2021. Disponível em: https://doi.org/10.21475/ajcs.21.15.12.p3269. Acesso em: 27 ago. 2022.

SINGH, V. *et al.* Plant Growth Promoting Rhizobacteria (PGPR): Current and Future Prospects for Development of Sustainable Agriculture. **J Microb Biochem Technol**, *[S. l.]*, v. 7, n. 2, p. 96–102, 2015. Disponível em: https://doi.org/10.4172/1948-5948.1000188

SOBRAL, M. F. *et al.* Basic slag and its effects on the concentration of nutrients and heavy metals in sugarcane. **Revista Brasileira de Engenharia Agricola e Ambiental**, *[S. l.]*, v. 15, n. 8, p. 867–872, 2011. Disponível em: https://doi.org/10.1590/S1415-43662011000800015

TEIXEIRA, D. A. *et al.* RHIZOBACTERIAL PROMOTION OF EUCALYPT ROOTING AND GROWTH. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 38, p. 118–123, 2007 a. Disponível em: www.cme.msu.edu/RDP/

TEIXEIRA, D. A. *et al.* Rhizobacterial promotion of eucalypt rooting and growth. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 38, n. 1, p. 118–123, 2007 b. Disponível em: https://doi.org/10.1590/S1517-83822007000100025

TIWARI, M.; BAJPAI, D. S.; DEWANGAN, D. U. Steel Slag Utilization — Overview in Indian Perspective. **International Journal of Advanced Research**, *[S. l.]*, v. 4, n. 8, p. 2232–2246, 2016. Disponível em: https://doi.org/10.21474/IJAR01/1442

TRUEMAN, S. J. Cytokinin and auxin effects on survival and rooting of Eucalyptus pellita and E. grandis × E. pellita cuttings. **Rhizosphere**, *[S. l.]*, v. 6, p. 74–76, 2018. Disponível em: https://doi.org/10.1016/J.RHISPH.2018.04.001

TRUEMAN, S. J.; RICHARDSON, D. M. Relationships between Indole-3-Butyric Acid, Photoinhibition and Adventitious Rooting of Corymbia torelliana, C. citriodora and F 1 Hybrid Cuttings. **Tree and Forestry Science and Biotechnology**, *[S. l.]*, v. 2, n. 1, p. 26–33, 2008.

TU, X. *et al.* Advantages and risks of using steel slag in preparing composts from raw organic waste. **Journal of Environmental Science and Health - Part B Pesticides, Food Contaminants, and Agricultural Wastes**, *[S. l.]*, v. 52, n. 1, p. 30–36, 2017. Disponível em: https://doi.org/10.1080/03601234.2016.1224701

VALENTE LIMA, J. *et al.* Rhizobacteria modify root architecture and improve nutrient uptake in oil palm seedlings despite reduced fertilizer. **Rhizosphere**, *[S. l.]*, v. 19, p. 100420, 2021. Disponível em: https://doi.org/10.1016/J.RHISPH.2021.100420

VEJAN, P. *et al.* Role of Plant Growth Promoting Rhizobacteria in Agricultural Sustainability—A Review. **Molecules 2016, Vol. 21, Page 573**, *[S. l.]*, v. 21, n. 5, p. 573, 2016. Disponível em: https://doi.org/10.3390/MOLECULES21050573. Acesso em: 13 fev. 2023.

VERMA, S.; KUILA, A. Bioremediation of heavy metals by microbial process. **Environmental Technology & Innovation**, *[S. l.]*, v. 14, p. 100369, 2019. Disponível em: https://doi.org/10.1016/j.eti.2019.100369

VOCCIANTE, M. *et al.* The Role of Plant Growth-Promoting Rhizobacteria (PGPR) in Mitigating Plant's Environmental Stresses. **Applied Sciences**, *[S. l.]*, v. 12, n. 3, p. 1231, 2022. Disponível em: https://doi.org/10.3390/app12031231

WANG, G. *et al.* The combined use of a plant growth promoting Bacillus sp. strain and GABA promotes the growth of rice under salt stress by regulating antioxidant enzyme system, enhancing photosynthesis and improving soil enzyme activities. **Microbiological Research**, *[S. l.]*, v. 266, p. 127225, 2023. Disponível em: https://doi.org/10.1016/J.MICRES.2022.127225

WANG, J. *et al. Bacillus velezensis* SX13 promoted cucumber growth and production by accelerating the absorption of nutrients and increasing plant photosynthetic metabolism. **Scientia Horticulturae**, *[S. l.]*, v. 301, p. 111151, 2022. Disponível em: https://doi.org/10.1016/J.SCIENTA.2022.111151

WANG, X. *et al.* Isolation of As-tolerant bacteria and their potentials of reducing As and Cd accumulation of edible tissues of vegetables in metal(loid)-contaminated soils. **Science of the Total Environment**, *[S. l.]*, v. 579, 2017. Disponível em: https://doi.org/10.1016/j.scitotenv.2016.10.239

WANG, X. *et al.* Environmental risks for application of iron and steel slags in soils in China: A review. **Pedosphere**, *[S. l.]*, v. 31, n. 1, p. 28–42, 2021. Disponível em: https://doi.org/10.1016/S1002-0160(20)60058-3

WANI, P. A.; KHAN, M. S. Bacillus species enhance growth parameters of chickpea (Cicer arietinum L.) in chromium stressed soils. **Food and Chemical Toxicology**, *[S. l.]*, v. 48, n. 11, p. 3262–3267, 2010. Disponível em: https://doi.org/10.1016/j.fct.2010.08.035

WHITE, B. *et al.* Effect of silicate slag application on wheat grown under two nitrogen rates. **Plants**, *[S. l.]*, v. 6, n. 4, 2017. Disponível em: https://doi.org/10.3390/plants6040047

WORLD STEEL ASSOCIATION. **Sustainable steel: Sustainability Indicators 2021**. Beijing: China: World Steel, 2022. Disponível em: https://worldsteel.org/publications/bookshop/sust2021/

YI, H. *et al.* An Overview of Utilization of Steel Slag. **Procedia Environmental**

Sciences, *[S. l.]*, v. 16, p. 791–801, 2012. Disponível em: https://doi.org/10.1016/j.proenv.2012.10.108

YILDIRIM, I. Z.; PREZZI, M. Chemical, Mineralogical, and Morphological Properties of Steel Slag. **Advances in Civil Engineering**, *[S. l.]*, v. 2011, p. 1–13, 2011. Disponível em: https://doi.org/10.1155/2011/463638

ZANONI DO PRADO, D. *et al.* Aspergillus flavipes as a novel biostimulant for rooting-enhancement of Eucalyptus. **Journal of Cleaner Production**, *[S. l.]*, v. 234, p. 681–689, 2019. Disponível em: https://doi.org/10.1016/J.JCLEPRO.2019.06.211

4. APLICAÇÃO COMBINADA DE *Bacillus* sp. E ESCÓRIA SIDERÚRGICA MELHORA A MORFOANATOMIA RADICULAR E O CRESCIMENTO DE *Corymbia* sp.

RESUMO

A reciclagem da escória de aciaria de forno elétrico a arco (EAE), um subproduto do refino do aço combinada a rizobactérias é pouco explorada para uso como bioinsumos na agricultura e informações sobre os efeitos desta combinação nos aspectos fisiológicos, morfoanatômicos e de crescimento da planta são fundamentais para a utilização como insumo agrícola. O objetivo deste trabalho foi investigar as alterações morfo-anatômicas em raízes e parâmetros de eficiência fotossintética em mudas de *Corymbia torelliana x Corymbia citriodora* desencadeadas pela inoculação da cepa *Bacillus* sp. CCMD862 associada a fertilização com (EAE). O delineamento foi inteiramente casualizado com quatro tratamentos: T1 – plantas controle sem inoculação e sem EAE; T2 – plantas inoculadas com *Bacillus* sp.; T3- inoculadas com *Bacillus* sp. e com EAE e T4 – plantas com EAE, com 8 repetições para cada tratamento, em casa de vegetação. Os resultados indicam a viabilidade de reciclagem de EAE na produção de mudas de *Corymbia* associada a inoculação de *Bacillus* sp. CCMD862, com ganhos de 41% em assimilação líquida de CO2, 71% em eficiência de carboxilação e 66% em eficiência intrínseca de uso da água, quando comparado a aplicação somente de EAE nas mudas. *Bacillus* sp. + EAE promoveu alterações na arquitetura radicular, aumentado em comprimento, diâmetro, área superficial, volume e número de pontas em relação aos demais tratamentos. Além disso, as alterações anatômicas com ganhos em diâmetro de raiz e xilema, bem como no diâmetro da fibras de floema resultaram em maior acúmulo de biomassa radicular e ganho de robustez. Nossos achados sugerem que a inoculação de *Bacillus* sp. em combinação com escória de aciaria de forno elétrico a arco pode ser um método potencial não apenas para a reciclagem deste resíduo, mas também para a produção segura de mudas florestais com maior robustes, contribuindo para a qualidade final e o estabelecimento em campo.

Palavras-chave: Performance fotossintética. Arquitetura radicular. PGPR. Eucalipto. Resíduo de aço.

COMBINED APPLICATION OF *Bacillus* sp. AND STEEL SLAG IMPROVES ROOT MORPHOANATOMY AND GROWTH OF *Corymbia* sp.

ABSTRACT

The recycling of electric arc furnace slag (EAE), a by-product of steel refining, combined with rhizobacteria is little explored for use as a bioimput in agriculture and information on the effects of this combination on physiological, morphoanatomical and on plant growth is essential for its use as an agricultural input. The aim of this study was to investigate the morpho-anatomical changes in roots and photosynthetic efficiency parameters in *Corymbia torelliana* x *Corymbia citriodora* seedlings triggered by the inoculation of *Bacillus* sp. strain CCMD862 associated with fertilization with EAE. The experimental design was entirely randomized with four treatments: T1 - control plants without inoculation and without EAE; T2 - plants inoculated with Bacillus sp.; T3- inoculated with Bacillus sp. and with EAE and T4 - plants with EAE, with 8 replicates for each treatment, in a greenhouse. The results indicate the viability of recycling EAE in the production of *Corymbia* seedlings associated with inoculation of *Bacillus* sp. CCMD862, with gains of 41% in CO2 net assimilation, 71% in carboxylation efficiency and 66% in intrinsic water use efficiency, when compared to the application of EAE alone. *Bacillus* sp. + EAE promoted changes in root architecture, increasing length, diameter, surface area, volume and number of tips compared to the other treatments. In addition, anatomical changes with increases in root and xylem diameter, as well as phloem fiber diameter, resulted in higher root biomass accumulation and increased robustness. Our findings suggest that the inoculation of *Bacillus* sp. in combination with electric arc furnace slag could be a potential method not only for recycling this waste, but also for the safe production of more robust forest seedlings, contributing to final quality and field establishment.

Keywords: Photosynthetic performance. Root architecture. PGPR. Eucalyptus. Steel waste.

4.1 Introdução

A área total de árvores plantadas no Brasil atingiu 9,93 milhões de hectares em 2021, dos quais 7,53 milhões continham plantações de eucalipto (INDÚSTRIA BRASILEIRA DE ÁRVORES, 2022). Os plantios de eucalipto são instalados geralmente em solos de baixa fertilidade, requerendo significativas quantidades de fertilizantes para produzir madeira com rendimentos economicamente esperados, os quais podem representar até metade dos custos de implementação, o que tem incentivado a pesquisa por fontes alternativas de nutrientes de menor custo (CARDOSO *et al.*, 2022) e outras técnicas que contribuam para o otimização dos programas de fertilização.

As escórias siderúrgicas geradas durante a produção do aço, contêm elevados teores de óxidos de cálcio e magnésio e outros elementos benéficos para a promoção do crescimento das plantas, como Si, P, K e Fe, podendo ser reaproveitadas como fertilizante e para melhoria da qualidade do solo (DAS *et al.*, 2020; GUO; BAO; WANG, 2018). Estes subprodutos também podem conter traços de Cu, Zn, Mo, Cr, Pb, Cd e Hg, no entanto estudos vem demonstrando que a liberação desses metais pesados em solos suplementados com escórias siderúrgicas é irrelevante em termos de poluição ambiental (CORRÊA *et al.*, 2008; WANG *et al.*, 2021).

A melhoria da fertilidade do solo e da nutrição de plantas promovida pela utilização de escórias siderúrgicas foi observada em espécies agronômicas como soja (SILVA *et al.*, 2021), trigo (SHI *et al.*, 2017; WHITE *et al.*, 2017), milho (RADIĆ *et al.*, 2013), alfafa (CAI *et al.*, 2022a) gerando efeitos positivos sobre o crescimento em altura da planta, biomassa total, crescimento radicular e produtividade das culturas.

A quantidade de trabalhos reportando a aplicação de escórias siderúrgicas em espécies florestais é reduzida, mas os resultados demonstram que estes subprodutos podem constituir uma alternativa de baixo custo que melhora a qualidade do solo e promove o crescimento das árvores, como observado para *Hymenaea courbaril*, *Lecythis pisonis*, *Carapa guianensis*, *Jacaranda mimosifolia* e *Eucalyptus urophylla* (BOLDT et al., 2021; GUERRINI *et al.*, 2017; MAGALHÃES *et al.*, 2011).

Os fertilizantes silicatados, como as escórias siderúrgicas, podem ter sua eficiência otimizada pela combinação com microrganismos promotores de crescimento vegetal (HU *et al.*, 2019) , os quais interferem positivamente no crescimento das plantas, representando uma solução sustentável e promissora para aumentar a produtividade, entre outras funcionalidades.

As rizobactérias promotoras de crescimento de plantas (PGPR) são bactérias de vida livre, que habitam a rizosfera e o rizoplano ou endofíticas que se alojam no interior dos tecidos vegetais e promovem o crescimento deste através da combinação complexa de mecanismos diretos como a FBN, solubilização de fosfatos e produção de fitormônios (ELSAYED *et al.*, 2022; WANG *et al.*, 2022; EGAMBERDIEVA *et al.*, 2017) e indiretos como a indução de resistência sistêmica (ETESAMI, 2020; GOSWAMI; DEKA, 2020), que afetam tanto o desenvolvimento quanto a nutrição das plantas (CALVO *et al.*, 2019).

Estes mecanismos complexos não totalmente elucidados, promovem alterações morfológicas e anatômicas, que desencadeiam respostas fisiológicas positivas das plantas favorecendo o crescimento e aumentando a produtividade das culturas (VEJAN *et al.*, 2016). Melhorias em comprimento, quantidade, espessura das raízes e densidade de pêlos absorventes resultam em maior capacidade da planta para explorar o solo em busca de água e nutrientes (ANGULO *et al.*, 2014b; CALVO et al., 2014; PII *et al.*, 2015).

Internamente, aumentos no diâmetro do cilindro vascular, incremento do número de elementos de vaso de metaxilema bem como seus diâmetros, são modificações induzidas por PGPR que estão relacionadas com maior transporte de água e translocação de íons e acúmulo de biomassa (PAN *et al.*, 2023; RÊGO *et al.*, 2014).

A hipótese testada foi que de mudas clonais de um híbrido de *Corymbia* tratadas com *Bacillus* sp. e fertilizadas com escória siderúrgica possuem alterações morfo-anatômicas e fisiológicas relacionadas a melhoria do crescimento. O objetivo deste trabalho foi investigar as alterações morfo-anatômicas em raízes e parâmetros de eficiência fotossintética em mudas de *Corymbia torelliana x Corymbia citriodora* desencadeadas pela inoculação da cepa *Bacillus* sp. CCMD862 associada a fertilização com escória de aciaria de forno elétrico a arco (EAE).

4.2 Material e métodos

4.2.1 Material vegetal e rizobactéria

As mudas híbridas obtidas do cruzamento entre os clones *Corymbia torelliana* (F.Muell.) K.D.Hill & L.A.S.Johnson x *Corymbia citriodora* (Hook.) K.D.Hill & L.A.S.Johnson foram adquiridas em viveiro comercial e apresentavam 40 dias de idade, tendo em média 24 cm de altura e com 6 pares de folhas, em excelente condição fitossanitária.

O isolado de *Bacillus* sp. (CCMD862) foi selecionado em experimentos prévios de promoção de crescimento e está depositado na Coleção de Cultura Maria de Lourdes Reis Duarte do Laboratório de Proteção de Plantas da Universidade Federal Rural da Amazônia, município de Belém, no estado do Pará.

O inóculo bacteriano foi preparado segundo a metodologia proposta por Castro et al. (2020), cultivando a rizobactéria em meio de cultivo ágar nutriente por 48h, à 28 ºC. Uma suspensão bacteriana foi posteriormente preparada com água estéril e a concentração ajustada em espectrofotômetro para 10^8 UFC mL^{-1}, no comprimento de onda de 540nm.

Para a inoculação, as mudas foram retiradas dos tubetes e tiveram o ápice do sistema radicular seccionado em aproximadamente 1cm, em seguida foram inoculadas com 10 mL da suspensão bacteriana por 30 minutos e imediatamente transplantadas. Após 15 dias, nova inoculação foi realizada, com 10mL da suspensão bacteriana via rega, no colo da planta. As plantas do tratamento Controle receberam o mesmo volume de água destilada estéril.

4.2.2 Delineamento experimental

O delineamento foi inteiramente casualizado com quatro tratamentos: T1 – plantas controle sem inoculação e sem EAE (escória de aciaria de forno elétrico a arco); T2 – plantas inoculadas com *Bacillus* sp.; T3- inoculadas com *Bacillus* sp. e com EAE e T4 – plantas com EAE, com 8 repetições para cada tratamento.

O experimento foi conduzido entre os meses de setembro e outubro de 2022, em casa de vegetação. Foi utilizado Latossolo Amarelo distrófico com textura média (P 60,5; K 38,0; Na 6,4 em mg dm^{-3}; Al 0,06; Ca 3,11; Ca+Mg 3,85 em $cmol_c$ dm^{-3}; Fe 85,5; Zn 2,5; Cu 2,2; Mn 20,2 em mg kg^{-1}, pH 5,7). Optou-se por não realizar adubação adicional além da escória siderúrgica incorporada.

O resíduo EAE foi gentilmente cedido pela Siderúrgica Norte Brasil S.A, localizada em Marabá, no estado do Pará. Após a moagem e peneiramento (fração ≤ 2mm), para compor o tratamento com EAE, foram incorporados 10 g de EAE para cada kg de substrato. Os substratos foram acondicionados em sacos plásticos e incubados por 20 dias, sendo homogeneizado a cada 2 dias e a umidade mantida em 50% da capacidade de retenção de água. Após este período, os substratos foram transferidos para vasos com capacidade para 1,5 dm^3, padronizados com 1,5 kg.

A condução do experimento foi de 21 dias e a rega foi diária e manual para repor perdas por evapotranspiração.

4.2.3 Análise de trocas gasosas

Os parâmetros de trocas gasosas e fotossintéticos foram estimados no primeiro ou segundo par de folhas fisiologicamente maduras e totalmente expandidas, do ápice para a base, quando as mudas apresentavam 61 dias. A taxa fotossintética (*A*), a condutância estomática ao vapor de água (*gs*), a concentração intercelular de CO_2 (*Ci*) e a taxa de transpiração foliar (*E*) foram medidas utilizando-se um analisador de gases a infravermelho em sistema aberto (IRGA – Infrared Gas Analyzer), modelo LI 6400XT (LI-COR, Lincoln, NE, EUA), com fluorômetro acoplado (LI-6400-40, LI-COR Inc.) no intervalo entre 10 e 12h, definido em análises preliminares nas mesmas condições experimentais. As condições ambientais na câmara do IRGA foram mantidas a 25 ºC e o déficit de pressão de vapor entre 1,2 e 1,8 kPa, a concentração de CO_2 de referência de 400 µmol mol^{-1}de ar e radiação ativa (PAR) de 1000 µmol de fótons $m^{-2}s^{-1}$. A quantidade de luz azul aplicada foi de 10% da densidade do fluxo fotossintético para maximizar a abertura estomática (MAXWELL; JOHNSON, 2000). As condições ambientais no interior da casa de vegetação durante as medições foram a temperatura do ar de 34 ± 2 °C, umidade relativa do ar de 53 ± 2%, radiação incidente de 680 ± 100 µmol m^{-2} s^{-1} e déficit de pressão de vapor do ar de 2,1 ± 0,14 KPa.

4.2.4 Descrição da anatomia e morfometria

Ao término do tempo experimental, as plantas foram retiradas dos vasos. As raízes foram cuidadosamente lavadas, amostras de aproximadamente 0,5 cm de comprimento de uma raiz adventícia de maior calibre foram coletadas no terço médio do sistema radicular e fixadas em FAA 50%. As amostras permaneceram por 24 horas no fixador, sendo então descartado e as amostras armazenadas em etanol 70% até o momento das análises.

As amostras foram reidratadas em série etanólica graduada para obtenção de cortes e analisadas por microscopia óptica. Cinco cortes transversais à mão livre foram obtidos das raízes. Os cortes foram clareados em hipoclorito de sódio NaClO 2% por 1 minuto, colocados em Astrablau 1% por 30 segundos e depois colocados em Fucsina Básica 1%. As medições foram realizadas utilizando o software ImageJ 1.54d (2012).

Além da descrição anatômica, foram medidos o diâmetro radicular, a espessura do córtex, o diâmetro do cilindro vascular, o número e o diâmetro de elementos de vasos do metaxilema e diâmetro das fibras do floema.

4.2.5 Arquitetura radicular

A arquitetura do sistema radicular das mudas foi avaliada no software WinRHIZO Pro 2007a (Régent Instrum. Quebec, Canadá), acoplado a um scanner profissional Epson XL 10000 equipado com unidade de luz adicional (TPU). Foi utilizada uma definição de 600 (dpi) para as medidas. As raízes foram transferidas da solução etanólica 30% para uma bandeja em acrílico de 30 cm de largura e 40 cm de comprimento contendo água. Foi possível a obtenção de imagens em escala de cinza, baseado em um método de esqueletização em três dimensões (largura, altura e profundidade) sendo evitada ao máximo a sobreposição das raízes. O sistema foi usado para obter as seguintes variáveis do sistema radicular: comprimento total, área superficial total, volume, diâmetro médio, número de pontas.

4.2.6 Biometria e biomassa

Ao final do tempo experimental foram realizadas análise biométricas de altura (régua graduada), diâmetro do coleto (paquímetro digital) e área foliar (medidor de área LICOR-3100). A biomassa seca foi determinada em balança analítica após a secagem em estufa a 50 °C até peso constante.

4.2.7 Análise estatística

Os dados foram submetidos as análises de normalidade e homoscedasticidade para atender as pressuposições da análise de variância (ANOVA), e quando significativos, as médias dos tratamentos foram comparadas pelo teste de Duncan ($P \leq 0,05$) utilizando o software RStudio 4.3.0 (2023).

4.3 Resultados

4.3.1 Trocas gasosas

A inoculação com *Bacillus* sp. influenciou significativamente os parâmetros de trocas gasosas, com incrementos de 21% em assimilação líquida de CO_2 (A), 16% em transpiração (*E*) e 19% em eficiência de carboxilação (*A/Ci*) em relação às plantas não inoculadas e não fertilizadas (Controle). As variações observadas em condutância estomática (*gs*), concentração intercelular de CO_2 (*Ci*) e eficiência intrínseca do uso da água (*A/gs*) não diferiram do tratamento controle (Figura 1).

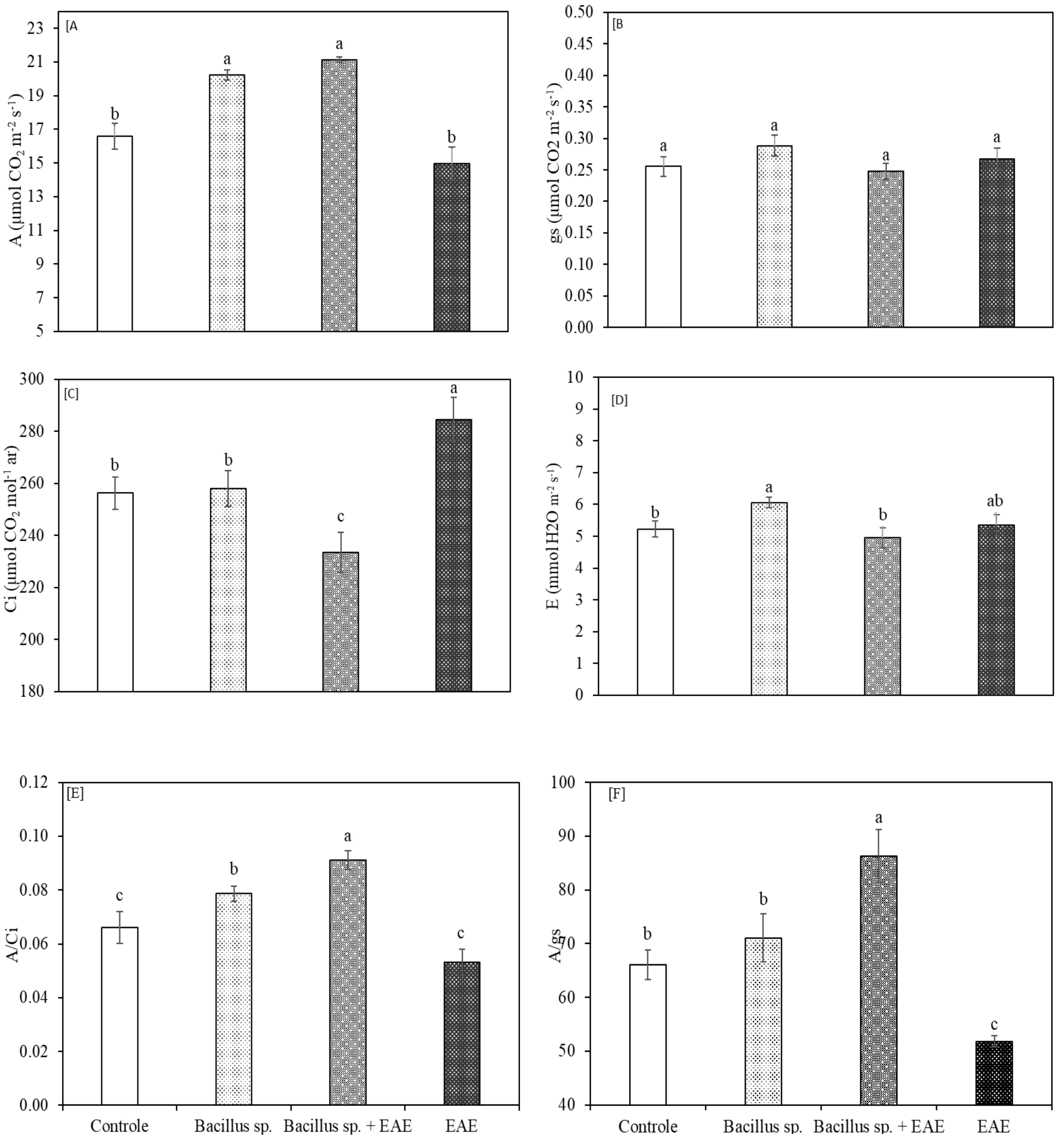


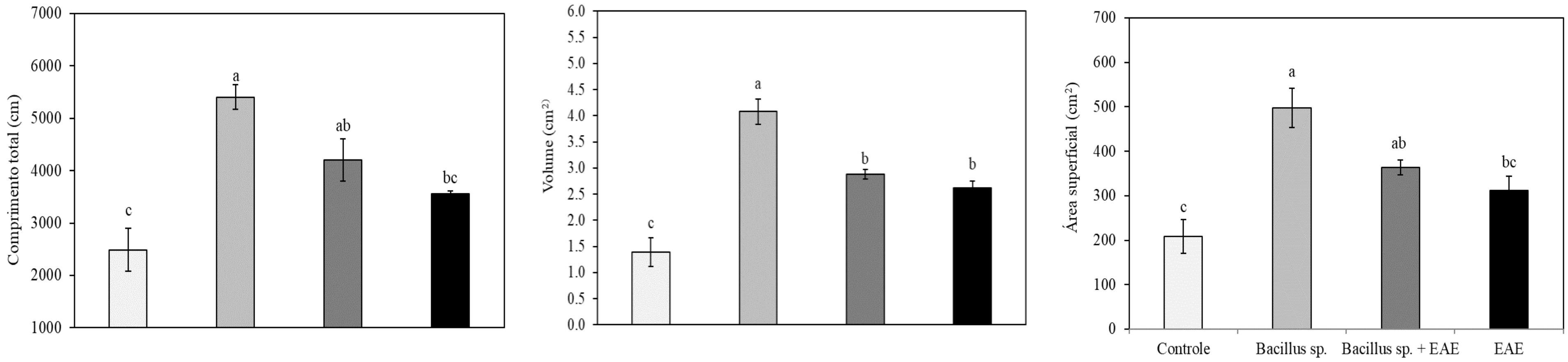
Figura 1. [A]: Taxa de assimilação líquida de CO2 (*A*), [B]: condutância estomática ao vapor de água (*gs*), [C]: concentração intercelular de CO2 (*Ci*), [D] transpiração (*E*), [E]: eficiência de carboxilação (*A/Ci*) e [F] eficiência intrínseca do uso da água (A/Gs) em mudas *Corymbia* tratadas com *Bacillus* sp., EAE, *Bacillus* sp.+ EAE. As médias com letras iguais não diferem significativamente de acordo com o teste de Duncan ($P < 0,05$).

Por outro lado, a fertilização com EAE reduziu em 10% a taxa de assimilação de CO_2 (A), em 22% A/Ci e 15% em A/gs e aumentou em 11% o Ci comparado ao Controle.

O tratamento *Bacillus* sp. + EAE resultou em aumentos que variaram de 27 a 41% em *A*, 38 a 71% em *A/Ci* e 31 a 66% em *A/gs* e reduções de 9 a 18% em *Ci* em relação ao Controle e EAE, respectivamente (Figura 1).

4.3.2 Arquitetura radicular

A rizobactéria *Bacillus* sp. promoveu aumentos de 117% em comprimento total de raiz, 138% em área superficial, 193% em volume de raiz, 51% em número de pontas e 28% em diâmetro médio comparado ao controle. A associação da rizobactéria à fertilização com EAE (*Bacillus* sp. + EAE) resultou em ganhos comparados tanto às plantas controle quanto às plantas apenas fertilizadas com EAE. Os incrementos variaram de 18 a 69% em comprimento total, de 17 a 74% em área superficial, de 11 a 31% o diâmetro médio, de 10 a 107% o volume e de 3 a 18% o número de pontas, comparando-se ao tratamento EAE e ao controle, respectivamente (Figura 2).



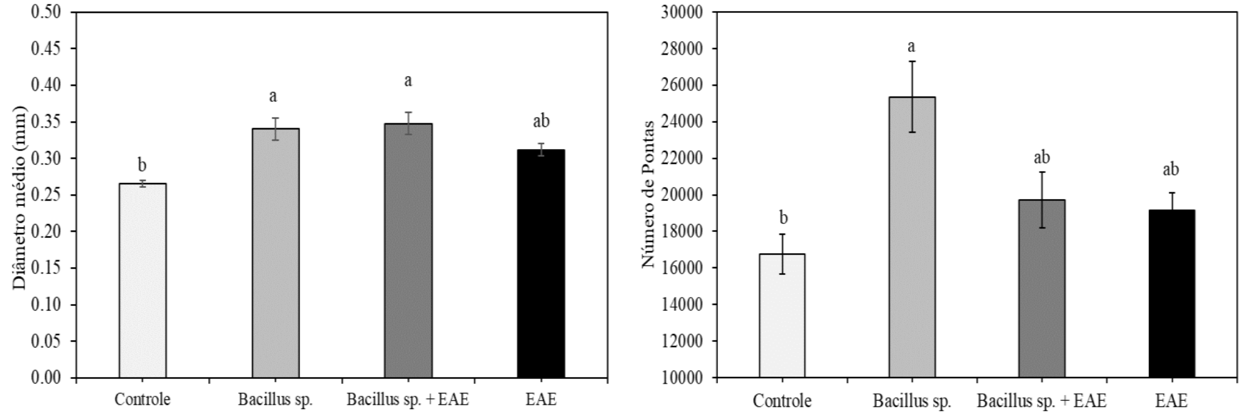


Figura 2. Características da arquitetura radicular em mudas de *Corymbia* submetidas a inoculação com *Bacillus* sp. e fertilização com escória de aciaria de forno a arco (EAE). Médias com letras iguais não diferem pelo teste de Duncan (P<0,05).

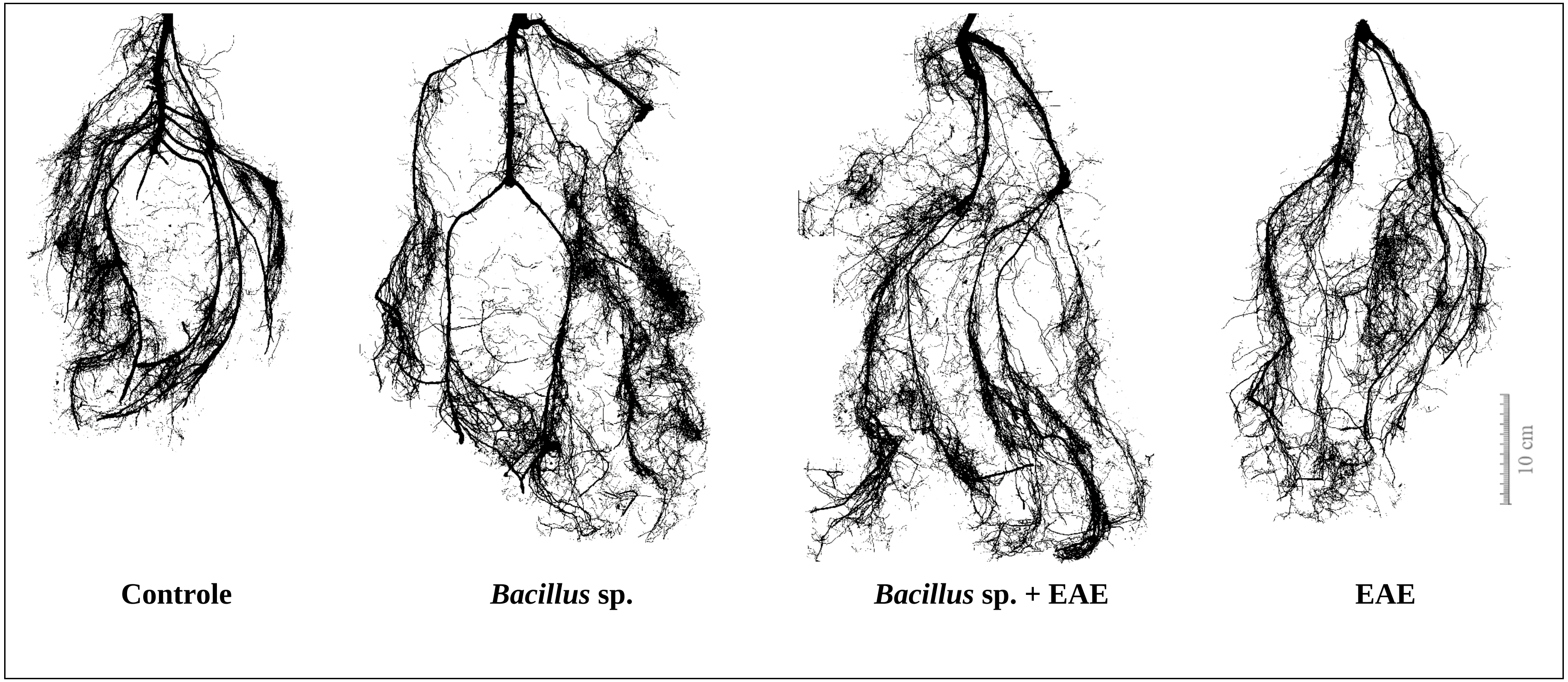


Figura 3. Arquitetura radicular de mudas do híbrido *C. torelliana x C. citriodora* inoculadas com *Bacillus* sp., fertilizadas com escória de aciaria de forno a arco (EAE) e a combinação destes (*Bacillus* sp. + EAE).

4.3.3 Descrição Anatômica e Morfometria

As raízes amostradas apresentavam crescimento secundário, apresentando o desenvolvimento inicial da periderme com camada suberizada ainda delgada, floema secundário, câmbio vascular e pêlos absorventes (Figura 4. d1-d2).

O diâmetro radicular aumentou em todos os tratamentos investigados. Esta alteração foi especialmente acentuada nos tratamentos com *Bacillus* sp + EAE e EAE, para os quais se observaram aumentos de 31% e 21% em relação ao controle (Tabela 1). Além disso, observações microscópicas e morfometria revelaram que algumas características anatômicas das raízes foram alteradas por todos os tratamentos, como espessura do córtex, diâmetro do cilindro vascular, diâmetro de elementos de vaso do metaxilema e diâmetro das fibras do floema (Figura 4).

A espessura do córtex foi 38% significativamente maior nas plantas fertilizadas com EAE. Os aumentos promovidos por *Bacillus* sp. + EAE (30%) e *Bacillus* sp. (21%) não diferiram do controle. Os tratamentos *Bacillus* sp, e *Bacillus* sp. + EAE promoveram cerca de 20% de aumento no diâmetro do cilindro vascular comparado ao controle (Tabela 1).

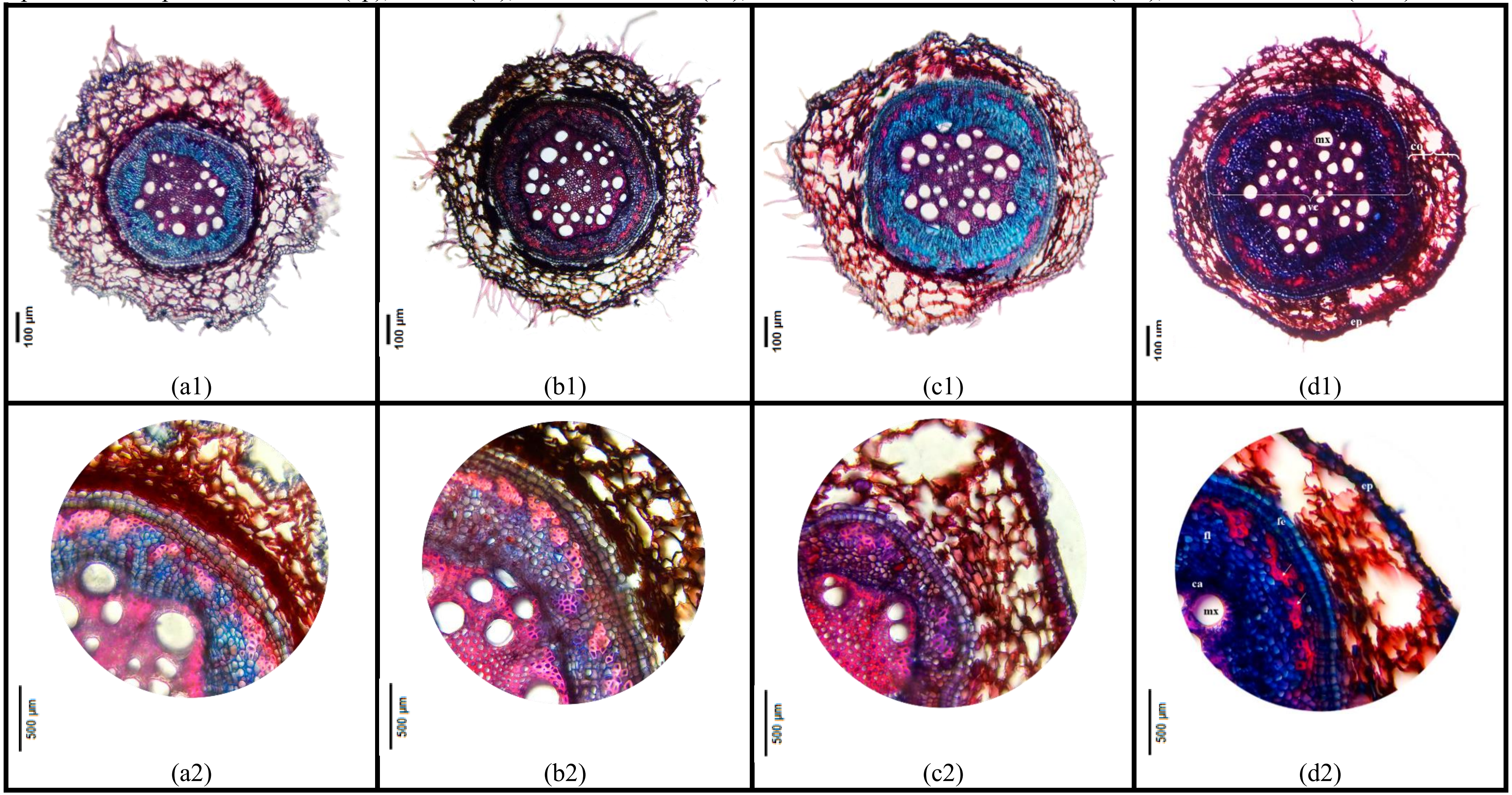
O tratamento *Bacillus* sp. + EAE aumentou o diâmetro de elementos de vaso de metaxilema em 40% em reação ao tratamento controle e em 63% comparado a EAE. O aumento de 24% gerado pela rizobactéria *Bacillus* sp. não diferiu estatisticamente do controle. Curiosamente a EAE reduziu esta variável em 14% em comparação ao controle. Os diâmetros das fibras do floema foram aumentados em 58% pela fertilização com EAE e em 49% pelo tratamento *Bacillus* sp. + EAE quando confrontados com o controle. Não foram observadas diferenças estatísticas entre os tratamentos quanto ao número de elementos de vaso de metaxilema (Tabela 1).

Tabela 1. Diâmetro radicular, espessura do córtex, diâmetro do cilindro vascular, número e diâmetro de elementos de vasos do metaxilema e diâmetro das fibras do floema de mudas de *Corymbia* inoculadas com *Bacillus* sp. e fertilizadas com escória de aciaria elétrica de forno a arco (EAE).

Tratamento	Diâmetro radicular (µm)	Espessura do Córtex (µm)	Diâmetro do Cilindro Vascular (µm)	Número de Elementos de Vaso de Metaxilema	Diâmetro de Elementos de Vaso de Metaxilema (µm)	Diâmetro Fibras do Floema (µm)
Controle	1375 ± 62.1[1] c [2]	230 ± 32.0 b	821 ± 51.4 b	24.3 ± 2.3 a	50 ± 3.1 bc	12 ± 1.2 b
Bacillus sp.	1556 ±19.6 bc	279 ± 5.0 ab	995 ± 15.8 a	28.7 ± 1.9 a	62 ± 2.6 b	15 ± 0.8 b
Bacillus sp. + EAE	1804 ±134.9 a	300 ± 28.3 ab	978 ± 39.1 a	26.0 ± 3.8 a	70 ± 10.3 a	17 ± 0.7 a
EAE	1675 ± 32.5 ab	317 ± 13.6 a	920 ± 22.4 ab	30.3 ± 4.9 a	43 ± 1.2 c	18 ± 2.4 a

Nota: [1] erro padrão ($P \leq 0.05$). [2] Médias seguidas da mesma letra na coluna não diferente entre si pelo teste de Duncan ($P \leq 0.05$). Número de amostras = 12.

Figura 4. Micrografias de raízes adventícias de *Corymbia* obtidas a partir de mudas clonais inoculadas com Bacillus sp. e fertilizadas com EAE, 21 dias após a inoculação. (a1-a2) Controle (não inoculado e não fertilizado), (b1-b2) *Bacillus* sp., (c1-c2) *Bacillus* sp. + EAE, (d1-d2) EAE. Epiderme com pêlos radiculares (ep), córtex (co), cilindro vascular (vc), elementos de vaso do metaxilema (mx), fibras do floema (setas).



4.3.4 Biometria e biomassa

As variáveis biométricas tiveram influência significativa dos tratamentos avaliados. A rizobactéria *Bacillus* sp. incrementou em 10% a altura (H), em 9% o diâmetro do coleto (DC) e em 21% a área foliar (AF) das mudas em relação ao controle (Tabela 2).

A fertilização com EAE proporcionou ganhos de 19% em h e 42% em AF e a combinação *Bacillus* sp. + EAE resultou em aumentos de 16% em H e 23% em AF, ambos comparados às plantas do tratamento controle. Curiosamente, *Bacillus* sp. + EAE promoveu redução de 14% em área foliar comparado as plantas fertilizadas com EAE. Os tratamentos EAE e *Bacillus* sp. + EAE melhoraram a robustez das mudas em 17% e 19%, respectivamente, enquanto *Bacillus* sp. não diferiu do controle para este parâmetro.

Tabela 2. Efeitos da inoculação da rizobactéria *Bacillus* sp. e da fertilização com escória de aciaria elétrica de forna a arco (EAE) sobre as variáveis de crescimento em mudas de *Corymbia*.

	Controle	*Bacillus* sp.	*Bacillus* sp.+ EAE	EAE	c.v. (%)
Biometria					
H (cm)	31.76 c	34.98 b	37.00 a	37.94 a	2
DC (mm)	3.42 b	3.74 a	3.53 b	3.48 b	4
AF (cm^2)	239 c	288 b	293 b	339 a	6
IR	9.3 b	9.3 b	11.1 a	10.8 a	3
Biomassa seca					
Raiz (g)	0.877 c	1.369 b	1.444 a	1.305 b	3
Caule (g)	0.543 b	0.658 ab	0.629 ab	0.774 a	12
Folha (g)	1.471 c	1.636 bc	1.816 ab	2.214 a	6
Total (g)	2.840 c	3.670 b	3.800 b	4.320 a	3

Nota: H: altura; DC: diâmetro do coleto; AF: área foliar; IR: índice de robustez. Médias com letras iguais não diferentem pelo teste de Duncan ($P < 0.05$)

A produção de biomassa foi influenciada positivamente por todos os tratamentos testados. A rizobactéria *Bacillus* sp. promoveu aumentos de 56% em biomassa seca de raiz, 21% de caule, 11% de folhas e 28% em biomassa seca total em relação ao Controle. Quando associado, *Bacillus* sp + EAE os aumentos observados foram de 64% em biomassa de raiz, 23% em folhas e 35% em biomassa seca total em relação ao controle. Resultados significativos também foram obtidos com EAE, com aumentos de 48% em biomassa seca de raiz, 42% em caule, 50% em folhas e 51% em biomassa seca total em comparação as plantas não fertilizadas e não inoculadas (Tabela 2).

4.4 Discussão

A gestão das escórias siderúrgicas estão voltados prioritariamente para a incorporação na construção civil (TIWARI; BAJPAI; DEWANGAN, 2016), todavia a natureza química confere valor agronômico a este resíduo, em função da capacidade corretiva de acidez do solo e conteúdo de nutrientes, podendo ser utilizado como substrato alternativos para a produção de mudas (OZA *et al.*, 2018), com ganhos significativos nas variáveis biométricas e de biomassa das plantas.

Neste estudo a incorporação de 1% de EAE ao substrato utilizado na produção de mudas de *Corymbia* e a simultânea inoculação de rizobactéria promotora de crescimento de plantas *Bacillus* sp. CCMD862 gerou resultados positivos no crescimento das plantas, o que amplia as possibilidades de destinação final, agregando valor ecológico e econômico ao resíduo e colaborando para a redução do passivo ambiental nas usinas.

A fertilização das mudas de *Corymbia* com EAE não alterou a taxa de fotossíntese líquida (*A*) e a atividade da enzima Rubisco (*A/Ci*), porém elevou os níveis de carbono intercelular (*Ci*) e reduziu da eficiência intrínseca de uso da água (*A/gs*). Ao associar a fertilização com EAE e a inoculação da rizobactéria *Bacillus* sp. foram observados aumentos significativos em *A*, *A/Ci* e *A/gs* e redução em *Ci*. Estes resultados indicam que a rizobactéria otimizou etapas da fotossíntese, melhorando o rendimento de carboxilação pela enzima rubisco.

Não foram observadas alterações na condutância estomática (*gs*) em nenhum dos tratamentos avaliados. Este resultado demonstra que não houve resistência a entrada estomática de CO_2. Entretanto os níveis mais elevados de *Ci* nas plantas fertilizadas com EAE apontam para menor carboxilação via Rubisco (*A/Ci*), corroborando com a redução em *A* observada nestas plantas comparado aos tratamentos bacterizados, embora não tenha diferido do tratamento controle.

As plantas inoculadas com *Bacillus* sp. apresentaram maior taxa de transpiração (*E*). A perda de água pelas folhas ocorre à medida que o CO_2 necessário à fotossíntese é absorvido da atmosfera (TAIZ et al., 2017). Neste trabalho, o pequeno aumento de *E* nas plantas inoculadas com *Bacillus* sp. pode estar ligado à elevação da taxa de fotossíntese líquida (A), mesmo sem aumento de *gs*. O aumento em área foliar observado nesta pesquisa confere maior área fotossintetizante, uma aspecto positivo para a aquisição de carbono, porém negativo considerando as perdas por transpiração. SAMANIEGO-GÁMEZ *et al.* (2016) observaram comportamento similar em plantas de pimenta onde a inoculação de *Bacillus* promoveu aumento da assimilação de CO2 com reduzida transpiração e condutância estomática, demonstrando que a inoculação promove ajustes fisiológicos no metabolismo vegetal.

Altos valores de eficiência intrínseca do uso da água (*A/gs*) garantem maiores absorções de CO_2 com mínimas perdas de água (CHAVES, 2002) . Nesta pesquisa, a melhoria observada em *A/gs* aparentemente está atrelada a maior eficiência de carboxilação (*A/Ci*) proporcionada pela inoculação bacteriana e não ao aumento da condutância estomática (*gs*). A atividade enzimática da Rubisco depende de suprimento adequado de ATP e NADPH gerados a partir da absorção de luz pelas clorofilas, logo o aumento de fotossíntese líquida pode ser associado ao maior conteúdo de clorofilas, como observado em açaí e arroz (DE CASTRO *et al.*, 2020; RÊGO *et al.*, 2018). O isolado de *Bacillus* sp. utilizado neste estudo foi capaz de promover aumento dos teores de *Chl a, Chl a+b* e na razão *Chl a/Chl b* em mudas de *Corymbia* em estudos anteriores (dados não mostrados).

A arquitetura radicular expressa a qualidade do sistema de fixação e define a capacidade da planta de explorar o solo em busca de água e nutrientes (BERBEL *et al.*, 2020; HALING *et al.*, 2013). Todos os tratamentos testados promoveram incrementos em comprimento total, área superficial, volume, diâmetro médio e número de pontas em relação ao controle, com destaque para os efeitos de *Bacillus* sp. e *Bacillus* sp. + EAE.

A produção de fitormônios por PGPR, principalmente auxinas tem sido um mecanismo constantemente associado às melhorias radiculares em plantas inoculadas. O ácido-3-indolacético (AIA) bacteriano soma-se ao AIA endógeno, estimulando a atividade meristemática radicular, o alongamento e a diferenciação celular (LÓPEZ-BUCIO *et al.*, 2007; TRUEMAN e RICHARDSON, 2008), a emissão de raízes laterais e adventícias (DUCA *et al.*, 2014), aumentando área superficial, número de pontas e a densidade de pêlos absorventes (VALENTE LIMA *et al.*, 2021; BERBEL *et al.*, 2020; TEIXEIRA *et al.*, 2007), favorecendo a exploração de maior volume de solo, ampliando o acesso e a captação dos recursos hídricos e nutricionais, ocasionando maior acúmulo de biomassa, como observados também nesta pesquisa.

Melhorias do enraizamento e sobrevivência de mudas clonais de *Eucalyptus* propagadas por estaquia foram obtidas com a inoculação de espécies de *Bacillus, Pseudomonas, Chryseobacterium, Mucilaginibacter* e *Rhodococcus* com ganhos na emissão de raízes adventícias e raízes laterais, aumentando a superfície de absorção de nutrientes e água, otimizando a porcentagem de enraizamento e sobrevivência das mudas (NWIGWE; FOSSEY;

DE SMIDT, 2023; ZANONI DO PRADO *et al.*, 2019; GONZÁLEZ *et al.*, 2018; MAFIA *et al.*, 2007).

Todos os tratamentos testados proporcionaram aumentos no diâmetro radicular, espessura do córtex e diâmetro do cilindro vascular das raízes, produzindo plantas mais altas, com maior área foliar e acúmulo de biomassa. Resultados semelhantes foram obtidos em arroz de terras altas inoculado com PGPR, onde o aumento do diâmetro radicular foi correlacionado com maior acúmulo de biomassa seca de folhas e raízes (RÊGO *et al.*, 2014).

Os aumentos nos diâmetros dos elementos de vaso de metaxilema proporcionados por *Bacillus* sp. e *Bacillus* sp. + EAE podem representar maior eficiência na condução de água, uma adaptação importante em ambientes onde há sazonalidade marcante na distribuição hídrica ou em regiões com predomínio de solos arenosos, como as novas fronteiras da eucaliptocultura no norte e nordeste do Brasil. O aumento do diâmetro dos vasos de xilema apresentaram correlação positiva com a absorção de N e P em 53 genótipos de alfafa (PAN *et al.*, 2023), indicando que esta adaptação anatômica pode contribuir com a maior translocação de nutrientes, através do maior fluxo hídrico no interior da planta.

Os tratamentos testados induziram ganhos no diâmetro das fibras do floema secundário (Figura 4 d2), com destaque para EAE e *Bacillus* sp. + EAE. Estas fibras gelatinosas foram observadas principalmente na porção externa do floema e tem função de sustentação e algumas vezes armazenamento. Gomide (2016) investigando a anatomia de raízes de *E. grandis* em diferentes profundidades e regimes hídricos, concluiu que as fibras gelatinosas são células esclerênquimáticas, composta principalmente por celulose, auxiliando em sustentação e resistência do órgão, as quais podem atuar no armazenamento de água, visto a maior incidência em raízes mais profundas, associado a natureza hidrofílica da celulose (CALDWELL; RICHARDS, 1989).

4.5 Conclusões

Nosso estudo é inovador em investigar os efeitos da fertilização com escória de aciaria elétrica de forna à arco (EAE) combinada com a inoculação de PGPR em mudas de *Corymbia* sp.

A EAE gerou melhorias no volume radicular, no crescimento e acúmulo de biomassa da mudas e as alterações anatômicas provocadas pelo resíduo podem estar relacionadas com a robustes das plantas.

Entretanto, a sinergia entre EAE e *Bacillus* sp. favoreceu a plasticidade radicular das mudas de *Corymbia*, através de melhorias sobre a arquitetura do sistema radicular. Estas mudanças foram acompanhadas de melhorias na performance fotossintética e adaptações anatômicas, resultando em maior crescimento das plantas.

Nossos achados sugerem que a inoculação de *Bacillus* sp. em combinação com escória de aciaria de forno elétrico a arco pode ser um método potencial não apenas para a reciclagem deste resíduo, mas também para a produção segura de mudas florestais com maior robustez, contribuindo para a qualidade final e o estabelecimento das mudas em campo.

REFERÊNCIAS

ABIRI, R. *et al.* The Prospect of Physiological Events Associated with the Micropropagation of Eucalyptus sp. **Forests**, *[S. l.]*, v. 11, n. 11, p. 1211, 2020. Disponível em: https://doi.org/10.3390/f11111211

AFZAL, M. J. *et al.* Combined application of Bacillus sp. MN-54 and phosphorus improved growth and reduced lead uptake by maize in the lead-contaminated soil. **Environmental Science and Pollution Research**, *[S. l.]*, v. 27, n. 35, p. 44528–44539, 2020. Disponível em: https://doi.org/10.1007/S11356-020-10372-4/FIGURES/7. Acesso em: 10 dez. 2022.

AGARWAL, P. *et al.* PGPR-induced OsASR6 improves plant growth and yield by altering root auxin sensitivity and the xylem structure in transgenic Arabidopsis thaliana. **Journal of Plant Physiology**, *[S. l.]*, v. 240, p. 153010, 2019. Disponível em: https://doi.org/10.1016/j.jplph.2019.153010

AHEMAD, M.; KIBRET, M. Mechanisms and applications of plant growth promoting rhizobacteria: Current perspective. **Journal of King Saud University - Science**, *[S. l.]*, v. 26, n. 1, p. 1–20, 2014. Disponível em: https://doi.org/10.1016/j.jksus.2013.05.001

AHKAMI, A. H. *et al.* **Rhizosphere engineering: Enhancing sustainable plant ecosystem productivity**. *[S. l.]*: Elsevier B.V., 2017. Disponível em: https://doi.org/10.1016/j.rhisph.2017.04.012

ALEKSANDROV, VG; BLAGODYR, RN; ILEV, I. Liberation of phosphoric acid from apatite by silicate bacteria. **Mikrobiol Z**, *[S. l.]*, 1967.

ALI, M. A.; OH, J. H.; KIM, P. J. Evaluation of silicate iron slag amendment on reducing methane emission from flood water rice farming. **Agriculture, Ecosystems & Environment**, *[S. l.]*, v. 128, n. 1–2, p. 21–26, 2008. Disponível em: https://doi.org/10.1016/j.agee.2008.04.014

ALMEIDA, R. A. M. de *et al.* Slag Foaming Fundamentals - A Critical Assessment. **Materials Research**, *[S. l.]*, v. 20, n. 2, p. 474–480, 2017. Disponível em: https://doi.org/10.1590/1980-5373-mr-2016-0059

ANGULO, V. C. *et al.* Caracterización de rizobacterias promotoras de crecimiento en plántulas de Eucalyptus nitens. **Revista Argentina de Microbiología**, *[S. l.]*, v. 46, n. 4, p. 338–347, 2014 a. Disponível em: https://doi.org/10.1016/S0325-7541(14)70093-8

ANGULO, V. C. *et al.* Caracterización de rizobacterias promotoras de crecimiento en plántulas de Eucalyptus nitens. **Revista Argentina de Microbiología**, *[S. l.]*, v. 46, n. 4, p. 338–347, 2014 b. Disponível em: https://doi.org/10.1016/S0325-7541(14)70093-8

ASSIS, T. F. de. Melhoramento genético de Eucalyptus: desafios e perspectivas. **Anais do SIMPÓSIO BRASILEIRO DE SILVICULTURA**, *[S. l.]*, v. 1, n. 1, p. 127–148, 2014.

ASSIS, T. F. de; ABAD, J. I. M.; AGUIAR, A. M. Melhoramento Genético do Eucalipto. *In*: MAURO VALDIR SCHUMACHER E MÁRCIO VIERA (ORG.) (org.). **Silvicultura do Eucalipto no Brasil**. 1. ed. Santa Maria: Editora UFSM, 2015. p. 225–247. *E-book*.

AUSTRALASIAN (IRON AND STEEL) SLAG ASSOCIATION INC. **Membership Annual Survey Results, 2019 Slag Sales and Production Survey, Australia.** *[S. l.: s. n.]* Disponível em: https://www.asa-inc.org.au/files/download/d787f72a84b5512

AVELAR, M. L. M. *et al.* In vitro ESTABLISHMENT OF Eucalyptus AND Corymbia species from epicormic shoots. **Revista Árvore**, *[S. l.]*, v. 44, 2020. Disponível em: https://doi.org/10.1590/1806-908820200000027

BELLINI, C.; PACURAR, D. I.; PERRONE, I. Adventitious Roots and Lateral Roots: Similarities and Differences. **Annual Review of Plant Biology**, *[S. l.]*, v. 65, n. 1, p. 639–666, 2014. Disponível em: https://doi.org/10.1146/annurev-arplant-050213-035645

BERBEL, A. *et al.* Changes in root morphological traits in soybean co-inoculated with Bradyrhizobium spp. and Azospirillum brasilense or treated with A. brasilense exudates. **Biology and Fertility of Soils**, *[S. l.]*, v. 56, p. 537–549, 2020. Disponível em: https://doi.org/10.1007/s00374-020-01453-0. Acesso em: 19 fev. 2023.

BRANCA, T. A. *et al.* Reuse and Recycling of By-Products in the Steel Sector: Recent Achievements Paving the Way to Circular Economy and Industrial Symbiosis in Europe. **Metals**, *[S. l.]*, v. 10, n. 3, p. 345, 2020 b. Disponível em: https://doi.org/10.3390/met10030345

COSMO, B.M.N. *et al.* Influência dos metais na fotossíntese. **Revista Agronomia Brasileira**, *[S. l.]*, v. 7, 2023. Disponível em: https://doi.org/10.29372/rab202347

CAI, S. *et al.* Fertilizer Efficiency and Risk Assessment of the Utilization of AOD Slag as a Mineral Fertilizer for Alfalfa (Medicago sativa L.) and Perennial Ryegrass (Lolium perenne L.) Planting. **Sustainability 2022, Vol. 14, Page 1575**, *[S. l.]*, v. 14, n. 3, p. 1575, 2022 a. Disponível em: https://doi.org/10.3390/SU14031575. Acesso em: 14 nov. 2022.

CAI, S. *et al.* Biochemical Analysis and Toxicity Assessment of Utilization of Argon Oxygen Decarbonization Slag as a Mineral Fertilizer for Tall Fescue (Festuca arundinacea Schreb) Planting. **Sustainability**, *[S. l.]*, v. 14, n. 15, p. 9286, 2022 b. Disponível em: https://doi.org/10.3390/su14159286

CALDWELL, M. M.; RICHARDS, J. H. Hydraulic lift: water efflux from upper roots improves effectiveness of water uptake by deep roots. **Oecologia**, *[S. l.]*, n. 79, p. 1–5, 1989.

CALVO, P. *et al.* Plant growth-promoting rhizobacteria induce changes in Arabidopsis thaliana gene expression of nitrate and ammonium uptake genes. **Journal of Plant**

Interactions, *[S. l.]*, v. 14, n. 1, p. 224–231, 2019. Disponível em: https://doi.org/10.1080/17429145.2019.1602887

CALVO, P.; NELSON, L.; KLOEPPER, J. W. Agricultural uses of plant biostimulants. **Plant and Soil**, *[S. l.]*, n. 383, p. 3+41, 2014. Disponível em: https://doi.org/10.1007/s11104-014-2131-8

CARDOSO, P. H. S. *et al.* Fertilization management with sewage sludge sustains Eucalyptus productivity in Cerrado infertile soil. **Forest Ecology and Management**, *[S. l.]*, v. 519, p. 120338, 2022. Disponível em: https://doi.org/10.1016/j.foreco.2022.120338

CASTRO, G. S. A.; CRUSCIOL, C. A. C. Effects of superficial liming and silicate application on soil fertility and crop yield under rotation. **Geoderma**, *[S. l.]*, v. 195–196, p. 234–242, 2013. Disponível em: https://doi.org/10.1016/J.GEODERMA.2012.12.006

CHAND, S.; PAUL, B.; KUMAR, M. An Overview of Use of Linz-Donawitz (LD) Steel Slag in Agriculture. **Current World Environment**, *[S. l.]*, v. 10, n. 3, p. 975–984, 2015. Disponível em: https://doi.org/10.12944/CWE.10.3.29

CHAVES, M. M. How Plants Cope with Water Stress in the Field? Photosynthesis and Growth. **Annals of Botany**, *[S. l.]*, v. 89, n. 7, p. 907–916, 2002. Disponível em: https://doi.org/10.1093/aob/mcf105

CHEN, G. *et al.* Competitive mechanism and influencing factors for the simultaneous removal of Cr(III) and Zn(II) in acidic aqueous solutions using steel slag: Batch and column experiments. **Journal of Cleaner Production**, *[S. l.]*, v. 230, p. 69–79, 2019. Disponível em: https://doi.org/10.1016/j.jclepro.2019.04.402

CHIEN-YING Y. *et al*. Back to Basic Slags as a Phosphorus Source and Liming Material. *In*: **Soil Amendments for Sustainability**. 1. ed. *[S. l.]*: CRC Press, 2018. p. 14. *E-book*.

CHOUDHARY, A. *et al.* Role of plant growth-promoting Rhizobacterium in adventitious root formation. *In*: **Environmental, Physiological and Chemical Controls of Adventitious Rooting in Cuttings**. *[S. l.]*: Elsevier, 2022. p. 159–181. *E-book*. Disponível em: https://doi.org/10.1016/B978-0-323-90636-4.00007-6

CORRÊA, J. C. *et al.* Disponibilidade de metais pesados em Latossolo com aplicação superficial de escória, lama cal, lodos de esgoto e calcário. **Pesquisa Agropecuaria Brasileira**, *[S. l.]*, v. 43, n. 3, p. 411–419, 2008. Disponível em: https://doi.org/10.1590/S0100-204X2008000300017. Acesso em: 28 abr. 2021.

CUNHA, T. Q. G. da *et al.* Eucalyptus expansion in Brazil: Energy yield in new forest frontiers. **Biomass and Bioenergy**, *[S. l.]*, v. 144, p. 105900, 2021. Disponível em: https://doi.org/10.1016/j.biombioe.2020.105900

DAS, S. *et al.* Taxonomic and functional responses of soil microbial communities to slag-based fertilizer amendment in rice cropping systems. **Environment International**, *[S. l.]*, v. 127, p. 531–539, 2019. Disponível em: https://doi.org/10.1016/J.ENVINT.2019.04.012

DAS, S. *et al.* Recycling of ferrous slag in agriculture: Potentials and challenges. **Critical**

Reviews in Environmental Science and Technology, *[S. l.]*, p. 1–35, 2020. Disponível em: https://doi.org/10.1080/10643389.2020.1853458. Acesso em: 20 abr. 2021.

DE ARAUJO, M. J. *et al.* Initial productivity and genetic parameters of three corymbia species in brazil: Designing a breeding strategy. **Canadian Journal of Forest Research**, *[S. l.]*, v. 51, n. 1, p. 25–30, 2021. Disponível em: https://doi.org/10.1139/CJFR-2019-0438/ASSET/IMAGES/CJFR-2019-0438IEQ2.GIF. Acesso em: 25 abr. 2023.

DE CASTRO, G. L. S. *et al.* Açaí palm seedling growth promotion by rhizobacteria inoculation. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 51, n. 1, p. 205–216, 2020. Disponível em: https://doi.org/10.1007/s42770-019-00159-2

DEUS, A. C. F. *et al.* Effects of lime and steel slag application on soil fertility and soybean yield under a no till-system. **Soil and Tillage Research**, *[S. l.]*, v. 196, 2020. Disponível em: https://doi.org/10.1016/j.still.2019.104422

DHALI, S. *et al.* Synergistic effect of Bacillus and Rhizobium on cytological and photosynthetic performance of Macrotyloma uniflorum (Lam.) Verdc. Grown in Cr (VI) contaminated soil. **Plant Physiology and Biochemistry**, *[S. l.]*, v. 190, p. 62–69, 2022. Disponível em: https://doi.org/10.1016/J.PLAPHY.2022.08.027

DÍAZ-PILONETA, M. *et al.* Application of Steel Slag for Degraded Land Remediation. **Land**, *[S. l.]*, v. 11, n. 2, p. 224, 2022. Disponível em: https://doi.org/10.3390/LAND11020224. Acesso em: 15 fev. 2022.

DOTANIYA, M. L.; MEENA, V. D. Rhizosphere Effect on Nutrient Availability in Soil and Its Uptake by Plants: A Review. **Proc. Natl. Acad. Sci., India, Sect. B Biol. Sci.**, *[S. l.]*, n. 1, p. 1–15, 2015. Disponível em: https://doi.org/10.1007/s40011-013-0297-0

DUCA, D. *et al.* Indole-3-acetic acid in plant–microbe interactions. **Antonie van Leeuwenhoek**, *[S. l.]*, v. 106, n. 1, p. 85–125, 2014. Disponível em: https://doi.org/10.1007/s10482-013-0095-y

EGAMBERDIEVA, D. *et al.* Phytohormones and beneficial microbes: Essential components for plants to balance stress and fitness. **Frontiers in Microbiology**, *[S. l.]*, v. 8, n. OCT, p. 2104, 2017. Disponível em: https://doi.org/10.3389/FMICB.2017.02104/BIBTEX

EL-MEIHY, R. M. *et al.* Efficiency of heavy metals-tolerant plant growth promoting bacteria for alleviating heavy metals toxicity on sorghum. **Environmental and Experimental Botany**, *[S. l.]*, v. 162, 2019. Disponível em: https://doi.org/10.1016/j.envexpbot.2019.03.005

ELSAYED, A. *et al.* Synergistic effects of Azospirillum brasilense and Bacillus cereus on plant growth, biochemical attributes and molecular genetic regulation of steviol glycosides biosynthetic genes in Stevia rebaudiana. **Plant Physiology and Biochemistry**, *[S. l.]*, v. 189, p. 24–34, 2022. Disponível em: https://doi.org/10.1016/J.PLAPHY.2022.08.016

ENEBAK, S. A.; WEI, G.; KLOEPPER, J. W. Effects of Plant Growth-Promoting Rhizobacteria on Loblolly and Slash Pine Seedlings. **Forest Science**, *[S. l.]*, v. 44, n. 1, p. 139–144, 1998. Disponível em: https://doi.org/https://doi.org/10.1093/forestscience/44.1.139. Acesso em: 20 fev. 2023.

ETESAMI, H. Plant–microbe interactions in plants and stress tolerance. **Plant Life Under Changing Environment**, *[S. l.]*, p. 355–396, 2020. Disponível em: https://doi.org/10.1016/B978-0-12-818204-8.00018-7

EUROPEAN COMMISSION, D.-G. for R. and I.; KUHN, M., SPIEGEL, H., LOPEZ, A. F., REX, M., ERDMANN, R. **Sustainable agriculture using blast furnace and steel slags as liming agents**. Luxembourg: Publications Office, 2006.

FILIPPI, M. C. C. *et al.* Leaf blast (Magnaporthe oryzae) suppression and growth promotion by rhizobacteria on aerobic rice in Brazil. **Biological Control**, *[S. l.]*, v. 58, n. 2, p. 160–166, 2011. Disponível em: https://doi.org/10.1016/J.BIOCONTROL.2011.04.016

GE, Y. *et al.* Biofilm-overproducing Bacillus subtilis B12ΔYwcc decreases Cd uptake in Chinese cabbage through increasing Cd-immobilizing related gene abundance and root surface colonization. **Journal of Environmental Sciences**, *[S. l.]*, v. 120, p. 84–93, 2022. Disponível em: https://doi.org/10.1016/J.JES.2022.01.016

MAFIA, R. G. *et al.* **Root colonization and interaction among growth … R. Árvore, Viçosa-MG, v.33, n.1**. *[S. l.: s. n.]*.

GONZÁLEZ, P. *et al.* Rhizobacteria strains as promoters of rooting in hybrids of Eucalyptus nitens × Eucalyptus globulus. **Chilean journal of agricultural research**, *[S. l.]*, v. 78, n. 1, p. 3–12, 2018. Disponível em: https://doi.org/10.4067/S0718-58392018000100003

GORDON, S. A.; WEBER, R. P. Colorimetric Estimation of indoleacetic Acid. **Plant Physiology**, *[S. l.]*, v. 26, n. 1, p. 192–195, 1951. Disponível em: https://doi.org/10.1104/PP.26.1.192. Acesso em: 19 abr. 2022.

GOSWAMI, M.; DEKA, S. Isolation of a novel rhizobacteria having multiple plant growth promoting traits and antifungal activity against certain phytopathogens. **Microbiological Research**, *[S. l.]*, v. 240, p. 126516, 2020. Disponível em: https://doi.org/10.1016/j.micres.2020.126516

GROBELAK, A. *et al.* Bacterial ACC deaminase activity in promoting plant growth on areas contaminated with heavy metals. **Journal of Ecological Engineering**, *[S. l.]*, v. 19, n. 5, p. 150–157, 2018. Disponível em: https://doi.org/10.12911/22998993/89818

GUERRINI, I. A. *et al.* Composted sewage sludge and steel mill slag as potential amendments for urban soils involved in afforestation programs. **Urban Forestry & Urban Greening**, *[S. l.]*, v. 22, p. 93–104, 2017. Disponível em: https://doi.org/10.1016/J.UFUG.2017.01.015

GUO, J.; BAO, Y.; WANG, M. **Steel slag in China: Treatment, recycling, and management**. *[S. l.]*: Elsevier Ltd, 2018. Disponível em: https://doi.org/10.1016/j.wasman.2018.04.045

GWON, H. S. *et al.* Environmental risk assessment of steel-making slags and the potential use of LD slag in mitigating methane emissions and the grain arsenic level in rice (Oryza sativa L.). **Journal of Hazardous Materials**, *[S. l.]*, v. 353, 2018. Disponível em:

https://doi.org/10.1016/j.jhazmat.2018.04.023

HALING, R. E. *et al.* Root hairs improve root penetration, root–soil contact, and phosphorus acquisition in soils of different strength. **Journal of Experimental Botany**, *[S. l.]*, v. 64, n. 12, p. 3711–3721, 2013. Disponível em: https://doi.org/10.1093/jxb/ert200

HAYNES, R. J.; ZHOU, Y. F. Effect of pH and added slag on the extractability of Si in two Si-deficient sugarcane soils. **Chemosphere**, *[S. l.]*, v. 193, p. 431–437, 2018. Disponível em: https://doi.org/10.1016/J.CHEMOSPHERE.2017.10.175

HE, H. *et al.* Effects of alkaline and bioorganic amendments on cadmium, lead, zinc, and nutrient accumulation in brown rice and grain yield in acidic paddy fields contaminated with a mixture of heavy metals. **Environmental Science and Pollution Research**, *[S. l.]*, v. 23, n. 23, p. 23551–23560, 2016. Disponível em: https://doi.org/10.1007/S11356-016-7593-1/TABLES/6. Acesso em: 1 dez. 2022.

HILTUNEN, R., AND HILTUNEN, A. "Environmental aspects of the utilization of steel industry slags". *In*: THE SOUTH AFRICAN INSTITUTE OF MINING AND METALLURGY (org.). **Proceedings of VII intenational conference on molten slags, fluxes and salts**. Cape Town, South Africa: *[s. n.]*, 2004. *E-book.*

HOSSEINI, S. *et al.* Environmental Technology Reviews Production and applications of electric-arc-furnace slag as solid waste in environmental technologies: a review. *[S. l.]*, 2016. Disponível em: https://doi.org/10.1080/21622515.2016.1147615

HU, Z. H. *et al.* Combined application of arbuscular mycorrhizal fungi and steel slag improves plant growth and reduces Cd, Pb accumulation in Zea mays. **International Journal of Phytoremediation**, *[S. l.]*, v. 21, n. 9, p. 857–865, 2019. Disponível em: https://doi.org/10.1080/15226514.2019.1577355

HUSSEINY, S. *et al.* Characterization of growth promoting bacterial endophytes isolated from Artemisia annua L. **South African Journal of Botany**, *[S. l.]*, v. 143, p. 238–247, 2021. Disponível em: https://doi.org/10.1016/J.SAJB.2021.07.042. Acesso em: 19 abr. 2022.

IBGE. **PEVS 2021: Produção da Extração Vegetal e da Silvicultura**. 36. ed. Rio de Janeiro: IBGE, 2022. *E-book.*

INDÚSTRIA BRASILEIRA DE ÁRVORES. **Relatório Anual 2021**. São Paulo: Fundação Getulio Vargas (FGV), 2022. Disponível em: https://www.iba.org/datafiles/publicacoes/relatorios/relatorio-anual-iba2022-compactado.pdf

INSTITUTO AÇO BRASIL. **Relatório de Sustentabilidade 2020**2021. p. 62. Disponível em: https://www.acobrasil.org.br/relatoriodesustentabilidade/

INSTITUTO AÇO BRASIL. **Estatísticas da siderurgia**. *[s. l.]*, 2023. Disponível em: https://acobrasil.org.br/site/.

ISLAM, F. *et al.* Influence of Pseudomonas aeruginosa as PGPR on oxidative stress tolerance in wheat under Zn stress. **Ecotoxicology and Environmental Safety**, *[S. l.]*, v. 104, p. 285–293, 2014. Disponível em: https://doi.org/10.1016/j.ecoenv.2014.03.008

ISLAM, Z. *et al.* Effect of Steel Slag on Soil Fertility and Plant Growth. **Journal of Agricultural Chemistry and Environment**, *[S. l.]*, 2022. Disponível em: https://doi.org/10.4236/jacen.2022.113014. Acesso em: 9 out. 2022.

ITO, K. Steelmaking Slag for Fertilizer Usage. **NIPPON STEEL & SUMITOMO METAL TECHNICAL REPORT**, *[S. l.]*, n. 109, p. 130–136, 2015. Disponível em: https://doi.org/UDC 669 . 184 . 28 : 631 . 82 /. 86

JI, S. H.; GURURANI, M. A.; CHUN, S. C. Isolation and characterization of plant growth promoting endophytic diazotrophic bacteria from Korean rice cultivars. **Microbiological Research**, *[S. l.]*, v. 169, n. 1, p. 83–98, 2014. Disponível em: https://doi.org/10.1016/J.MICRES.2013.06.003

JING, Y.; HE, Z.; YANG, X. Role of soil rhizobacteria in phytoremediation of heavy metal contaminated soils. **Journal of Zhejiang University SCIENCE B**, *[S. l.]*, v. 8, n. 3, p. 192–207, 2007. Disponível em: https://doi.org/10.1631/jzus.2007.B0192

BOLDT, J.L. et al. Inoculation of tree species with mycorrhizal fungi fertilized with organic fertilizer and steel slag in a legal reserve area. **Agroecossistemas**, *[S. l.]*, v. 13, n. 1, p. 31–45, 2021.

KAVAMURA, V. N.; MELO, I. S. de. Effects of different osmolarities on bacterial biofilm formation. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 45, n. 2, p. 627–631, 2014. Disponível em: https://doi.org/10.1590/S1517-83822014000200034

KHODAVERDILOO, H. *et al.* Potentially toxic element contamination of arid and semi-arid soils and its phytoremediation. **Arid Land Research and Management**, *[S. l.]*, v. 34, n. 4, p. 361–391, 2020. Disponível em: https://doi.org/10.1080/15324982.2020.1746707. Acesso em: 20 mar. 2021.

KUMAR, P.; SHUKLA, S. Utilization of steel slag waste as construction material: A review. **Materials Today: Proceedings**, *[S. l.]*, v. 78, p. 145–152, 2023. Disponível em: https://doi.org/10.1016/j.matpr.2023.01.015

KURNIATI, E. O.; PEDERSON, F.; KIM, H.-J. Application of steel slags, ferronickel slags, and copper mining waste as construction materials: A review. **Resources, Conservation and Recycling**, *[S. l.]*, v. 198, p. 107175, 2023. Disponível em: https://doi.org/10.1016/j.resconrec.2023.107175

LARRABURU, E. E.; APÓSTOLO, N. M.; LLORENTE, B. E. Anatomy and morphology of photinia (Photinia × fraseri Dress) in vitro plants inoculated with rhizobacteria. **Trees**, *[S. l.]*, v. 24, n. 4, p. 635–642, 2010. Disponível em: https://doi.org/10.1007/s00468-010-0433-x

LEE, D. J. Achievements in forest tree genetic improvement in Australia and New Zealand 2: Development of Corymbia species and hybrids for plantations in eastern Australia. **http://dx.doi.org/10.1080/00049158.2007.10676256**, *[S. l.]*, v. 70, n. 1, p. 11–16, 2013. Disponível em: https://doi.org/10.1080/00049158.2007.10676256. Acesso em: 25 abr. 2023.

LI, Y. *et al.* Toxic effects of cadmium on the physiological and biochemical attributes of plants, and phytoremediation strategies: A review. **Environmental Pollution**, *[S. l.]*, v. 325, p. 121433, 2023. Disponível em: https://doi.org/10.1016/j.envpol.2023.121433

LIMA, J. V. *et al.* Rhizobacteria modify root architecture and improve nutrient uptake in oil palm seedlings despite reduced fertilizer. **Rhizosphere**, *[S. l.]*, p. 100420, 2021. Disponível em: https://doi.org/10.1016/j.rhisph.2021.100420. Acesso em: 12 ago. 2021.

LINCOLN TAIZ; EDUARDO ZEIGER; IAN MAX MØLLER; ANGUS MURPHY. **Fisiologia e desenvolvimento vegetal**. 6. ed. Porto Alegre: Artmed, 2017. *E-book*.

LÓPEZ-BUCIO, J. *et al.* Bacillus megaterium Rhizobacteria Promote Growth and Alter Root-System Architecture Through an Auxin- and Ethylene-Independent Signaling Mechanism in Arabidopsis thaliana. **Molecular Plant-Microbe Interactions®**, *[S. l.]*, v. 20, n. 2, p. 207–217, 2007. Disponível em: https://doi.org/10.1094/MPMI-20-2-0207

MA, Y. *et al.* Plant growth promoting rhizobacteria and endophytes accelerate phytoremediation of metalliferous soils. **Biotechnology Advances**, *[S. l.]*, v. 29, n. 2, p. 248–258, 2011. Disponível em: https://doi.org/10.1016/J.BIOTECHADV.2010.12.001

MA, Y. *et al.* **Beneficial role of bacterial endophytes in heavy metal phytoremediation**. *[S. l.: s. n.]* Disponível em: https://doi.org/10.1016/j.jenvman.2016.02.047

MADHAIYAN, M.; POONGUZHALI, S.; SA, T. Metal tolerating methylotrophic bacteria reduces nickel and cadmium toxicity and promotes plant growth of tomato (Lycopersicon esculentum L.). **Chemosphere**, *[S. l.]*, v. 69, n. 2, p. 220–228, 2007. Disponível em: https://doi.org/10.1016/j.chemosphere.2007.04.017

MAËLA PERON GOMIDE. **Anatomia de raízes de Eucalyptus grandis Hill ex Maiden em diferentes profundidades do solo sob um contexto de redução de chuvas**. 2016. - Universidade Estadual Paulista "Júlio de Mesquita Filho", *[s. l.]*, 2016.

MAFIA, R. G. *et al.* Rooting induction and growth of eucalyptus by rhizobacteria: Effect of addition of nutrient source and growing substrate composition. **Revista Arvore**, *[S. l.]*, v. 31, n. 4, p. 589–597, 2007 a. Disponível em: https://doi.org/10.1590/s0100-67622007000400003. Acesso em: 22 abr. 2021.

MAFIA, R. G. *et al.* Compatibility among rhizobacteria isolates and effect of mixture of isolates in inducing rooting and growth of eucalyptus clones. **Revista Arvore**, *[S. l.]*, v. 31, n. 4, p. 635–643, 2007 b. Disponível em: https://doi.org/10.1590/S0100-67622007000400008

MAFIA, R. G. *et al.* Indução do enraizamento e crescimento do eucalipto por rizobactérias: efeito da adição de fonte alimentar e da composição do substrato de enraizamento. **Revista Árvore**, *[S. l.]*, v. 31, n. 4, p. 589–597, 2007 c. Disponível em: https://doi.org/10.1590/S0100-67622007000400003

MAFIA, R. G. *et al.* Microbiolização e interação entre rizobactérias promotoras do crescimento e clones de eucalipto. **Revista Árvore**, *[S. l.]*, v. 33, n. 5, p. 789–797, 2009. Disponível em: https://doi.org/10.1590/S0100-67622009000500002

MAGALHÃES, M. O. L. *et al.* Potencial de duas espécies de eucalipto na fitoestabilização de solo contaminado com zinco. **Revista Ciência Agronômica**, *[S. l.]*, v. 42, n. 3, p. 805–812, 2011. Disponível em: https://doi.org/10.1590/S1806-66902011000300029

MARTÍNEZ-VIVEROS, O. *et al.* MECHANISMS AND PRACTICAL CONSIDERATIONS INVOLVED IN PLANT GROWTH PROMOTION BY RHIZOBACTERIA. **Journal of soil science and plant nutrition**, *[S. l.]*, v. 10, n. 3, 2010. Disponível em: https://doi.org/10.4067/S0718-95162010000100006

MARTINS, M. S. **Inovações tecnológicas da indústria 4.0: aplicações e implicações para a siderurgia brasileira**. 2019. - Universidade Federal de Uberlândia, *[s. l.]*, 2019.

MATSUMOTO, S. *et al.* Inhibition of arsenic accumulation in Japanese rice by the application of iron and silicate materials. **CATENA**, *[S. l.]*, v. 135, p. 328–335, 2015. Disponível em: https://doi.org/10.1016/J.CATENA.2015.07.004

MEENA, K. K. *et al.* Abiotic Stress Responses and Microbe-Mediated Mitigation in Plants: The Omics Strategies. **Frontiers in Plant Science**, *[S. l.]*, v. 8, 2017 a. Disponível em: https://doi.org/10.3389/fpls.2017.00172

MEENA, V. S. *et al.* Plant beneficial rhizospheric microorganism (PBRM) strategies to improve nutrients use efficiency: A review. **Ecological Engineering**, *[S. l.]*, v. 107, p. 8–32, 2017 b. Disponível em: https://doi.org/10.1016/j.ecoleng.2017.06.058

MOURÃO, M. **Introdução à siderurgia**. *[S. l.: s. n.]*. *E-book.*

NAUTIYAL, C. S. An efficient microbiological growth medium for screening phosphate solubilizing microorganisms. **FEMS Microbiology Letters**, *[S. l.]*, v. 170, n. 1, p. 265–270, 1999. Disponível em: https://doi.org/10.1111/J.1574-6968.1999.TB13383.X. Acesso em: 19 abr. 2022.

NING, D. *et al.* Impacts of steel-slag-based silicate fertilizer on soil acidity and silicon availability and metals-immobilization in a paddy soil. **PLoS ONE**, *[S. l.]*, v. 11, n. 12, 2016. Disponível em: https://doi.org/10.1371/journal.pone.0168163

NJOKU, K. L.; AKINYEDE, O. R.; OBIDI, O. F. Microbial Remediation of Heavy Metals Contaminated Media by Bacillus megaterium and Rhizopus stolonifer. **Scientific African**, *[S. l.]*, v. 10, p. e00545, 2020. Disponível em: https://doi.org/10.1016/j.sciaf.2020.e00545

NWIGWE, C.; FOSSEY, A.; DE SMIDT, O. Effects of Plant Growth Promoting Rhizospheric Bacteria (PGPR) on Survival, Growth and Rooting Architecture of Eucalyptus Hybrid Clones. **Forests**, *[S. l.]*, v. 14, n. 9, p. 1848, 2023. Disponível em: https://doi.org/10.3390/f14091848

O'CONNOR, J. *et al.* Production, characterisation, utilisation, and beneficial soil application of steel slag: A review. **Journal of Hazardous Materials**, *[S. l.]*, v. 419, p. 126478, 2021. Disponível em: https://doi.org/10.1016/J.JHAZMAT.2021.126478

OLEŃSKA, E. *et al.* Beneficial features of plant growth-promoting rhizobacteria for improving plant growth and health in challenging conditions: A methodical review. **Science of The Total Environment**, *[S. l.]*, v. 743, p. 140682, 2020. Disponível em:

https://doi.org/10.1016/J.SCITOTENV.2020.140682

OLIVEIRA, A. M. de; **Cenário brasileiro de inoculantes e aplicações de bactérias endofíticas em Corymbia**. 2023. - UNIVERSIDADE FEDERAL DOS VALES DO JEQUITINHONHA E MUCURI, *[s. l.]*, 2023.

OZA, E. F. *et al.* Aproveitamento de escória de siderurgia em substratos alternativos para produção de mudas de pimenteira Dedo-de-moça. **Revista Ceres**, *[S. l.]*, v. 65, n. 1, p. 104–109, 2018. Disponível em: https://doi.org/10.1590/0034-737x201865010014

PAN, X. *et al.* Exploring root system architecture and anatomical variability in alfalfa (Medicago sativa L.) seedlings. **BMC Plant Biology**, *[S. l.]*, v. 23, n. 1, p. 449, 2023. Disponível em: https://doi.org/10.1186/s12870-023-04469-4

PANDIT, A. *et al.* Microbial biofilms in nature: unlocking their potential for agricultural applications. **Journal of applied microbiology**, *[S. l.]*, v. 129, n. 2, p. 199–211, 2020. Disponível em: https://doi.org/10.1111/JAM.14609. Acesso em: 12 dez. 2022.

PAZ, I. C. P. *et al.* Eucalyptus growth promotion by endophytic Bacillus spp. **Genetics and Molecular Research**, *[S. l.]*, v. 11, n. 4, p. 3711–3720, 2012. Disponível em: https://doi.org/10.4238/2012.August.17.9

PERALTA, K. D. *et al.* Production of phytohormones, siderophores and population fluctuation of two root-promoting rhizobacteria in Eucalyptus globulus cuttings. **World Journal of Microbiology and Biotechnology**, *[S. l.]*, v. 28, n. 5, p. 2003–2014, 2012. Disponível em: https://doi.org/10.1007/s11274-012-1003-8. Acesso em: 22 abr. 2021.

PIATAK, N. M.; PARSONS, M. B.; SEAL, R. R. Characteristics and environmental aspects of slag: A review. **Applied Geochemistry**, *[S. l.]*, v. 57, p. 236–266, 2015. Disponível em: https://doi.org/10.1016/j.apgeochem.2014.04.009

PII, Y. *et al.* **Microbial interactions in the rhizosphere: beneficial influences of plant growth-promoting rhizobacteria on nutrient acquisition process. A review**. *[S. l.: s. n.]* Disponível em: https://doi.org/10.1007/s00374-015-0996-1

PISHCHIK, V. N. *et al.* **Mechanisms of plant and microbial adaptation to heavy metals in plant–microbial systems**. *[S. l.]*: Maik Nauka Publishing / Springer SBM, 2016. Disponível em: https://doi.org/10.1134/S0026261716030097. Acesso em: 1 abr. 2020.

PRADO, R. de M.; FERNANDES, F. M. Efeito da escória de siderurgia e calcário na disponibilidade de fósforo de um Latossolo Vermelho-Amarelo cultivado com cana-de-açúcar. **Pesquisa Agropecuária Brasileira**, *[S. l.]*, v. 36, n. 9, p. 1199–1204, 2001. Disponível em: https://doi.org/10.1590/S0100-204X2001000900014

RAASCH, L. D.; BONALDO, S. M.; OLIVEIRA, A. A. F. de. Bacillus subtilis: Enraizamento e crescimento de miniestacas de eucalipto em Sinop, norte de Mato Grosso, Brasil. **Biosci. J.**, *[S. l.]*, v. 29, n. Supplement 1, p. 1446–1457, 2013.

RADIĆ, S. *et al.* Effect of electric arc furnace slag on growth and physiology of maize (Zea mays L.). **Acta Biologica Hungarica**, *[S. l.]*, v. 64, n. 4, p. 490–499, 2013. Disponível em:

https://doi.org/10.1556/ABiol.64.2013.4.8

RAJKUMAR, M.; FREITAS, H. Influence of metal resistant-plant growth-promoting bacteria on the growth of Ricinus communis in soil contaminated with heavy metals. **Chemosphere**, *[S. l.]*, v. 71, n. 5, p. 834–842, 2008. Disponível em: https://doi.org/10.1016/j.chemosphere.2007.11.038. Acesso em: 13 nov. 2022.

RAJKUMAR, M.; MA, Y.; FREITAS, H. Improvement of Ni phytostabilization by inoculation of Ni resistant Bacillus megaterium SR28C. **Journal of Environmental Management**, *[S. l.]*, v. 128, p. 973–980, 2013. Disponível em: https://doi.org/10.1016/J.JENVMAN.2013.07.001

RÊGO, M. C. F. *et al.* Morphoanatomical and Biochemical Changes in the Roots of Rice Plants Induced by Plant Growth-Promoting Microorganisms. **Journal of Botany**, *[S. l.]*, v. 2014, p. 1–10, 2014. Disponível em: https://doi.org/10.1155/2014/818797

RÊGO, M. C. F. *et al.* The role of rhizobacteria in rice plants: Growth and mitigation of toxicity. **Journal of Integrative Agriculture**, *[S. l.]*, v. 17, n. 12, p. 2636–2647, 2018. Disponível em: https://doi.org/10.1016/S2095-3119(18)62039-8

REIS, C. A. F. *et al.* **Corymbia citriodora: estado da arte de pesquisas no Brasil**. *[s. l.]*, 2013. Disponível em: https://ainfo.cnptia.embrapa.br/digital/bitstream/item/94547/1/Doc.-255-Reis-Ainfo1.pdf. Acesso em: 10 out. 2023.

REIS, C. A. F. *et al.* **Corymbia torelliana: estado da arte de pesquisas no Brasil**. Colombo: Embrapa Florestas, 2014.

RODRÍGUEZ-DÍAZ, M. *et al*. Uma revisão sobre a taxonomia e possíveis características de triagem de rizobactérias promotoras de crescimento de plantas. *In*: AHMAD, J. PICHTEL, J., S. H. (org.). **Interações Planta-Bactérias: Estratégias e Técnicas para Promover o Crescimento das Plantas**. Weinheim, Alemanh: Wiley-VCH Verlag GmbH & Co., 2008. p. 55–80. *E-book*.

ROZEFELDS, A. C. Eucalyptus phylogeny and history: A Brief Summary. **Tasforests**, *[S. l.]*, v. 8, p. 11, 1996.

SAMANIEGO-GÁMEZ, B. Y. *et al.* Bacillus spp. inoculation improves photosystem II efficiency and enhances photosynthesis in pepper plants. **Chilean journal of agricultural research**, *[S. l.]*, v. 76, n. 4, p. 409–416, 2016. Disponível em: https://doi.org/10.4067/S0718-58392016000400003. Acesso em: 18 out. 2022.

SANTOS, L. F. dos; OLIVARES, F. L. Microbial inoculants in agriculture and its effects on plant microbiome. *In*: **Rhizosphere Engineering**. *[S. l.]*: Elsevier, 2022. p. 151–169. *E-book*. Disponível em: https://doi.org/10.1016/B978-0-323-89973-4.00016-8

SCHWYN, B.; NEILANDS, J. B. Universal chemical assay for the detection and determination of siderophores. **Analytical Biochemistry**, *[S. l.]*, v. 160, n. 1, p. 47–56, 1987. Disponível em: https://doi.org/10.1016/0003-2697(87)90612-9

SHARMA, P. Role and significance of biofilm-forming microbes in phytoremediation -A review. **Environmental Technology & Innovation**, *[S. l.]*, v. 25, p. 102182, 2022. Disponível em: https://doi.org/10.1016/J.ETI.2021.102182

SHENG, X.-F. *et al.* Characterization of heavy metal-resistant endophytic bacteria from rape (Brassica napus) roots and their potential in promoting the growth and lead accumulation of rape. **Environmental Pollution**, *[S. l.]*, v. 156, n. 3, p. 1164–1170, 2008. Disponível em: https://doi.org/10.1016/j.envpol.2008.04.007

SHEPHERD, M. *et al.* Mapping species differences for adventitious rooting in a Corymbia torelliana × Corymbia citriodora subspecies variegata hybrid. **Tree Genetics & Genomes**, *[S. l.]*, v. 4, n. 4, p. 715–725, 2008. Disponível em: https://doi.org/10.1007/s11295-008-0145-1

SHI, R. yong *et al.* Effects of biomass ash, bone meal, and alkaline slag applied alone and combined on soil acidity and wheat growth. **Journal of Soils and Sediments**, *[S. l.]*, v. 17, n. 8, p. 2116–2126, 2017. Disponível em: https://doi.org/10.1007/S11368-017-1673-9/FIGURES/6. Acesso em: 3 nov. 2022.

SHIN, M.-N. *et al.* Characterization of lead resistant endophytic Bacillus sp. MN3-4 and its potential for promoting lead accumulation in metal hyperaccumulator Alnus firma. **Journal of Hazardous Materials**, *[S. l.]*, v. 199–200, p. 314–320, 2012. Disponível em: https://doi.org/10.1016/j.jhazmat.2011.11.010

SHU, K.; SASAKI, K. Occurrence of steel converter slag and its high value-added conversion for environmental restoration in China: A review. **Journal of Cleaner Production**, *[S. l.]*, v. 373, p. 133876, 2022. Disponível em: https://doi.org/10.1016/J.JCLEPRO.2022.133876

SILVA, A. V. da *et al.* Physiological, biochemical and nutritional changes in soybean in response to application of steel slag. **Australian Journal of Crop Science**, *[S. l.]*, v. 15, n. 15(12):2021, p. 1399–1405, 2021. Disponível em: https://doi.org/10.21475/ajcs.21.15.12.p3269. Acesso em: 27 ago. 2022.

SINGH, V. *et al.* Plant Growth Promoting Rhizobacteria (PGPR): Current and Future Prospects for Development of Sustainable Agriculture. **J Microb Biochem Technol**, *[S. l.]*, v. 7, n. 2, p. 96–102, 2015. Disponível em: https://doi.org/10.4172/1948-5948.1000188

SOBRAL, M. F. *et al.* Basic slag and its effects on the concentration of nutrients and heavy metals in sugarcane. **Revista Brasileira de Engenharia Agricola e Ambiental**, *[S. l.]*, v. 15, n. 8, p. 867–872, 2011. Disponível em: https://doi.org/10.1590/S1415-43662011000800015

TEIXEIRA, D. A. *et al.* Rhizobacterial promotion of eucalypt rooting and growth. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 38, n. 1, p. 118–123, 2007. Disponível em: https://doi.org/10.1590/S1517-83822007000100025

TIWARI, M.; BAJPAI, D. S.; DEWANGAN, D. U. Steel Slag Utilization — Overview in Indian Perspective. **International Journal of Advanced Research**, *[S. l.]*, v. 4, n. 8, p. 2232–2246, 2016. Disponível em: https://doi.org/10.21474/IJAR01/1442

TRUEMAN, S. J. Cytokinin and auxin effects on survival and rooting of Eucalyptus pellita and E. grandis × E. pellita cuttings. **Rhizosphere**, *[S. l.]*, v. 6, p. 74–76, 2018. Disponível em:

https://doi.org/10.1016/J.RHISPH.2018.04.001

TRUEMAN, S. J.; RICHARDSON, D. M. Relationships between Indole-3-Butyric Acid, Photoinhibition and Adventitious Rooting of Corymbia torelliana, C. citriodora and F 1 Hybrid Cuttings. **Tree and Forestry Science and Biotechnology**, *[S. l.]*, v. 2, n. 1, p. 26–33, 2008.

TU, X. *et al.* Advantages and risks of using steel slag in preparing composts from raw organic waste. **Journal of Environmental Science and Health - Part B Pesticides, Food Contaminants, and Agricultural Wastes**, *[S. l.]*, v. 52, n. 1, p. 30–36, 2017. Disponível em: https://doi.org/10.1080/03601234.2016.1224701

VALENTE LIMA, J. *et al.* Rhizobacteria modify root architecture and improve nutrient uptake in oil palm seedlings despite reduced fertilizer. **Rhizosphere**, *[S. l.]*, v. 19, p. 100420, 2021. Disponível em: https://doi.org/10.1016/J.RHISPH.2021.100420

VEJAN, P. *et al.* Role of Plant Growth Promoting Rhizobacteria in Agricultural Sustainability—A Review. **Molecules 2016, Vol. 21, Page 573**, *[S. l.]*, v. 21, n. 5, p. 573, 2016. Disponível em: https://doi.org/10.3390/MOLECULES21050573. Acesso em: 13 fev. 2023.

VERMA, S.; KUILA, A. Bioremediation of heavy metals by microbial process. **Environmental Technology & Innovation**, *[S. l.]*, v. 14, p. 100369, 2019. Disponível em: https://doi.org/10.1016/j.eti.2019.100369

VOCCIANTE, M. *et al.* The Role of Plant Growth-Promoting Rhizobacteria (PGPR) in Mitigating Plant's Environmental Stresses. **Applied Sciences**, *[S. l.]*, v. 12, n. 3, p. 1231, 2022. Disponível em: https://doi.org/10.3390/app12031231

WANG, G. *et al.* The combined use of a plant growth promoting Bacillus sp. strain and GABA promotes the growth of rice under salt stress by regulating antioxidant enzyme system, enhancing photosynthesis and improving soil enzyme activities. **Microbiological Research**, *[S. l.]*, v. 266, p. 127225, 2023. Disponível em: https://doi.org/10.1016/J.MICRES.2022.127225

WANG, J. *et al.* Bacillus velezensis SX13 promoted cucumber growth and production by accelerating the absorption of nutrients and increasing plant photosynthetic metabolism. **Scientia Horticulturae**, *[S. l.]*, v. 301, p. 111151, 2022. Disponível em: https://doi.org/10.1016/J.SCIENTA.2022.111151

WANG, X. *et al.* Isolation of As-tolerant bacteria and their potentials of reducing As and Cd accumulation of edible tissues of vegetables in metal(loid)-contaminated soils. **Science of the Total Environment**, *[S. l.]*, v. 579, 2017. Disponível em: https://doi.org/10.1016/j.scitotenv.2016.10.239

WANG, X. *et al.* Environmental risks for application of iron and steel slags in soils in China: A review. **Pedosphere**, *[S. l.]*, v. 31, n. 1, p. 28–42, 2021. Disponível em: https://doi.org/10.1016/S1002-0160(20)60058-3

WANI, P. A.; KHAN, M. S. Bacillus species enhance growth parameters of chickpea (Cicer arietinum L.) in chromium stressed soils. **Food and Chemical Toxicology**, *[S. l.]*, v. 48, n. 11, p. 3262–3267, 2010. Disponível em: https://doi.org/10.1016/j.fct.2010.08.035

WHITE, B. *et al.* Effect of silicate slag application on wheat grown under two nitrogen rates. **Plants**, *[S. l.]*, v. 6, n. 4, 2017. Disponível em: https://doi.org/10.3390/plants6040047

WORLD STEEL ASSOCIATION. **Sustainable steel: Sustainability Indicators 2021**. Beijing: China: World Steel, 2022. Disponível em: https://worldsteel.org/publications/bookshop/sust2021/

YI, H. *et al.* An Overview of Utilization of Steel Slag. **Procedia Environmental Sciences**, *[S. l.]*, v. 16, p. 791–801, 2012. Disponível em: https://doi.org/10.1016/j.proenv.2012.10.108

YILDIRIM, I. Z.; PREZZI, M. Chemical, Mineralogical, and Morphological Properties of Steel Slag. **Advances in Civil Engineering**, *[S. l.]*, v. 2011, p. 1–13, 2011. Disponível em: https://doi.org/10.1155/2011/463638

ZANONI DO PRADO, D. *et al.* Aspergillus flavipes as a novel biostimulant for rooting-enhancement of Eucalyptus. **Journal of Cleaner Production**, *[S. l.]*, v. 234, p. 681–689, 2019. Disponível em: https://doi.org/10.1016/J.JCLEPRO.2019.06.211

5. CONSIDERAÇÕES FINAIS

Este estudo apreseta um caráter inovador ao investigar diferentes efeitos da fertilização de mudas de *Corymbia* com escória de aciaria elétrica de forna à arco (EAE) associada a inoculação de rizobactéria promotora de crescimento vegetal.

A combinação *Bacillus* sp.+ EAE como um bioinsumo, promoveu o crescimento através de um conjunto de melhorias fisiológicas, bioquímicas, anatômicas e nutricionais.

A menor acumulação de metais pesados nas raízes e a redução da translocação para os tecidos aéreos confirma a baixa mobilidade destes elementos presentes na EAE e o efeito protetor de fitotoxidez desencadeado pelo *Bacillus* sp.

A qualidade de mudas está intimamente relacionada a qualidade do sistema radicular. Este trabalho demonstrou que a EAE gera ganhos em volume de raiz e robustes das mudas e quando combinada com *Bacillus* sp., modificações mais expressivas em toda a arquitetura radicular são estabelecidas, amenizando as dificuldades de enraizamento existentes no gênero *Corymbia*.

Nossos achados indicam que a *Bacillus* sp. + EAE pode ser um método potencial não apenas para a reciclagem deste resíduo, mas também para a produção segura de mudas florestais com maior robustes, contribuindo o estabelecimento em campo e produtividade dos plantios.

A validação desta combinação em ambiente comercial compreende a fase futura desta pesquisa. Novos testes para otimizar o processo de inoculação e fertilização, com acompanhamento pós-transplantio são prioritários.

Por fim, esperamos que os resultados apresentados até o momento sejam úteis para fomentar a economia circular nas usinas siderúrgicas, como uma estratégia de gestão, além de reforçar novas parcerias público-privadas, fundamentais para a resolução de gargalos e para o avanço da ciência.

REFERÊNCIAS

ABIRI, R. *et al.* The Prospect of Physiological Events Associated with the Micropropagation of Eucalyptus sp. **Forests**, *[S. l.]*, v. 11, n. 11, p. 1211, 2020. Disponível em: https://doi.org/10.3390/f11111211

AFZAL, M. J. *et al.* Combined application of Bacillus sp. MN-54 and phosphorus improved growth and reduced lead uptake by maize in the lead-contaminated soil. **Environmental Science and Pollution Research**, *[S. l.]*, v. 27, n. 35, p. 44528–44539, 2020. Disponível em: https://doi.org/10.1007/S11356-020-10372-4/FIGURES/7. Acesso em: 10 dez. 2022.

AGARWAL, P. *et al.* PGPR-induced OsASR6 improves plant growth and yield by altering root auxin sensitivity and the xylem structure in transgenic Arabidopsis thaliana. **Journal of Plant Physiology**, *[S. l.]*, v. 240, p. 153010, 2019. Disponível em: https://doi.org/10.1016/j.jplph.2019.153010

AHEMAD, M.; KIBRET, M. Mechanisms and applications of plant growth promoting rhizobacteria: Current perspective. **Journal of King Saud University - Science**, *[S. l.]*, v. 26, n. 1, p. 1–20, 2014. Disponível em: https://doi.org/10.1016/j.jksus.2013.05.001

AHKAMI, A. H. *et al.* **Rhizosphere engineering: Enhancing sustainable plant ecosystem productivity**. *[S. l.]*: Elsevier B.V., 2017. Disponível em: https://doi.org/10.1016/j.rhisph.2017.04.012

ALEKSANDROV, VG; BLAGODYR, RN; ILEV, I. Liberation of phosphoric acid from apatite by silicate bacteria. **Mikrobiol Z**, *[S. l.]*, 1967.

ALI, M. A.; OH, J. H.; KIM, P. J. Evaluation of silicate iron slag amendment on reducing methane emission from flood water rice farming. **Agriculture, Ecosystems & Environment**, *[S. l.]*, v. 128, n. 1–2, p. 21–26, 2008. Disponível em: https://doi.org/10.1016/j.agee.2008.04.014

ALMEIDA, R. A. M. de *et al.* Slag Foaming Fundamentals - A Critical Assessment. **Materials Research**, *[S. l.]*, v. 20, n. 2, p. 474–480, 2017. Disponível em: https://doi.org/10.1590/1980-5373-mr-2016-0059

ANGULO, V. C. *et al.* Caracterización de rizobacterias promotoras de crecimiento en plántulas de Eucalyptus nitens. **Revista Argentina de Microbiología**, *[S. l.]*, v. 46, n. 4, p. 338–347, 2014 a. Disponível em: https://doi.org/10.1016/S0325-7541(14)70093-8

ASSIS, T. F. de. Melhoramento genético de Eucalyptus: desafios e perspectivas. **Anais do SIMPÓSIO BRASILEIRO DE SILVICULTURA**, *[S. l.]*, v. 1, n. 1, p. 127–148, 2014.

ASSIS, T. F. de; ABAD, J. I. M.; AGUIAR, A. M. Melhoramento Genético do Eucalipto. *In*: MAURO VALDIR SCHUMACHER E MÁRCIO VIERA (ORG.) (org.). **Silvicultura do Eucalipto no Brasil**. 1. ed. Santa Maria: Editora UFSM, 2015. p. 225–247. *E-book*.

AUSTRALASIAN (IRON AND STEEL) SLAG ASSOCIATION INC. **Membership Annual Survey Results, 2019 Slag Sales and Production Survey, Australia.** *[S. l.: s. n.]* Disponível em: https://www.asa-inc.org.au/files/download/d787f72a84b5512

AVELAR, M. L. M. *et al.* In vitro ESTABLISHMENT OF Eucalyptus AND Corymbia SPECIES FROM EPICORMIC SHOOTS. **Revista Árvore**, *[S. l.]*, v. 44, 2020. Disponível em: https://doi.org/10.1590/1806-908820200000027

BELLINI, C.; PACURAR, D. I.; PERRONE, I. Adventitious Roots and Lateral Roots: Similarities and Differences. **Annual Review of Plant Biology**, *[S. l.]*, v. 65, n. 1, p. 639–666, 2014. Disponível em: https://doi.org/10.1146/annurev-arplant-050213-035645

BERBEL, A. *et al.* Changes in root morphological traits in soybean co-inoculated with Bradyrhizobium spp. and Azospirillum brasilense or treated with A. brasilense exudates.

Biology and Fertility of Soils, *[S. l.]*, v. 56, p. 537–549, 2020. Disponível em: https://doi.org/10.1007/s00374-020-01453-0. Acesso em: 19 fev. 2023.

BRANCA, T. A. *et al.* Reuse and Recycling of By-Products in the Steel Sector: Recent Achievements Paving the Way to Circular Economy and Industrial Symbiosis in Europe. **Metals**, *[S. l.]*, v. 10, n. 3, p. 345, 2020 a. Disponível em: https://doi.org/10.3390/met10030345

BRUNO MARCOS NUNES COSMO *et al.* Influência dos metais na fotossíntese. **Revista Agronomia Brasileira**, *[S. l.]*, v. 7, 2023. Disponível em: https://doi.org/10.29372/rab202347

CAI, S. *et al.* Fertilizer Efficiency and Risk Assessment of the Utilization of AOD Slag as a Mineral Fertilizer for Alfalfa (Medicago sativa L.) and Perennial Ryegrass (Lolium perenne L.) Planting. **Sustainability 2022, Vol. 14, Page 1575**, *[S. l.]*, v. 14, n. 3, p. 1575, 2022 a. Disponível em: https://doi.org/10.3390/SU14031575. Acesso em: 14 nov. 2022.

CAI, S. *et al.* Biochemical Analysis and Toxicity Assessment of Utilization of Argon Oxygen Decarbonization Slag as a Mineral Fertilizer for Tall Fescue (Festuca arundinacea Schreb) Planting. **Sustainability**, *[S. l.]*, v. 14, n. 15, p. 9286, 2022 b. Disponível em: https://doi.org/10.3390/su14159286

CALDWELL, M. M.; RICHARDS, J. H. Hydraulic lift: water efflux from upper roots improves effectiveness of water uptake by deep roots. **Oecologia**, *[S. l.]*, n. 79, p. 1–5, 1989.

CALVO, P. *et al.* Plant growth-promoting rhizobacteria induce changes in Arabidopsis thaliana gene expression of nitrate and ammonium uptake genes. **Journal of Plant Interactions**, *[S. l.]*, v. 14, n. 1, p. 224–231, 2019. Disponível em: https://doi.org/10.1080/17429145.2019.1602887
CALVO, P.; NELSON, L.; KLOEPPER, J. W. Agricultural uses of plant biostimulants. **Plant and Soil**, *[S. l.]*, n. 383, p. 3+41, 2014. Disponível em: https://doi.org/10.1007/s11104-014-2131-8

CARDOSO, P. H. S. *et al.* Fertilization management with sewage sludge sustains Eucalyptus productivity in Cerrado infertile soil. **Forest Ecology and Management**, *[S. l.]*, v. 519, p. 120338, 2022. Disponível em: https://doi.org/10.1016/j.foreco.2022.120338

CASTRO, G. S. A.; CRUSCIOL, C. A. C. Effects of superficial liming and silicate application on soil fertility and crop yield under rotation. **Geoderma**, *[S. l.]*, v. 195–196, p. 234–242, 2013. Disponível em: https://doi.org/10.1016/J.GEODERMA.2012.12.006

CHAND, S.; PAUL, B.; KUMAR, M. An Overview of Use of Linz-Donawitz (LD) Steel Slag in Agriculture. **Current World Environment**, *[S. l.]*, v. 10, n. 3, p. 975–984, 2015. Disponível em: https://doi.org/10.12944/CWE.10.3.29

CHAVES, M. M. How Plants Cope with Water Stress in the Field? Photosynthesis and Growth. **Annals of Botany**, *[S. l.]*, v. 89, n. 7, p. 907–916, 2002. Disponível em: https://doi.org/10.1093/aob/mcf105

CHEN, G. *et al.* Competitive mechanism and influencing factors for the simultaneous removal of Cr(III) and Zn(II) in acidic aqueous solutions using steel slag: Batch and column experiments. **Journal of Cleaner Production**, *[S. l.]*, v. 230, p. 69–79, 2019. Disponível em: https://doi.org/10.1016/j.jclepro.2019.04.402

YANG, C.Y. *et al*. B. Back to Basic Slags as a Phosphorus Source and Liming Material. *In*: **Soil Amendments for Sustainability**. 1. ed. *[S. l.]*: CRC Press, 2018. p. 14. *E-book*.

CHOUDHARY, A. *et al.* Role of plant growth-promoting Rhizobacterium in adventitious root formation. *In*: **Environmental, Physiological and Chemical Controls of Adventitious Rooting in Cuttings**. *[S. l.]*: Elsevier, 2022. p. 159–181. *E-book*. Disponível em: https://doi.org/10.1016/B978-0-323-90636-4.00007-6

CORRÊA, J. C. *et al.* Disponibilidade de metais pesados em Latossolo com aplicação superficial de escória, lama cal, lodos de esgoto e calcário. **Pesquisa Agropecuaria Brasileira**, *[S. l.]*, v. 43, n. 3, p. 411–419, 2008. Disponível em: https://doi.org/10.1590/S0100-204X2008000300017. Acesso em: 28 abr. 2021.

CUNHA, T. Q. G. da *et al.* Eucalyptus expansion in Brazil: Energy yield in new forest frontiers. **Biomass and Bioenergy**, *[S. l.]*, v. 144, p. 105900, 2021. Disponível em: https://doi.org/10.1016/j.biombioe.2020.105900

DAS, S. *et al.* Taxonomic and functional responses of soil microbial communities to slag-based fertilizer amendment in rice cropping systems. **Environment International**, *[S. l.]*, v. 127, p. 531–539, 2019. Disponível em: https://doi.org/10.1016/J.ENVINT.2019.04.012

DAS, S. *et al.* Recycling of ferrous slag in agriculture: Potentials and challenges. **Critical Reviews in Environmental Science and Technology**, *[S. l.]*, p. 1–35, 2020. Disponível em: https://doi.org/10.1080/10643389.2020.1853458. Acesso em: 20 abr. 2021.

DE ARAUJO, M. J. *et al.* Initial productivity and genetic parameters of three corymbia species in brazil: Designing a breeding strategy. **Canadian Journal of Forest Research**, *[S. l.]*, v. 51, n. 1, p. 25–30, 2021. Disponível em: https://doi.org/10.1139/CJFR-2019-0438/ASSET/IMAGES/CJFR-2019-0438IEQ2.GIF. Acesso em: 25 abr. 2023.

DE CASTRO, G. L. S. *et al.* Açaí palm seedling growth promotion by rhizobacteria inoculation. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 51, n. 1, p. 205–216, 2020. Disponível em: https://doi.org/10.1007/s42770-019-00159-2

DEUS, A. C. F. *et al.* Effects of lime and steel slag application on soil fertility and soybean yield under a no till-system. **Soil and Tillage Research**, *[S. l.]*, v. 196, 2020. Disponível em: https://doi.org/10.1016/j.still.2019.104422

DHALI, S. *et al.* Synergistic effect of Bacillus and Rhizobium on cytological and photosynthetic performance of Macrotyloma uniflorum (Lam.) Verdc. Grown in Cr (VI) contaminated soil. **Plant Physiology and Biochemistry**, *[S. l.]*, v. 190, p. 62–69, 2022. Disponível em: https://doi.org/10.1016/J.PLAPHY.2022.08.027

DÍAZ-PILONETA, M. *et al.* Application of Steel Slag for Degraded Land Remediation. **Land**, *[S. l.]*, v. 11, n. 2, p. 224, 2022. Disponível em: https://doi.org/10.3390/LAND11020224. Acesso em: 15 fev. 2022.

DOTANIYA, M. L.; MEENA, V. D. Rhizosphere Effect on Nutrient Availability in Soil and Its Uptake by Plants: A Review. **Proc. Natl. Acad. Sci., India, Sect. B Biol. Sci.**, *[S. l.]*, n. 1, p. 1–15, 2015. Disponível em: https://doi.org/10.1007/s40011-013-0297-0

DUCA, D. *et al.* Indole-3-acetic acid in plant–microbe interactions. **Antonie van Leeuwenhoek**, *[S. l.]*, v. 106, n. 1, p. 85–125, 2014. Disponível em: https://doi.org/10.1007/s10482-013-0095-y

EGAMBERDIEVA, D. *et al.* Phytohormones and beneficial microbes: Essential components for plants to balance stress and fitness. **Frontiers in Microbiology**, *[S. l.]*, v. 8, n. OCT, p. 2104, 2017. Disponível em: https://doi.org/10.3389/FMICB.2017.02104/BIBTEX

EL-MEIHY, R. M. *et al.* Efficiency of heavy metals-tolerant plant growth promoting bacteria for alleviating heavy metals toxicity on sorghum. **Environmental and Experimental Botany**, *[S. l.]*, v. 162, 2019. Disponível em: https://doi.org/10.1016/j.envexpbot.2019.03.005

ELSAYED, A. *et al.* Synergistic effects of Azospirillum brasilense and Bacillus cereus on plant growth, biochemical attributes and molecular genetic regulation of steviol glycosides biosynthetic genes in Stevia rebaudiana. **Plant Physiology and Biochemistry**, *[S. l.]*, v. 189, p. 24–34, 2022. Disponível em: https://doi.org/10.1016/J.PLAPHY.2022.08.016

ENEBAK, S. A.; WEI, G.; KLOEPPER, J. W. Effects of Plant Growth-Promoting Rhizobacteria on Loblolly and Slash Pine Seedlings. **Forest Science**, *[S. l.]*, v. 44, n. 1, p. 139–144, 1998. Disponível em: https://doi.org/https://doi.org/10.1093/forestscience/44.1.139. Acesso em: 20 fev. 2023.

ETESAMI, H. Plant–microbe interactions in plants and stress tolerance. **Plant Life Under Changing Environment**, *[S. l.]*, p. 355–396, 2020. Disponível em: https://doi.org/10.1016/B978-0-12-818204-8.00018-7

EUROPEAN COMMISSION, D.-G. for R. and I.; KUHN, M., SPIEGEL, H., LOPEZ, A. F., REX, M., ERDMANN, R. **Sustainable agriculture using blast furnace and steel slags as liming agents**. Luxembourg: Publications Office, 2006.

FILIPPI, M. C. C. *et al.* Leaf blast (Magnaporthe oryzae) suppression and growth promotion by rhizobacteria on aerobic rice in Brazil. **Biological Control**, *[S. l.]*, v. 58, n. 2, p. 160–166, 2011. Disponível em: https://doi.org/10.1016/J.BIOCONTROL.2011.04.016

GE, Y. *et al.* Biofilm-overproducing Bacillus subtilis B12ΔYwcc decreases Cd uptake in Chinese cabbage through increasing Cd-immobilizing related gene abundance and root surface colonization. **Journal of Environmental Sciences**, *[S. l.]*, v. 120, p. 84–93, 2022. Disponível em: https://doi.org/10.1016/J.JES.2022.01.016

GONÇALVES MAFIA, R. *et al.* **Root colonization and interaction among growth … R. Árvore, Viçosa-MG, v.33, n.1**. *[S. l.: s. n.]*.

GONZÁLEZ, P. *et al.* Rhizobacteria strains as promoters of rooting in hybrids of Eucalyptus nitens × Eucalyptus globulus. **Chilean journal of agricultural research**, *[S. l.]*, v. 78, n. 1, p. 3–12, 2018. Disponível em: https://doi.org/10.4067/S0718-58392018000100003

GORDON, S. A.; WEBER, R. P. Colorimetric Estimation of indoleacetic Acid. **Plant Physiology**, *[S. l.]*, v. 26, n. 1, p. 192–195, 1951. Disponível em: https://doi.org/10.1104/PP.26.1.192. Acesso em: 19 abr. 2022.

GOSWAMI, M.; DEKA, S. Isolation of a novel rhizobacteria having multiple plant growth promoting traits and antifungal activity against certain phytopathogens. **Microbiological Research**, *[S. l.]*, v. 240, p. 126516, 2020. Disponível em: https://doi.org/10.1016/j.micres.2020.126516

GROBELAK, A. *et al.* Bacterial ACC deaminase activity in promoting plant growth on areas contaminated with heavy metals. **Journal of Ecological Engineering**, *[S. l.]*, v. 19, n. 5, p. 150–157, 2018. Disponível em: https://doi.org/10.12911/22998993/89818

GUERRINI, I. A. *et al.* Composted sewage sludge and steel mill slag as potential amendments for urban soils involved in afforestation programs. **Urban Forestry & Urban Greening**, *[S. l.]*, v. 22, p. 93–104, 2017. Disponível em: https://doi.org/10.1016/J.UFUG.2017.01.015

GUO, J.; BAO, Y.; WANG, M. **Steel slag in China: Treatment, recycling, and management**. *[S. l.]*: Elsevier Ltd, 2018. Disponível em: https://doi.org/10.1016/j.wasman.2018.04.045

GWON, H. S. *et al.* Environmental risk assessment of steel-making slags and the potential use of LD slag in mitigating methane emissions and the grain arsenic level in rice (Oryza sativa L.). **Journal of Hazardous Materials**, *[S. l.]*, v. 353, 2018. Disponível em: https://doi.org/10.1016/j.jhazmat.2018.04.023

HALING, R. E. *et al.* Root hairs improve root penetration, root–soil contact, and phosphorus acquisition in soils of different strength. **Journal of Experimental Botany**, *[S. l.]*, v. 64, n. 12, p. 3711–3721, 2013. Disponível em: https://doi.org/10.1093/jxb/ert200

HAYNES, R. J.; ZHOU, Y. F. Effect of pH and added slag on the extractability of Si in two Si-deficient sugarcane soils. **Chemosphere**, *[S. l.]*, v. 193, p. 431–437, 2018. Disponível em: https://doi.org/10.1016/J.CHEMOSPHERE.2017.10.175

HE, H. *et al.* Effects of alkaline and bioorganic amendments on cadmium, lead, zinc, and nutrient accumulation in brown rice and grain yield in acidic paddy fields contaminated with a mixture of heavy metals. **Environmental Science and Pollution Research**, *[S. l.]*, v. 23, n. 23, p. 23551–23560, 2016. Disponível em: https://doi.org/10.1007/S11356-016-7593-1/TABLES/6. Acesso em: 1 dez. 2022.

HILTUNEN, R., AND HILTUNEN, A. "Environmental aspects of the utilization of steel industry slags". *In*: THE SOUTH AFRICAN INSTITUTE OF MINING AND METALLURGY (org.). **Proceedings of VII intenational conference on molten slags, fluxes and salts**. Cape Town, South Africa: *[s. n.]*, 2004. *E-book.*

HOSSEINI, S. *et al.* Environmental Technology Reviews Production and applications of electric-arc-furnace slag as solid waste in environmental technologies: a review. *[S. l.]*, 2016. Disponível em: https://doi.org/10.1080/21622515.2016.1147615

HU, Z. H. *et al.* Combined application of arbuscular mycorrhizal fungi and steel slag improves plant growth and reduces Cd, Pb accumulation in Zea mays. **International Journal of Phytoremediation**, *[S. l.]*, v. 21, n. 9, p. 857–865, 2019. Disponível em: https://doi.org/10.1080/15226514.2019.1577355

HUSSEINY, S. *et al.* Characterization of growth promoting bacterial endophytes isolated from Artemisia annua L. **South African Journal of Botany**, *[S. l.]*, v. 143, p. 238–247, 2021. Disponível em: https://doi.org/10.1016/J.SAJB.2021.07.042. Acesso em: 19 abr. 2022.

IBGE. **PEVS 2021: Produção da Extração Vegetal e da Silvicultura**. 36. ed. Rio de Janeiro: IBGE, 2022. *E-book*.

INDÚSTRIA BRASILEIRA DE ÁRVORES. **Relatório Anual 2021**. São Paulo: Fundação Getulio Vargas (FGV), 2022. Disponível em: https://www.iba.org/datafiles/publicacoes/relatorios/relatorio-anual-iba2022-compactado.pdf

INSTITUTO AÇO BRASIL. **Relatório de Sustentabilidade 2021**. p. 62. Disponível em: https://www.acobrasil.org.br/relatoriodesustentabilidade/

INSTITUTO AÇO BRASIL. **Estatísticas da siderurgia**. *[s. l.]*, 2023. Disponível em: https://acobrasil.org.br/site/.

ISLAM, F. *et al.* Influence of Pseudomonas aeruginosa as PGPR on oxidative stress tolerance in wheat under Zn stress. **Ecotoxicology and Environmental Safety**, *[S. l.]*, v. 104, p. 285–293, 2014. Disponível em: https://doi.org/10.1016/j.ecoenv.2014.03.008

ISLAM, Z. *et al.* Effect of Steel Slag on Soil Fertility and Plant Growth. **Journal of Agricultural Chemistry and Environment**, *[S. l.]*, 2022. Disponível em: https://doi.org/10.4236/jacen.2022.113014. Acesso em: 9 out. 2022.

ITO, K. Steelmaking Slag for Fertilizer Usage. **NIPPON STEEL & SUMITOMO METAL TECHNICAL REPORT**, *[S. l.]*, n. 109, p. 130–136, 2015. Disponível em: https://doi.org/UDC 669 . 184 . 28 : 631 . 82 /. 86

JI, S. H.; GURURANI, M. A.; CHUN, S. C. Isolation and characterization of plant growth promoting endophytic diazotrophic bacteria from Korean rice cultivars. **Microbiological Research**, *[S. l.]*, v. 169, n. 1, p. 83–98, 2014. Disponível em: https://doi.org/10.1016/J.MICRES.2013.06.003

JING, Y.; HE, Z.; YANG, X. Role of soil rhizobacteria in phytoremediation of heavy metal contaminated soils. **Journal of Zhejiang University SCIENCE B**, *[S. l.]*, v. 8, n. 3, p. 192–207, 2007. Disponível em: https://doi.org/10.1631/jzus.2007.B0192

BOLDT, J.L. *et al*. Inoculation of tree species with mycorrhizal fungi fertilized with organic fertilizer and steel slag in a legal reserve area. **Agroecossistemas**, *[S. l.]*, v. 13, n. 1, p. 31–45, 2021.

KAVAMURA, V. N.; MELO, I. S. de. Effects of different osmolarities on bacterial biofilm formation. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 45, n. 2, p. 627–631, 2014. Disponível em: https://doi.org/10.1590/S1517-83822014000200034

KHODAVERDILOO, H. *et al.* Potentially toxic element contamination of arid and semi-arid soils and its phytoremediation. **Arid Land Research and Management**, *[S. l.]*, v. 34, n. 4, p. 361–391, 2020. Disponível em: https://doi.org/10.1080/15324982.2020.1746707. Acesso em: 20 mar. 2021.

KUMAR, P.; SHUKLA, S. Utilization of steel slag waste as construction material: A review. **Materials Today: Proceedings**, *[S. l.]*, v. 78, p. 145–152, 2023. Disponível em: https://doi.org/10.1016/j.matpr.2023.01.015

KURNIATI, E. O.; PEDERSON, F.; KIM, H.-J. Application of steel slags, ferronickel slags, and copper mining waste as construction materials: A review. **Resources, Conservation and Recycling**, *[S. l.]*, v. 198, p. 107175, 2023. Disponível em: https://doi.org/10.1016/j.resconrec.2023.107175

LARRABURU, E. E.; APÓSTOLO, N. M.; LLORENTE, B. E. Anatomy and morphology of photinia (Photinia × fraseri Dress) in vitro plants inoculated with rhizobacteria. **Trees**, *[S. l.]*, v. 24, n. 4, p. 635–642, 2010. Disponível em: https://doi.org/10.1007/s00468-010-0433-x

LEE, D. J. Achievements in forest tree genetic improvement in Australia and New Zealand 2: Development of Corymbia species and hybrids for plantations in eastern Australia. **http://dx.doi.org/10.1080/00049158.2007.10676256**, *[S. l.]*, v. 70, n. 1, p. 11–16, 2013. Disponível em: https://doi.org/10.1080/00049158.2007.10676256. Acesso em: 25 abr. 2023.

LI, Y. *et al.* Toxic effects of cadmium on the physiological and biochemical attributes of plants, and phytoremediation strategies: A review. **Environmental Pollution**, *[S. l.]*, v. 325, p. 121433, 2023. Disponível em: https://doi.org/10.1016/j.envpol.2023.121433

LIMA, J. V. *et al.* Rhizobacteria modify root architecture and improve nutrient uptake in oil palm seedlings despite reduced fertilizer. **Rhizosphere**, *[S. l.]*, p. 100420, 2021. Disponível em: https://doi.org/10.1016/j.rhisph.2021.100420. Acesso em: 12 ago. 2021.

LINCOLN TAIZ; EDUARDO ZEIGER; IAN MAX MØLLER; ANGUS MURPHY. **Fisiologia e desenvolvimento vegetal**. 6. ed. Porto Alegre: Artmed, 2017. *E-book*.

LÓPEZ-BUCIO, J. *et al.* Bacillus megaterium Rhizobacteria Promote Growth and Alter Root-System Architecture Through an Auxin- and Ethylene-Independent Signaling Mechanism in Arabidopsis thaliana. **Molecular Plant-Microbe Interactions®**, *[S. l.]*, v. 20, n. 2, p. 207–217, 2007. Disponível em: https://doi.org/10.1094/MPMI-20-2-0207

MA, Y. *et al.* Plant growth promoting rhizobacteria and endophytes accelerate phytoremediation of metalliferous soils. **Biotechnology Advances**, *[S. l.]*, v. 29, n. 2, p. 248–258, 2011. Disponível em: https://doi.org/10.1016/J.BIOTECHADV.2010.12.001

MA, Y. *et al.* **Beneficial role of bacterial endophytes in heavy metal phytoremediation**. *[S. l.: s. n.]* Disponível em: https://doi.org/10.1016/j.jenvman.2016.02.047

MADHAIYAN, M.; POONGUZHALI, S.; SA, T. Metal tolerating methylotrophic bacteria reduces nickel and cadmium toxicity and promotes plant growth of tomato (Lycopersicon esculentum L.). **Chemosphere**, *[S. l.]*, v. 69, n. 2, p. 220–228, 2007. Disponível em:

https://doi.org/10.1016/j.chemosphere.2007.04.017

MAËLA PERON GOMIDE. **ANATOMIA DE RAÍZES DE Eucalyptus grandis Hill ex Maiden EM DIFERENTES PROFUNDIDADES DO SOLO SOB UM CONTEXTO DE REDUÇÃO DE CHUVAS**. 2016. - Universidade Estadual Paulista “Júlio de Mesquita Filho”, *[s. l.]*, 2016.

MAFIA, R. G. *et al.* Rooting induction and growth of eucalyptus by rhizobacteria: Effect of addition of nutrient source and growing substrate composition. **Revista Arvore**, *[S. l.]*, v. 31, n. 4, p. 589–597, 2007 a. Disponível em: https://doi.org/10.1590/s0100-67622007000400003. Acesso em: 22 abr. 2021.

MAFIA, R. G. *et al.* Compatibility among rhizobacteria isolates and effect of mixture of isolates in inducing rooting and growth of eucalyptus clones. **Revista Arvore**, *[S. l.]*, v. 31, n. 4, p. 635–643, 2007 b. Disponível em: https://doi.org/10.1590/S0100-67622007000400008

MAFIA, R. G. *et al.* Indução do enraizamento e crescimento do eucalipto por rizobactérias: efeito da adição de fonte alimentar e da composição do substrato de enraizamento. **Revista Árvore**, *[S. l.]*, v. 31, n. 4, p. 589–597, 2007 c. Disponível em: https://doi.org/10.1590/S0100-67622007000400003

MAFIA, R. G. *et al.* Microbiolização e interação entre rizobactérias promotoras do crescimento e clones de eucalipto. **Revista Árvore**, *[S. l.]*, v. 33, n. 5, p. 789–797, 2009. Disponível em: https://doi.org/10.1590/S0100-67622009000500002

MAGALHÃES, M. O. L. *et al.* Potencial de duas espécies de eucalipto na fitoestabilização de solo contaminado com zinco. **Revista Ciência Agronômica**, *[S. l.]*, v. 42, n. 3, p. 805–812, 2011. Disponível em: https://doi.org/10.1590/S1806-66902011000300029

MARTÍNEZ-VIVEROS, O. *et al.* MECHANISMS AND PRACTICAL CONSIDERATIONS INVOLVED IN PLANT GROWTH PROMOTION BY RHIZOBACTERIA. **Journal of soil science and plant nutrition**, *[S. l.]*, v. 10, n. 3, 2010. Disponível em: https://doi.org/10.4067/S0718-95162010000100006

MARTINS, M. S. **INOVAÇÕES TECNOLÓGICAS DA INDÚSTRIA 4.0: APLICAÇÕES E IMPLICAÇÕES PARA A SIDERURGIA BRASILEIRA**. 2019. - Universidade Federal de Uberlândia, *[s. l.]*, 2019.

MATSUMOTO, S. *et al.* Inhibition of arsenic accumulation in Japanese rice by the application of iron and silicate materials. **CATENA**, *[S. l.]*, v. 135, p. 328–335, 2015. Disponível em: https://doi.org/10.1016/J.CATENA.2015.07.004

MEENA, K. K. *et al.* Abiotic Stress Responses and Microbe-Mediated Mitigation in Plants: The Omics Strategies. **Frontiers in Plant Science**, *[S. l.]*, v. 8, 2017 a. Disponível em: https://doi.org/10.3389/fpls.2017.00172

MEENA, V. S. *et al.* Plant beneficial rhizospheric microorganism (PBRM) strategies to improve nutrients use efficiency: A review. **Ecological Engineering**, *[S. l.]*, v. 107, p. 8–32, 2017 b. Disponível em: https://doi.org/10.1016/j.ecoleng.2017.06.058

MOURÃO, M. **Introdução à siderurgia**. *[S. l.: s. n.]*. *E-book*.

NAUTIYAL, C. S. An efficient microbiological growth medium for screening phosphate solubilizing microorganisms. **FEMS Microbiology Letters**, *[S. l.]*, v. 170, n. 1, p. 265–270, 1999. Disponível em: https://doi.org/10.1111/J.1574-6968.1999.TB13383.X. Acesso em: 19 abr. 2022.

NING, D. *et al.* Impacts of steel-slag-based silicate fertilizer on soil acidity and silicon availability and metals-immobilization in a paddy soil. **PLoS ONE**, *[S. l.]*, v. 11, n. 12, 2016. Disponível em: https://doi.org/10.1371/journal.pone.0168163

NJOKU, K. L.; AKINYEDE, O. R.; OBIDI, O. F. Microbial Remediation of Heavy Metals Contaminated Media by Bacillus megaterium and Rhizopus stolonifer. **Scientific African**, *[S. l.]*, v. 10, p. e00545, 2020. Disponível em: https://doi.org/10.1016/j.sciaf.2020.e00545

NWIGWE, C.; FOSSEY, A.; DE SMIDT, O. Effects of Plant Growth Promoting Rhizospheric Bacteria (PGPR) on Survival, Growth and Rooting Architecture of Eucalyptus Hybrid Clones. **Forests**, *[S. l.]*, v. 14, n. 9, p. 1848, 2023. Disponível em: https://doi.org/10.3390/f14091848

O'CONNOR, J. *et al.* Production, characterisation, utilisation, and beneficial soil application of steel slag: A review. **Journal of Hazardous Materials**, *[S. l.]*, v. 419, p. 126478, 2021. Disponível em: https://doi.org/10.1016/J.JHAZMAT.2021.126478

OLEŃSKA, E. *et al.* Beneficial features of plant growth-promoting rhizobacteria for improving plant growth and health in challenging conditions: A methodical review. **Science of The Total Environment**, *[S. l.]*, v. 743, p. 140682, 2020. Disponível em: https://doi.org/10.1016/J.SCITOTENV.2020.140682

OLIVEIRA, A. M. de. **CENÁRIO BRASILEIRO DE INOCULANTES E APLICAÇÕES DE BACTÉRIAS ENDOFÍTICAS EM Corymbia**. 2023. - UNIVERSIDADE FEDERAL DOS VALES DO JEQUITINHONHA E MUCURI, *[s. l.]*, 2023.

OZA, E. F. *et al.* Aproveitamento de escória de siderurgia em substratos alternativos para produção de mudas de pimenteira Dedo-de-moça. **Revista Ceres**, *[S. l.]*, v. 65, n. 1, p. 104–109, 2018. Disponível em: https://doi.org/10.1590/0034-737x201865010014

PAN, X. *et al.* Exploring root system architecture and anatomical variability in alfalfa (Medicago sativa L.) seedlings. **BMC Plant Biology**, *[S. l.]*, v. 23, n. 1, p. 449, 2023. Disponível em: https://doi.org/10.1186/s12870-023-04469-4

PANDIT, A. *et al.* Microbial biofilms in nature: unlocking their potential for agricultural applications. **Journal of applied microbiology**, *[S. l.]*, v. 129, n. 2, p. 199–211, 2020. Disponível em: https://doi.org/10.1111/JAM.14609. Acesso em: 12 dez. 2022.

PAZ, I. C. P. *et al.* Eucalyptus growth promotion by endophytic Bacillus spp. **Genetics and Molecular Research**, *[S. l.]*, v. 11, n. 4, p. 3711–3720, 2012. Disponível em: https://doi.org/10.4238/2012.August.17.9

PERALTA, K. D. *et al.* Production of phytohormones, siderophores and population fluctuation of two root-promoting rhizobacteria in Eucalyptus globulus cuttings. **World**

Journal of Microbiology and Biotechnology, *[S. l.]*, v. 28, n. 5, p. 2003–2014, 2012. Disponível em: https://doi.org/10.1007/s11274-012-1003-8. Acesso em: 22 abr. 2021.

PIATAK, N. M.; PARSONS, M. B.; SEAL, R. R. Characteristics and environmental aspects of slag: A review. **Applied Geochemistry**, *[S. l.]*, v. 57, p. 236–266, 2015. Disponível em: https://doi.org/10.1016/j.apgeochem.2014.04.009

PII, Y. *et al.* **Microbial interactions in the rhizosphere: beneficial influences of plant growth-promoting rhizobacteria on nutrient acquisition process. A review**. *[S. l.: s. n.]* Disponível em: https://doi.org/10.1007/s00374-015-0996-1

PISHCHIK, V. N. *et al.* **Mechanisms of plant and microbial adaptation to heavy metals in plant–microbial systems**. *[S. l.]*: Maik Nauka Publishing / Springer SBM, 2016. Disponível em: https://doi.org/10.1134/S0026261716030097. Acesso em: 1 abr. 2020.

PRADO, R. de M.; FERNANDES, F. M. Efeito da escória de siderurgia e calcário na disponibilidade de fósforo de um Latossolo Vermelho-Amarelo cultivado com cana-de-açúcar. **Pesquisa Agropecuária Brasileira**, *[S. l.]*, v. 36, n. 9, p. 1199–1204, 2001. Disponível em: https://doi.org/10.1590/S0100-204X2001000900014

RAASCH, L. D.; BONALDO, S. M.; OLIVEIRA, A. A. F. de. Bacillus subtilis: ENRAIZAMENTO E CRESCIMENTO DE MINIESTACAS DE EUCALIPTO EM SINOP, NORTE DE MATO GROSSO, BRASIL. **Biosci. J.**, *[S. l.]*, v. 29, n. Supplement 1, p. 1446–1457, 2013.

RADIĆ, S. *et al.* Effect of electric arc furnace slag on growth and physiology of maize (Zea mays L.). **Acta Biologica Hungarica**, *[S. l.]*, v. 64, n. 4, p. 490–499, 2013. Disponível em: https://doi.org/10.1556/ABiol.64.2013.4.8

RAJKUMAR, M.; FREITAS, H. Influence of metal resistant-plant growth-promoting bacteria on the growth of Ricinus communis in soil contaminated with heavy metals. **Chemosphere**, *[S. l.]*, v. 71, n. 5, p. 834–842, 2008. Disponível em: https://doi.org/10.1016/j.chemosphere.2007.11.038. Acesso em: 13 nov. 2022.

RAJKUMAR, M.; MA, Y.; FREITAS, H. Improvement of Ni phytostabilization by inoculation of Ni resistant Bacillus megaterium SR28C. **Journal of Environmental Management**, *[S. l.]*, v. 128, p. 973–980, 2013. Disponível em: https://doi.org/10.1016/J.JENVMAN.2013.07.001

RÊGO, M. C. F. *et al.* Morphoanatomical and Biochemical Changes in the Roots of Rice Plants Induced by Plant Growth-Promoting Microorganisms. **Journal of Botany**, *[S. l.]*, v. 2014, p. 1–10, 2014. Disponível em: https://doi.org/10.1155/2014/818797

RÊGO, M. C. F. *et al.* The role of rhizobacteria in rice plants: Growth and mitigation of toxicity. **Journal of Integrative Agriculture**, *[S. l.]*, v. 17, n. 12, p. 2636–2647, 2018. Disponível em: https://doi.org/10.1016/S2095-3119(18)62039-8

REIS, C. A. F. *et al.* **Corymbia citriodora: estado da arte de pesquisas no Brasil**. *[s. l.]*, 2013. Disponível em: https://ainfo.cnptia.embrapa.br/digital/bitstream/item/94547/1/Doc.-255-Reis-Ainfo1.pdf. Acesso em: 10 out. 2023.

REIS, C. A. F. *et al.* **Corymbia torelliana: estado da arte de pesquisas no Brasil**. Colombo: Embrapa Florestas, 2014.

RODRÍGUEZ-DÍAZ, M. *et al*. Uma revisão sobre a taxonomia e possíveis características de triagem de rizobactérias promotoras de crescimento de plantas. *In*: AHMAD, J. PICHTEL, J., S. H. (org.). **Interações Planta-Bactérias: Estratégias e Técnicas para Promover o Crescimento das Plantas**. Weinheim, Alemanh: Wiley-VCH Verlag GmbH & Co., 2008. p. 55–80. *E-book*.

ROZEFELDS, A. C. Eucalyptus phylogeny and history: A Brief Summary. **Tasforests**, *[S. l.]*, v. 8, p. 11, 1996.

SAMANIEGO-GÁMEZ, B. Y. *et al.* Bacillus spp. inoculation improves photosystem II efficiency and enhances photosynthesis in pepper plants. **Chilean journal of agricultural research**, *[S. l.]*, v. 76, n. 4, p. 409–416, 2016. Disponível em: https://doi.org/10.4067/S0718-58392016000400003. Acesso em: 18 out. 2022.

SANTOS, L. F. dos; OLIVARES, F. L. Microbial inoculants in agriculture and its effects on plant microbiome. *In*: **Rhizosphere Engineering**. *[S. l.]*: Elsevier, 2022. p. 151–169. *E-book*. Disponível em: https://doi.org/10.1016/B978-0-323-89973-4.00016-8

SCHWYN, B.; NEILANDS, J. B. Universal chemical assay for the detection and determination of siderophores. **Analytical Biochemistry**, *[S. l.]*, v. 160, n. 1, p. 47–56, 1987. Disponível em: https://doi.org/10.1016/0003-2697(87)90612-9

SHARMA, P. Role and significance of biofilm-forming microbes in phytoremediation -A review. **Environmental Technology & Innovation**, *[S. l.]*, v. 25, p. 102182, 2022. Disponível em: https://doi.org/10.1016/J.ETI.2021.102182

SHENG, X.-F. *et al.* Characterization of heavy metal-resistant endophytic bacteria from rape (Brassica napus) roots and their potential in promoting the growth and lead accumulation of rape. **Environmental Pollution**, *[S. l.]*, v. 156, n. 3, p. 1164–1170, 2008. Disponível em: https://doi.org/10.1016/j.envpol.2008.04.007

SHEPHERD, M. *et al.* Mapping species differences for adventitious rooting in a Corymbia torelliana × Corymbia citriodora subspecies variegata hybrid. **Tree Genetics & Genomes**, *[S. l.]*, v. 4, n. 4, p. 715–725, 2008. Disponível em: https://doi.org/10.1007/s11295-008-0145-1

SHI, R. yong *et al.* Effects of biomass ash, bone meal, and alkaline slag applied alone and combined on soil acidity and wheat growth. **Journal of Soils and Sediments**, *[S. l.]*, v. 17, n. 8, p. 2116–2126, 2017. Disponível em: https://doi.org/10.1007/S11368-017-1673-9/FIGURES/6. Acesso em: 3 nov. 2022.

SHIN, M.-N. *et al.* Characterization of lead resistant endophytic Bacillus sp. MN3-4 and its potential for promoting lead accumulation in metal hyperaccumulator Alnus firma. **Journal of Hazardous Materials**, *[S. l.]*, v. 199–200, p. 314–320, 2012. Disponível em: https://doi.org/10.1016/j.jhazmat.2011.11.010

SHU, K.; SASAKI, K. Occurrence of steel converter slag and its high value-added conversion for environmental restoration in China: A review. **Journal of Cleaner Production**, *[S. l.]*, v.

373, p. 133876, 2022. Disponível em: https://doi.org/10.1016/J.JCLEPRO.2022.133876

SILVA, A. V. da *et al.* Physiological, biochemical and nutritional changes in soybean in response to application of steel slag. **Australian Journal of Crop Science**, *[S. l.]*, v. 15, n. 15(12):2021, p. 1399–1405, 2021. Disponível em: https://doi.org/10.21475/ajcs.21.15.12.p3269. Acesso em: 27 ago. 2022.

SINGH, V. *et al.* Plant Growth Promoting Rhizobacteria (PGPR): Current and Future Prospects for Development of Sustainable Agriculture. **J Microb Biochem Technol**, *[S. l.]*, v. 7, n. 2, p. 96–102, 2015. Disponível em: https://doi.org/10.4172/1948-5948.1000188

SOBRAL, M. F. *et al.* Basic slag and its effects on the concentration of nutrients and heavy metals in sugarcane. **Revista Brasileira de Engenharia Agricola e Ambiental**, *[S. l.]*, v. 15, n. 8, p. 867–872, 2011. Disponível em: https://doi.org/10.1590/S1415-43662011000800015

TEIXEIRA, D. A. *et al.* Rhizobacterial promotion of eucalypt rooting and growth. **Brazilian Journal of Microbiology**, *[S. l.]*, v. 38, n. 1, p. 118–123, 2007 b. Disponível em: https://doi.org/10.1590/S1517-83822007000100025

TIWARI, M.; BAJPAI, D. S.; DEWANGAN, D. U. Steel Slag Utilization — Overview in Indian Perspective. **International Journal of Advanced Research**, *[S. l.]*, v. 4, n. 8, p. 2232–2246, 2016. Disponível em: https://doi.org/10.21474/IJAR01/1442

TRUEMAN, S. J. Cytokinin and auxin effects on survival and rooting of Eucalyptus pellita and E. grandis × E. pellita cuttings. **Rhizosphere**, *[S. l.]*, v. 6, p. 74–76, 2018. Disponível em: https://doi.org/10.1016/J.RHISPH.2018.04.001

TRUEMAN, S. J.; RICHARDSON, D. M. Relationships between Indole-3-Butyric Acid, Photoinhibition and Adventitious Rooting of Corymbia torelliana, C. citriodora and F 1 Hybrid Cuttings. **Tree and Forestry Science and Biotechnology**, *[S. l.]*, v. 2, n. 1, p. 26–33, 2008.

TU, X. *et al.* Advantages and risks of using steel slag in preparing composts from raw organic waste. **Journal of Environmental Science and Health - Part B Pesticides, Food Contaminants, and Agricultural Wastes**, *[S. l.]*, v. 52, n. 1, p. 30–36, 2017. Disponível em: https://doi.org/10.1080/03601234.2016.1224701

VALENTE LIMA, J. *et al.* Rhizobacteria modify root architecture and improve nutrient uptake in oil palm seedlings despite reduced fertilizer. **Rhizosphere**, *[S. l.]*, v. 19, p. 100420, 2021. Disponível em: https://doi.org/10.1016/J.RHISPH.2021.100420

VEJAN, P. *et al.* Role of Plant Growth Promoting Rhizobacteria in Agricultural Sustainability—A Review. **Molecules 2016, Vol. 21, Page 573**, *[S. l.]*, v. 21, n. 5, p. 573, 2016. Disponível em: https://doi.org/10.3390/MOLECULES21050573. Acesso em: 13 fev. 2023.

VERMA, S.; KUILA, A. Bioremediation of heavy metals by microbial process. **Environmental Technology & Innovation**, *[S. l.]*, v. 14, p. 100369, 2019. Disponível em: https://doi.org/10.1016/j.eti.2019.100369

VOCCIANTE, M. *et al.* The Role of Plant Growth-Promoting Rhizobacteria (PGPR) in Mitigating Plant's Environmental Stresses. **Applied Sciences**, *[S. l.]*, v. 12, n. 3, p. 1231, 2022. Disponível em: https://doi.org/10.3390/app12031231

WANG, G. *et al.* The combined use of a plant growth promoting Bacillus sp. strain and GABA promotes the growth of rice under salt stress by regulating antioxidant enzyme system, enhancing photosynthesis and improving soil enzyme activities. **Microbiological Research**, *[S. l.]*, v. 266, p. 127225, 2023. Disponível em: https://doi.org/10.1016/J.MICRES.2022.127225

WANG, J. *et al.* Bacillus velezensis SX13 promoted cucumber growth and production by accelerating the absorption of nutrients and increasing plant photosynthetic metabolism. **Scientia Horticulturae**, *[S. l.]*, v. 301, p. 111151, 2022. Disponível em: https://doi.org/10.1016/J.SCIENTA.2022.111151

WANG, X. *et al.* Isolation of As-tolerant bacteria and their potentials of reducing As and Cd accumulation of edible tissues of vegetables in metal(loid)-contaminated soils. **Science of the Total Environment**, *[S. l.]*, v. 579, 2017. Disponível em: https://doi.org/10.1016/j.scitotenv.2016.10.239

WANG, X. *et al.* Environmental risks for application of iron and steel slags in soils in China: A review. **Pedosphere**, *[S. l.]*, v. 31, n. 1, p. 28–42, 2021. Disponível em: https://doi.org/10.1016/S1002-0160(20)60058-3

WANI, P. A.; KHAN, M. S. Bacillus species enhance growth parameters of chickpea (Cicer arietinum L.) in chromium stressed soils. **Food and Chemical Toxicology**, *[S. l.]*, v. 48, n. 11, p. 3262–3267, 2010. Disponível em: https://doi.org/10.1016/j.fct.2010.08.035

WHITE, B. *et al.* Effect of silicate slag application on wheat grown under two nitrogen rates. **Plants**, *[S. l.]*, v. 6, n. 4, 2017. Disponível em: https://doi.org/10.3390/plants6040047

WORLD STEEL ASSOCIATION. **Sustainable steel: Sustainability Indicators 2021**. Beijing: China: World Steel, 2022. Disponível em: https://worldsteel.org/publications/bookshop/sust2021/

YI, H. *et al.* An Overview of Utilization of Steel Slag. **Procedia Environmental Sciences**, *[S. l.]*, v. 16, p. 791–801, 2012. Disponível em: https://doi.org/10.1016/j.proenv.2012.10.108

YILDIRIM, I. Z.; PREZZI, M. Chemical, Mineralogical, and Morphological Properties of Steel Slag. **Advances in Civil Engineering**, *[S. l.]*, v. 2011, p. 1–13, 2011. Disponível em: https://doi.org/10.1155/2011/463638

ZANONI DO PRADO, D. *et al.* Aspergillus flavipes as a novel biostimulant for rooting-enhancement of Eucalyptus. **Journal of Cleaner Production**, *[S. l.]*, v. 234, p. 681–689, 2019. Disponível em: https://doi.org/10.1016/J.JCLEPRO.2019.06.211