

Characterization of *Bacillus*, *Paenibacillus*, *Peribacillus*, and *Alkalihalobacillus* strains for indole-3-acetic acid production

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The biostimulation mechanisms of plants by microorganisms can be direct (acquisition of nutrients and production of phytohormones) and indirect (production of siderophores, antioxidants, enzymes, peptides, various secondary metabolites, volatile organic compounds, which can control phytopathogens and herbivores, in addition of giving the plant the ability to tolerate water, heat, and heavy metal stress, or activating systemic resistance pathways, giving the plants a priming effect). Not all beneficial microorganisms exhibit the above-mentioned mechanisms, challenging understanding their interactions with plants. Bacterial auxins (indole-3-acetic acid) may influence plant growth, affecting cell division, elongation, tropism, apical dominance, senescence, flowering, and stress response. We evaluated *in vitro* different *Bacillus*, *Paenibacillus*, *Peribacillus*, and *Alkalihalobacillus* strains for their ability to produce indole-3-acetic acid (IAA). A total of 187 endophytic, epiphytic, and rhizospheric strains isolated from native and cultivated plant species from different Brazilian biomes were obtained from the "Collection of Microorganisms and Microalgae Applied to Agroenergy and Biorefineries (CMMAABio)". To prepare the pre-inoculum, a single colony was inoculated into a 125 mL Erlenmeyer flask containing 25 mL of Tryptic Soy Broth (TSB). Cultivation was carried out overnight at 30 °C and 300 rpm. The 187 strains were inoculated [initial optical density (600 nm) adjusted to 0.2] in 125 mL Erlenmeyer flasks containing 25 mL of 10 % TSB with and without L-tryptophan precursor supplementation (5 mM) and incubated in the dark at 30 °C and 300 rpm for 72 hours (primary screening with one repetition). After the incubation period, 1 mL of each culture was centrifuged, and the supernatant was used for determination of indole compounds by colorimetric assay using the Salkowski reagent in miniaturized tests in 96-well microplates. The absorbance reading was performed using a spectrophotometer (520 nm). The concentration of indole compounds in the supernatant was determined using a standard curve, calculated based on known concentrations of IAA (5, 10, 25, 50, 100 µg/mL). Among the 187 strains, 142 were positive (75.9 %) in the primary screening. Twenty-two strains presented values above 5 µg/mL and were selected for triplicate cultivation. In secondary screening, the *Bacillus* sp. MGB2254 strain achieved the highest average IAA production in the supernatant (9.56 µg/mL). The selected strains will be evaluated for their potential to promote the growth of pre-sprouted sugarcane seedlings.

Key words: *Bacillus*; Indole-3-Acetic Acid; Sugarcane; Auxin.

Caracterização de linhagens de *Bacillus*, *Paenibacillus*, *Peribacillus* e *Alkalihalobacillus* para produção de ácido-3-indol acético

Linhagens de *Bacillus*, *Paenibacillus*, *Peribacillus* e *Alkalihalobacillus* foram avaliadas quanto à capacidade de produzir ácido-3-indol acético (AIA). Na triagem primária de 187 bactérias pelo método colorimétrico (uma repetição), 142 (75,9 %) produziram compostos indol no sobrenadante do meio TSB 10 % contendo 5 mM de L-triptofano. Na triagem secundária foram avaliadas (em triplicata) 22 linhagens que haviam produzido mais que 5 µg/mL de AIA. A quantidade de compostos indol no sobrenadante alcançou 9,56 µg/mL para *Bacillus* sp. MGB2254. As linhagens selecionadas serão avaliadas quanto ao potencial em promover o crescimento de mudas pré-brotadas de cana-de-açúcar.

Palavras-chave: *Bacillus*; Ácido-3-indol acético; Cana-de-açúcar; Auxina.

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