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Diazotrophic bacteria from a tropical forest soil with additional plant growth promoting traits: Prospective bioinoculants

K.T. Abeytunga, M.M. Wijesooriya and S.M.K. Widana Gamage*

Department of Botany, Faculty of Science, University of Ruhuna, Matara, Sri Lanka

Abstract

Soil microbes play a key role in ecosystem functioning by being part of numerous biogeochemical cycles and organic matter decomposition. For this reason, microbial inoculants could contribute immensely to the sustainable agriculture as natural nutrient mobilizers. In this study, we isolated and characterized nitrogen-fixing bacteria extracted from soil to investigate their potential to be used as bioinoculants to improve crop growth and development. Fifteen isolates of bacteria were isolated from the Kekanadura Forest Reserve in Matara, Sri Lanka (5°98'51.79"N-80°59'12.90"E). Their N-fixing potential was confirmed by the widely used peptone water qualitative assay. Agriculturally important traits such as phosphate solubilization, synthesis of indole-3-aceti acid (IAA) and antagonisms against plant pathogen, *Sclerotium rolfsii* were also tested. Phosphate solubilization was tested by growing the bacteria in Pikovskayas agar medium, which contains calcium phosphate as the insoluble phosphate source. Strain DzpC, DzpE, DzpF, DzpQ and Azp1 showed clear zone surrounding bacterial growth indicating solubilization of phosphate in the medium. The IAA production was tested spectrophotometrically in broth cultures supplemented with tryptophan as the precursor molecule. Five strains, DzpC, DzpE, DzpF, DzpI and DzpQ produced IAA in the range of 9.9-64.1 mg/mL. The antagonistic potential of strains was tested in dual culture plate assay. Thirteen strains showed growth inhibition of *S. rolfsii* at varying degrees. Strain DzpC, DzpP and Azp1 showed the strongest antagonistic activity. When overall results were considered, DzpC, DzpE, DzpF, DzpI and DzpQ were the best performing strains. Bacteria were identified by their colony morphology and sequencing of 16S rRNA gene. Seven out of fifteen strains were morphologically similar to actinomycetes. Four strains were identified as *Bacillus toyonensis*, *Lysinibacillus macroides*, *Rhizobium tropici* and *Rhizobium* sp. by DNA sequencing. Since actinomycetes and endospore-forming bacteria such as *Bacillus* and *Lysinibacllus* are generally resistant to adverse environmental conditions, potential application of majority of the strains isolated in this study for commercial formulation in crop cultivation is high. Furthermore, since bacterial strains were isolated from the native soil, they are expected to have better adaptability to Sri Lankan soil compared to exotic strains.

Keywords: Biofertilizer, Diazotrophs, Nitrogen-fixation, Plant growth promotion

****Corresponding Author:*** shirani@bot.ruh.ac.lk