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"Development on the margin"

Genetic Identification of Rhizobia Isolates: Behaviour on Bean (*Phaseolus vulgaris* L.) Genotypes

Ariany Colás Sánchez[1], Roldán Torres Gutiérrez[1], Bettina Eichler-Loebermann[2], Klever Ivan Granda Mora[3], Yenisey Gutiérrez Sánchez[1], Anne Willems[4], Jan Michiels[5], Jos Vanderleyden[5]

[1] *Central University of Las Villas, Faculty of Agricultural Sciences, Cuba*
[2] *University of Rostock, Faculty of Agricultural and Environmental Sciences, Germany*
[3] *Universidad Nacional de Loja, Area de Recursos Naturales, Ecuador*
[4] *Gent University, Laboratory of Microbiology, Belgium*
[5] *Katholieke Universiteit Leuven, Centre for Microbial and Plant Genetics, Belgium*

Abstract

The work was carried out with the aim to identify and characterise isolates of *Rhizobium* genus from 13 sampling areas in central provinces of Cuba (Sancti Spiritus, Villa Clara and Cienfuegos), and to determine the effect of the isolates on phenotypic and growth parameters of common bean genotypes under controlled conditions. The morphological analysis was based on determining the differences of the colonies obtained from isolation, which evaluated the Gram stain, growth type, colour, slime production, edge and elevation. The genetic identification of isolates was performed by molecular tools using 16S rRNA gene sequencing. Under controlled conditions the nodulation, phenotypic parameters, biomass and nitrogen content were evaluated on ICA Pijao and BAT-304 common bean genotypes. The results showed a wide morphological diversity of native strains in the soils of central Cuba. A total of 33 isolates showed different characteristics in at least one parameter measured. A total of 22 sequences were analysed for genetic identification, turning out six genus of bacteria matching to *Arthrobacter*, *Chryseobacterium*, *Enterobacter*, *Stenotrophomonas*, *Pseudomonas* and *Rhizobium* genus. Rhizobia were the only bacteria with differences at species level. From eight sequences aligned belonging to the genus *Rhizobium*, four species (*Rhizobium* sp., *R. etli*, *R. pisi* and *R. radiobacter*) were identified including the first report of *R. pisi* for Cuban soils. Phenotypic analysis showed the beneficial effect of the species *R. etli* and *R. pisi*, respectively, increasing the growth parameters, biomass production and nitrogen content on bean genotypes. Genotypic variability demonstrated the close correlation among these strains and genotype BAT-304 compared with ICA Pijao.

Keywords: Genetic identification, growth parameters, nitrogen content, *Phaseolus vulgaris*, *Rhizobium*, strains

Contact Address: Bettina Eichler-Loebermann, University of Rostock, Faculty of Agricultural and Environmental Sciences, J. von Liebig Weg 6, 18059 Rostock, Germany, e-mail: bettina.eichler@uni-rostock.de