

TITLE: COLONIZATION OF MAIZE BY *BACILLUS* SP. RZ2MS9, A PLANT GROWTH-PROMOTING RHIZOBACTERIUM FROM GUARANA

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Corn is one of the most economically important crops of Brazil, which produced almost 67 million tons of grains in the harvest of 2015/2016. In contrast, this crop requires an intensive amount of mineral fertilizers, mainly nitrogen fertilizers. One of the alternatives to reduce the use of this chemical inputs is the use of nitrogen-fixing bacteria, such as *Azospirillum brasilense*, already marketed to be used in corn, but with some inconsistent results. It's already known that microorganisms do not act alone, both on the ground or inside the plants there are millions of bacteria interacting in complex ways. In the search for other microorganisms capable of performing similar task, *Bacillus* sp. strain RZ2MS9, originally isolated from the rhizosphere of Brazilian guarana, was previously reported as corn and soybean growth promoter. To understand the interaction of RZ2MS9 with corn, this strain was GFP-labeled. The electroporation was performed using integrative and replicative plasmids (pNKGFP and pBAV1k-T5gfp). Corn seeds were inoculated with the strain RZ2MS9 labeled with the plasmid pNKGFP (RZ2MS9::pNKGFP) and also co-inoculated with RZ2MS9::pNKGFP and *Azospirillum brasilense* (Ab-v5::pWM1013), previously labeled with red fluorescence, aiming to understand if the co-inoculation changes the behavior of the strains. The transformant RZ2MS9::pNKGFP demonstrated a behavior similar to the wild, demonstrating the same ability to plant growth-promoting in corn. The plants cuts were analyzed by fluorescent optical microscopy, 15 and 30 days after germination (DAG). For quantification of the strain RZ2MS9::pNKGFP in corn shoot and roots, qPCR was performed with the primers PPNKF (5 'CCTTCATTACAGAAACGGC 3') and PPNKRII (5 'GGTGATGCGTGATCTGATCC 3'). The microscopic images, as well as the qPCR revealed that RZ2MS9::pNKGFP colonizes the whole corn plant, being more abundant in the leaves. These results will help us to determine the best way to apply RZ2MS9 in corn. The co-inoculation analysis showed a reduction of the RZ2MS9 colonization at 15 DAG, which was not observed at 30 DAG.

Keywords: Plant growth-promoting, Monitoring, GFP, qPCR,

Development Agency: FAPESP