Herbaspirillum seropedicae rfbB and *rfbC* Genes Are Required for Maize Colonization.

Balsanelli, E.[1], Serrato, R.V.[1], Baura, V.A.[1], Sassaki, G.[1], Wassem, R.[2], Yates, M.G.[1], Pedrosa, F.O.[1], Souza, E.M.[1] and Monteiro, R.A.[1]

[1] Department of Biochemistry and Molecular Biology, Universidade Federal do Paraná, Curitiba, PR, Brazil; [2] Department of Genetic, Universidade Federal do Paraná, Curitiba, PR, Brazil

Herbaspirillum seropedicae is a diazotrophic endophyte known to associate with many agriculturally important poaceae. Although large numbers of *H. seropedicae* cells are found in internal plant tissues, the molecular mechanisms of colonization are not understood. Lipopolysaccharide, the outermost component of the cell envelope of gram-negative bacteria, may be involved in the early stages of plant colonization by bacteria. Since rhamnose is a monosaccharide frequently found in LPS, we disrupted two genes, *rfbB* and *rfbC*, involved in its biosynthesis, to test whether LPS has a role in **the** plant-*H. seropedicae* interaction. The **obtained** mutant strains had a very different LPS **pattern from the** wild-type strain. Plant inoculation assays showed that the attachment step of the colonization process depends on **the** *H. seropedicae* surface molecules, since the number of wild-type bacteria attached to maize root surfaces was approximately 100-fold higher than that of the mutant strains. The addition of isolated wild type LPS, glucosamine or N-acetyl glucosamine **when the wild-type** strain was used as an inoculant for maize also led to decreased attachment, suggesting that these additions blocked bacterial attachment sites. LPS seems to have a specific participation in *H. seropedicae* attachment to plant roots, since attachment to glass fiber was not altered by the mutations. The number of wild type bacteria colonizing the internal plant tissues **was 2- and 3-fold higher than the mutants one and three days respectively after inoculation.** The finding that *H. seropedicae rfbB* and *rfbC* **gene** knockout decreases endophytic association suggests that attachment and recognition of the bacteria by the plant involves surface molecules, most probably LPS.