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Differential efficiency of nitrate respiration between Bradyrhizbium diazoefficiens and Bradyrhizobium japonicum

F. Siqueira, Arthur[1], S_nchez, Cristina[1], Itakura, Manabu[1], Minamisawa, Kiwamu[1]

1Touhoku University - Graduate School of Life Sciences

Denitrification is an alternative respiratory mechanism that occurs in limitation of oxygen, characterized by using nitrate or other nitrogen oxide as electron acceptor, reducing it sequentially in $NO_3 \rightarrow NO_2 \rightarrow NO \rightarrow N_2O \rightarrow N_2$ by nitrate reductase, nitrite reductase, nitric oxide reductase and nitrous oxide reductase (encoded by *nosZ* gene), respectively. This process is found in *Bradyrhizobium diazoefficiens* and *B. japonicum* that nodulate soybean plants for symbiotic nitrogen fixation. *B. diazoefficiens* strains contain *nos* gene cluster (*nosZ*+) and are able of complete denitrification, while *B. japonicum* strains lack nos gene cluster (*nosZ*-). Shiina et al. (2014) observed *B. diazoefficiens* (Bd) dominance in alluvial soils, characterized as anaerobic environments. Moreover *B. japonicum* (Bj) strains were dominant in Andosol soils, characterized as aerobic environment, suggesting that different ability of nitrate respiration might drive the distribution of Bj and Bd strains in soybean fields. Growth experiments performed under free-living conditions showed that Bj strain USDA 6 (natural *nosZ*-) and Bd strain USDA 110 (natural *nosZ*+) have the same growth in HMM medium under aerobiosis, however when in anaerobic nitrate-respiring condition (10mM KNO_3), USDA 6 grew less than USDA 110. The lack of *nosZ* gene seems not to be the cause of this difference since *nosZ* mutant of USDA 110 had the same growth as the wild-type. To further investigate this and extrapolate as global phenomena, the aerobic, microaerobic (2% O_2) and anaerobic growth of randomly selected Bd and Bj strains was tested. In total, 27 strains were selected, 12 of Bj and 15 of Bd. Anaerobic growth curves in presence of nitrate were statistically different between Bd and Bj strains, however no difference was observed in aerobic or microaerobic growth curves. Our results strongly suggest a differential response of nitrate reduction to N_2O between Bd and Bj strains.

keywords:Denitrification,Bradyrhizobia,Nitrate-respiring,Nitrous Oxide,Anaerobic,