

Screening of acid aluminium tolerant *Bradyrhizobium japonicum* strains: analysis of marker genes and competition in planta

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Affiliations

Bogor Agricultural University, Indonesia. Faculty of Mathematics and Sciences. Dept, of
Biology

Bogor Agricultural University, Indonesia. Faculty of Mathematics and Sciences. Dept, of
Biology

Bogor Agricultural University, Indonesia. Faculty of Mathematics and Sciences. Dept, of
Biology

Bogor Agricultural University, Indonesia. Faculty of Mathematics and Sciences. Dept, of
Biology

Abstract

The genes, *inaZ* from *Pseudomonas syringae*, *xylE* from *P. putida* and the kanamycin resistance gene (KmR) of mini-Tn5Km1, were studied for their capabilities and reliabilities as molecular markers in three acid tolerant *Bradyrhizobium japonicum* strains. Conjugation was used to transfer each of these genes to *B. japonicum* strains. Mating was conducted on membrane filter (0.45 gm, Millipore) using modified Luria Agar. The results showed that all of the genes were able to be transferred to acid tolerant *B. japonicum* strains by conjugation. All of these bacteria were able to express the *inaZ* and KmR genes. However, none of the exconjugants was able to express the *xylE* gene. Acid tolerant *B. japonicum* strains that carried each of these genes were able to form root nodules in either siratro or soybean plant. The KmR gene had high stability as tested after nodulation. experiments. This gene was subsequently used as a genetic marker in a competition study of acid tolerant *B. japonicum* strains for nodule occupancy in soybean plant. This study was conducted using acid tolerant soybean cultivar (B 09) grown in Leonard jars using nitrogen free nutrient solution (pH 4.5 + Al 50 μ M). Mixtures of acid tolerant *B. japonicum* strains and USDA 110 were inoculated in 1:1 ratio and nodules were harvested 30 days after planting. The results indicated that one exconjugant strain (11.71Km) dominated root nodules. Thus this strain has the potential to be developed as soybean inoculant in acid soils.