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UNRAVELING PSYMA MOBILIZATION IN *Sinorhizobium meliloti* LPU88: INSIGHTS INTO CONJUGATION SYSTEMS

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Rhizobia are nitrogen-fixing bacteria capable of establishing a facultative mutualist relationship with legume plants. In such interaction, rhizobia convert atmospheric nitrogen into reduced forms, that are supplied to the plants in exchange of carbohydrates. This process plays a crucial role in the nitrogen cycle and contribute to the development of a sustainable agriculture. The symbiotic interaction between *Medicago sativa* and *Sinorhizobium meliloti* serves as a model system for exploring the genetic and evolutionary aspects of rhizobial-legume interactions, as well as the nitrogen fixation process. Strains of *S. meliloti* possess at least three replicons: a chromosome and two megaplasmids pSymA and pSymB. Additionally, some strains may harbor accessory plasmids. Comparative genomics studies have shown that pSymA is one of the replicons contributing the greatest diversity among *S. meliloti* strains, although the specific genes responsible for this diversity are not yet characterized. Also, pSymA may have been acquired through Horizontal Gene Transfer (HGT) due to its lower GC content compared to the other replicon. In our laboratory, we characterized the *S. meliloti* LPU88 strain and recently obtained the complete genome sequence. LPU88 presents five circular replicons: a chromosome, two megaplasmids (pSymA and pSymB) and two accessory plasmids (p88a and p88b). Since one of our research interests is plasmid conjugation, we propose to study the conjugation systems in the LPU88 strain, with a primary focus on pSymA (the main source of diversity among *S. meliloti* strains). In previous studies, we demonstrated that pSymALPU88 conjugates from the environment of strain LPU88 at a conjugation frequency of $10^{??}$. Then, the remobilization of pSymALPU88 to another strain of *Agrobacterium tumefaciens* UBAPF2 (Gm^R) occurred at frequencies of 10^{-10} . Conjugative systems are composed of the DNA transfer and replication (Dtr) and Mating pair formation (Mpf) genes. These genes encode proteins responsible of DNA processing and conjugative pore formation, respectively. LPU88 strain contains 4 Dtr and 3 Mpf systems distributed across pSymA, pSymB, p88a and p88b. Given that pSymALPU88 is transferred at higher frequencies from the LPU88 strain environment, we wondered which conjugation system might be responsible for this observed phenotype. For that purpose, we compared the conjugation frequency of pSymALPU88 from Sma818R strains harboring only the pSymALPU88 or said replicon with different combinations of accessory

plasmids of LPU88. As a result, the conjugation frequency from the constructed genetic backgrounds was approximately 10^{-1} . This suggests that there might be a crosstalk in the genetic background of the LPU88 strain that we could not replicate in the constructed strains. As a next step, we aim to construct mutants in the genes involved in conjugation to identify the conjugation system responsible for the mobilization of pSymALPU88.

Palabras clave: *Sinorhizobium meliloti* - Conjugation systems - pSymA