Triparental Mating of *Sinorhizobium meliloti*

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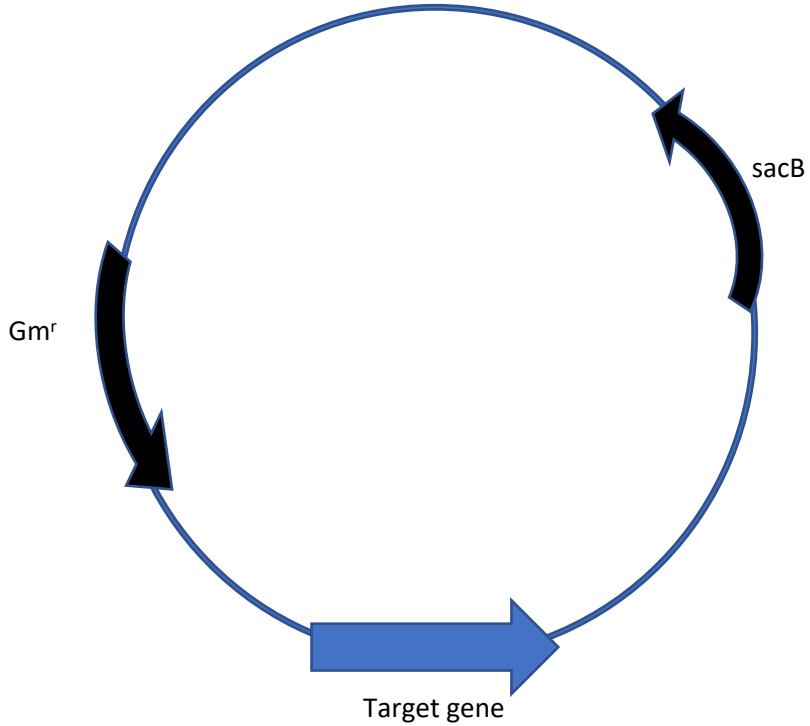
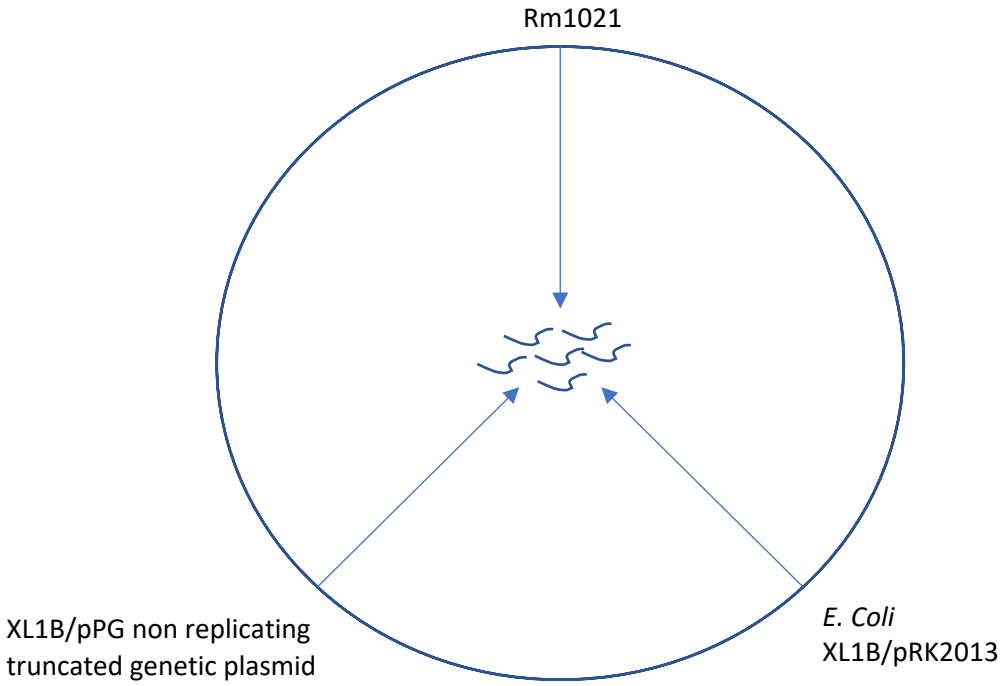
Plasmid Example



Inoculated LB Petri Plate



The triparental mating process for *Sinorhizobium meliloti*. *S. meliloti* is a bacterium which is commonly known for the beneficial symbiotic relationship it establishes with legume hosts. This relationship involves the legume host providing bacteria with an abundant carbon source, and *S. meliloti* responding with the conversion of di-nitrogen to ammonium, which would otherwise be unavailable to the host. We have been analyzing the Sma0113/Sma0114 two component signal transduction system, consisting of a sensor histidine kinase and a response regulator. Via bioinformatic analysis, we have identified genes that encode proteins which we believe carry out the same function as Sma0113 or Sma0114. We have used a triple mating protocol to knock out these duplicative genes in the wild-type host Rm-1021, using a plasmid with a truncated gene that will not replicate in *S. meliloti*. It will integrate into the chromosome via homologous recombination. The knockouts and integration of plasmid duplicates allows the analysis of a variety of different phenotypic responses in *S. meliloti* and are helping to further understand the Sma0113/Sma0114 two component system.