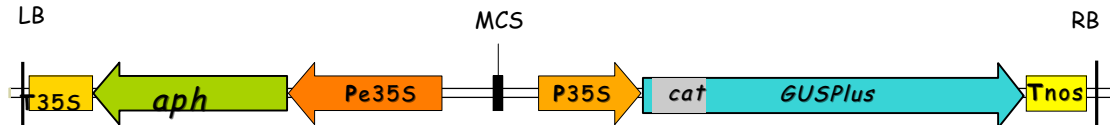


Construction and maps of binary vectors

T-DNA map for pCAMBIA vectors pC1105.1, pC1105.1r

T-DNA contains the hygromycin resistance gene (*aph*) for plant selection and the *GUSPlus* gene as a reporter to investigate gene transfer. P35S and Pe35S, promoter and enhancer+promotor from CaMV35S respectively; Tnos and T35S, terminators from *A. tumefaciens* nopaline synthase gene and CaMV35S respectively; *cat*, catalase intron; LB and RB, left and right T-DNA border sequence.



Multiple cloning site (MCS) sequences in pCAMBIA1105.1 used in *A. tumefaciens*, and pCAMBIA1105.1R, exclusively used in non-*Agrobacterium* bacterial species, respectively. Primers used to distinguish between pCAMBIA1105.1 ('non-R') and pCAMBIA1105.1R ('R') are 5'-CTGGCACGACAGGTTTC-3' and 5'-TACGGCGAGTTCTGTTAGGT-3', encompassing the MCS region. These give PCR products of 491bp (pCAMBIA1105.1) and 572bp (pCAMBIA1105.1R).

pCAMBIA1105.1 (2638)ATTACGaattcgagct...N39 ...gcaagcttggCACTGG

pCAMBIA1105.1R (2638)ATTACGccaagcttgg...N138...tattacaattCACTGG

- pCAMBIA1105.1 was derived from pCAMBIA1405.1 by removal of the kanamycin marker so only the spectinomycin/streptomycin marker was retained.
- pCAMBIA1105.1R was derived from pCAMBIA1105.1 by removing the *PvuII*-*PvuII* MCS fragment in pCAMBIA1105.1 and replacing it with the larger *PvuII*-*PvuII* MCS from pCR11, following re-ligation of the *EcoRI* sites.

Schematic diagrams of pCambia1105 and pCambia1105.1r vectors

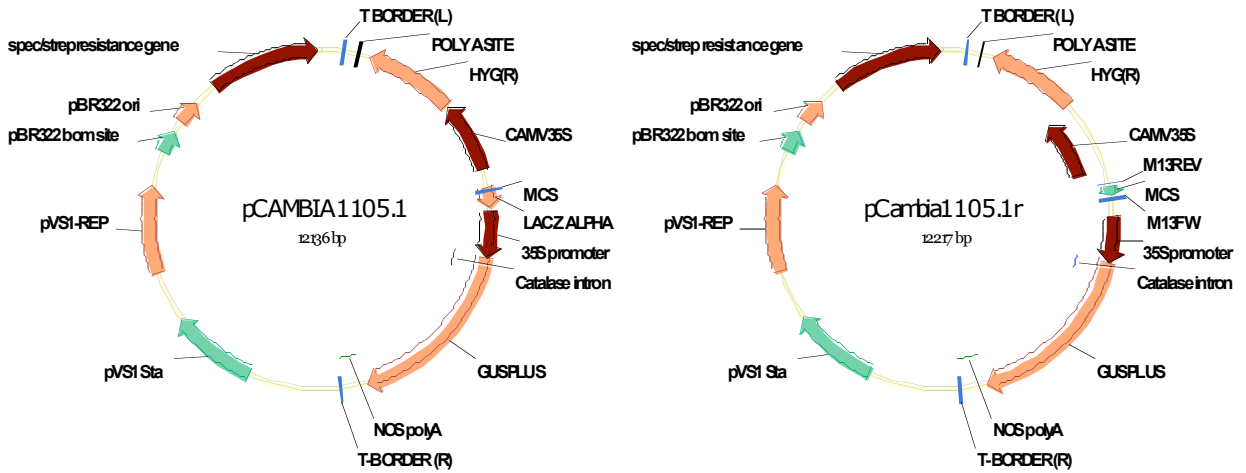


Figure shows typical PCR confirming presence of Ti plasmid and binary vectors and absence of *Agrobacterium* contamination in rhizobial spp.

