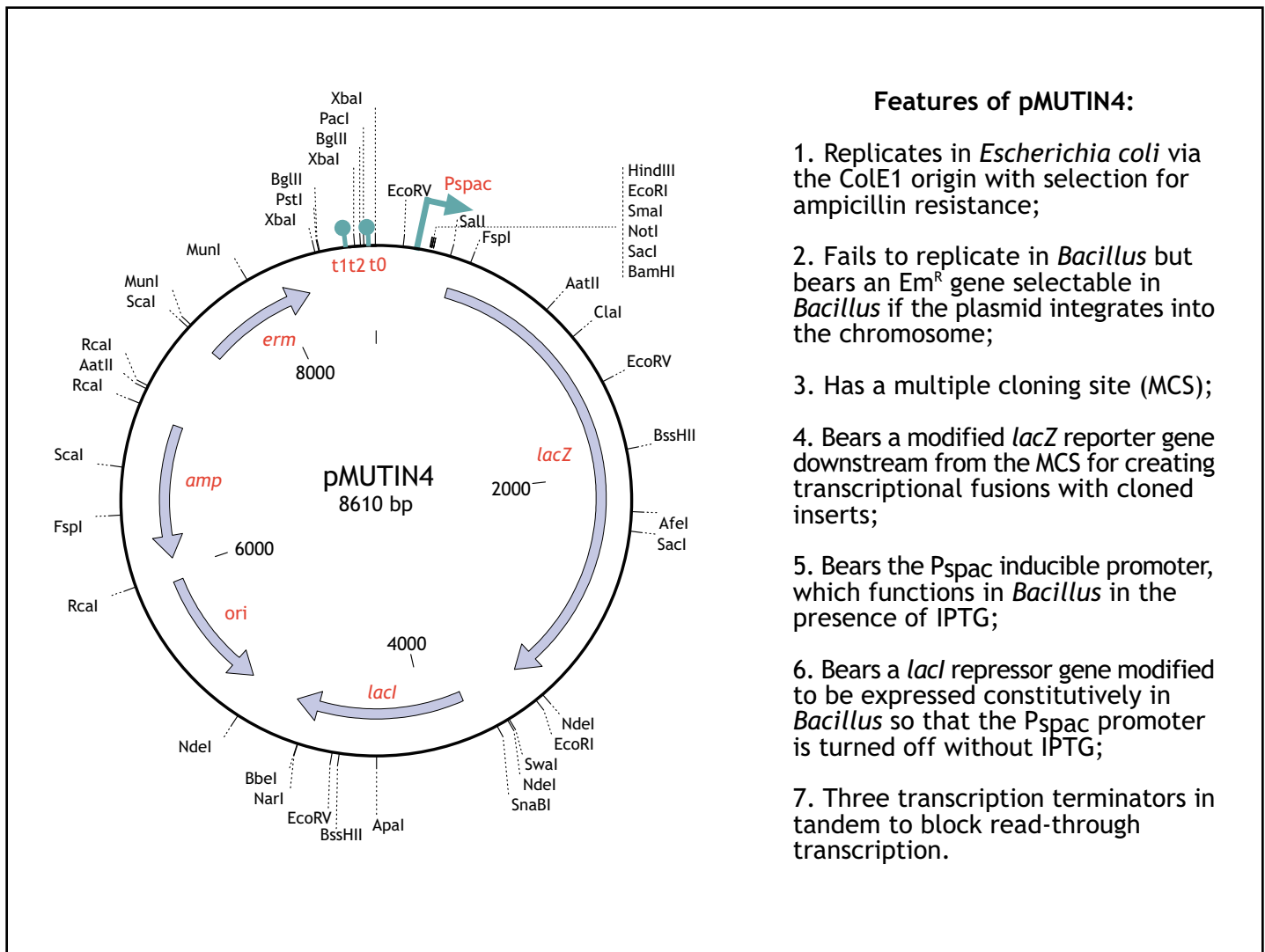


A Vector Useful for Gram-Positive Genomics



From the *Bacillus subtilis* genome project have come several tools that should prove useful to any researcher studying Gram-positive genomics. Among these tools is the integrative vector, pMUTIN4 (Vagner, V., E. Dervyn and S. D. Ehrlich. 1998. A vector for systematic gene inactivation in *Bacillus subtilis*. *Microbiol.* 144:3097-3104).

In principle, pMUTIN4 should allow the researcher to produce knockout or conditional expression mutations in any unknown coding sequence. To use this vector, one inserts a chromosomal fragment of interest into the multiple cloning site. Upon transformation into the Gram-positive host, the recombinant plasmid integrates into the chromosome by homology to the insert. If the insert is completely internal to a coding sequence, a knockout mutation is produced. If the insert instead contains regulatory sequences upstream from a coding sequence, the *lacZ* reporter gene is placed under their control. If the insertion event separates downstream genes from their promoter, their expression becomes dependent on the IPTG-inducible Pspac promoter. The pMUTIN4 vector may be obtained from the *Bacillus* Genetic Stock Center in *Escherichia coli* host strain **ECE139**.

