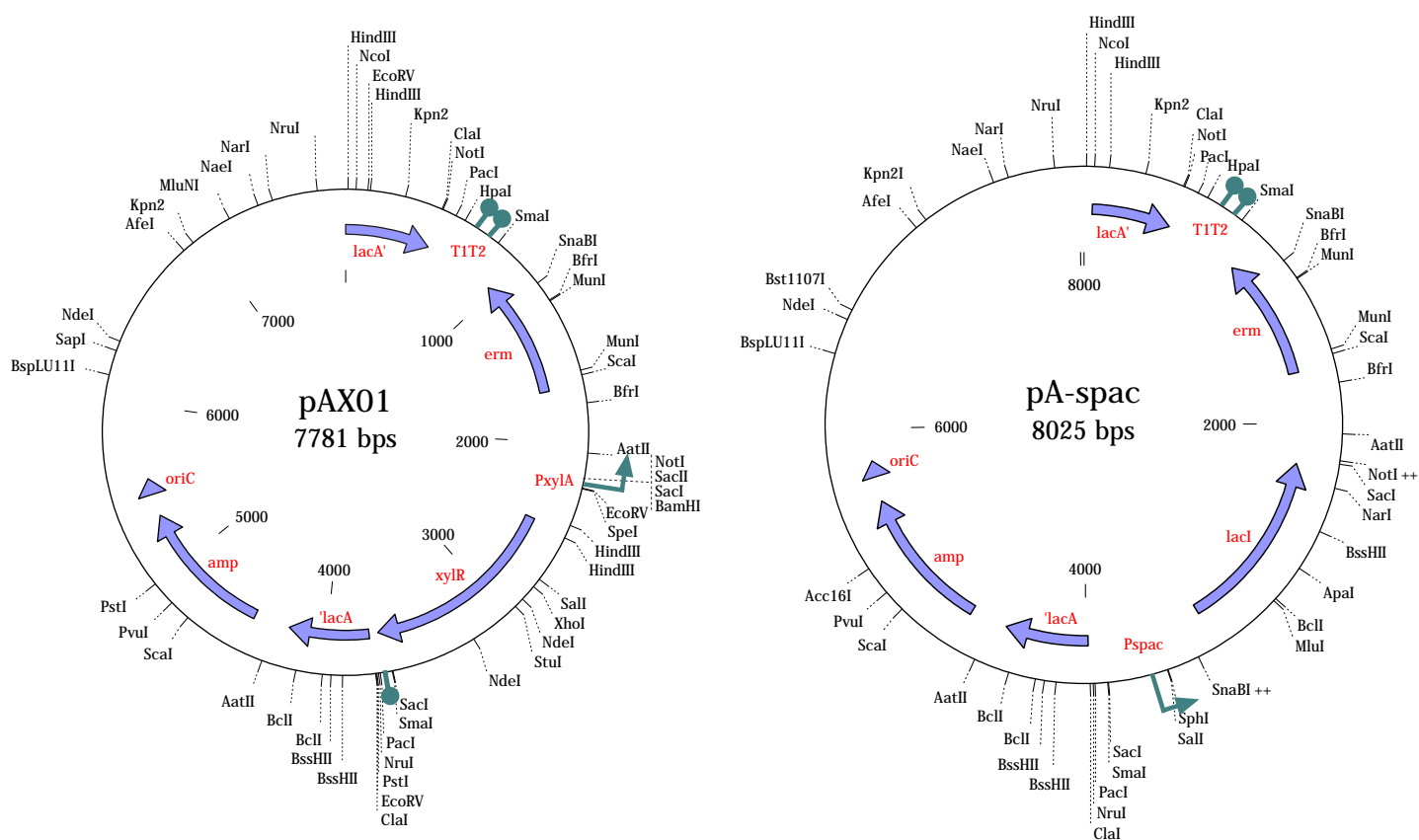


## Bacillus subtilis Integration Vectors with Inducible Expression of Cloned Inserts



From the laboratory of Wolfgang Schumann at the University of Bayreuth come two new expression vectors capable of integrating into the *Bacillus subtilis* chromosome at the *lacA* locus. Each allows for regulated expression of cloned inserts. The first, pAX01, features xylose induction, while the second, pA-spac, feature IPTG induction. To use these plasmids, a PCR replicon or subcloned fragment is inserted into an appropriate restriction site. The recombinant plasmid is transformed into one of the supplied *B. subtilis* host strains, with selection for erythromycin resistance. A double recombination event between the plasmid and chromosomal *lacA* sequences delivers the expression cassette into the chromosome, replacing a spectinomycin resistance gene inserted into the host *lacA* locus. One of the hosts also contains a chloramphenicol resistance gene in the chromosomal *amyE* locus, allowing for integration of a second plasmid (such as the pDG series in strains ECE112-ECE116) in this separate locus. For a more complete description of these vectors and Härtl, B., W. Wehrl, T. Wiegert, G. Homuth, and W. Schumann. 2001. *J. Bacteriol.* 183:2696-2700. See also the Schumann web site at <http://btbgn1.bio.uni-bayreuth.de/lsgenetik1/frames.htm>.

### BGSC Strains

- ECE137 contains pAX01, a 7781 bp plasmid in *E. coli* JM109; integrates into the *B. subtilis* chromosome at *lacA* with ; allows for xylose-inducible expression of cloned insert; for use with recipient strains 1A785
- ECE138 contains pA-spac, 8025 bp plasmid in *E. coli* JM109; integrates into the *B. subtilis* chromosome at *lacA* with selection for Em; allows for IPTG-inducible expression of cloned insert; for use with recipient strains 1A785 and 1A786
- 1A785 original code: IHA01; genotype: *lacA::spec leuB8 metB5 hsd<sub>R</sub>IR-M<sub>-</sub> Spr*. Recipient strain for ectopic integration into *lacA*; use with plasmids pA-spac or pAX01
- 1A786 original code: IHA02; genotype: *amyE::cat lacA::spec leuB8 metB5 hsd<sub>R</sub>IR-M<sub>+</sub> Cm<sub>R</sub> Spr*. Recipient strain for ectopic integration into *amyE* and/or *lacA*; use with plasmids pA-spac or pAX01 for *lacA* integration