

Methods for Metabolic Engineering of *Thermoanaerobacterium saccharolyticum*

Background

- Thermoanaerobacterium saccharolyticum* is an anaerobic thermophilic bacterium that can ferment C6 sugars, C5 sugars, and C5 sugar polymers. Techniques for genetic modification of this organism have been developed over the last decade but were published across many different periodicals.

Approach

- Best practices were summarized in this chapter for several important techniques, including:
 - 16S analysis
 - Positive and negative selection systems
 - Natural competence transformation protocol
 - Chromosomal modification by homologous recombination
 - Phenotype analysis by fermentation and enzyme assays
 - Heterologous protein expression

Significance

- A single resource for techniques for metabolic engineering of *T. saccharolyticum* will make it easier for researchers unfamiliar with this organism to start using it.

