An Investigation of Potential Gene Over-Expression and siRNA Methods for Causing Cell Lysis and Stasis

Hines, Jordan

This research determined methods for both cell lysis and cell stasis based on simple methods in metabolic engineering. These methods could be useful to maximize the amount of product produced by cells and to allow for the extraction of products that cannot exit the cell. One method, using small interfering RNA to bind to targeted mRNA and thus blocking production of targeted proteins, rendered no lysis or stasis with any of combinations of genes related to cell wall production. The other method, over-expressing individual genes using plasmids from the ASKA collection, found multiple successful options. Over-expression of dacB was found to cause cell lysis at early and middle stages of E. coli growth and showed a similar effect to adding ampicillin. Two similar genes, dacA and dacB, showed similar results but took longer to take effect. Over-expression of sulA was found to cause cell stasis at all stages of growth. In addition, the concentration of IPTG (the inducer) added to the flasks of E. coli was optimized. Among other genes tested, ypjF and mrdA also showed interesting results. Tests measuring the concentration of L-Phenylalanine in the cultures did not show a clear correlation between the time at which sulA was over-expressed and the L-Phenylalanine concentration, contrary to what was predicted.

Awards Won: