

(1) The central aim of the project was to improve the *E. coli* genome by streamlining/re-structuring its genome. We sought to answer the long-standing question whether fitness of the bacterium can be improved by removing genes unused under selected conditions.

Results: A selection-driven, random deletion method was developed. It allows streamlining of the genome in a cyclic fashion, produces deletions randomly in terms of location and size, produces one deletion per cycle, and can be applied to a relatively large population of the cell. The procedure involves selection steps for growth rate.

We performed streamlining experiments in parallel populations in up to 5 cycles. All together 65 genomes were sequenced for analysis of the deletions obtained.

We found that deletions were formed by a variety of processes, including spontaneous loss of prophages, homologous recombination between repeat sequences, and repairing a double stranded break by an alternative end joining mechanism, using microhomologies spread over the genome. The process serves as a model of natural processes. Propensity of genomic segments for elimination is hierarchic (some segments are readily/frequently lost).

We proved that loss of some genomic segments improves fitness. The choice of such genes/regions is, however, limited (similar deletions appear in parallel populations). A typical fitness-improving change is the deletion of the energy-consuming flagellar apparatus, unnecessary in shaking, rich media cultures.

Improvements in fitness under the conditions applied throughout the cycles are accompanied by rapid decrease of fitness under other conditions.

The work resulted in a publication in *Scientific Reports*, and served as the basis of a PhD thesis.

(2) In a side project, aimed at the genetic adjustment of the codon bias of *E. coli*, we engineered a strain that proved to be a superior host for expression of heterologous proteins. The work was published in *ACS Synthetic Biology* (both corresponding authors are from the group).