Laboratory of Systems and Synthetic Biology
Wageningen University and Research Center
Dreijenplein 10, Wageningen
6703 HB, the Netherlands

Thesis project:

Synthetic Biology - Characterizing promoters in Pseudomonas putida KT2440

Stamatios Damalas
E: stamatios.damalas@wur.nl
Building 316
Room 0.023

Background

Pseudomonas putida is a gram-negative soil bacterium that is renowned for its metabolic flexibility. This metabolic flexibility underlies an inherent toxin and solvent tolerance, which make P. putida an organism of interest for laboratory, industrial and environmental applications. It can (i) tolerate toxic intermediates, (ii) survive in biphase production processes, (iii) produce a large range of products, and (iv) handle harsh growth conditions. Besides this flexibility, and in comparison to its relatively close model organism Escherichia coli, the available synthetic biology tools are very little.

Goal

We want to screen and characterize different promoters for gene expression in P. putida. Robust gene expression based on characterized modular systems is critical in Synthetic Biology. In this context we need to observe and document the expression behavior of specific promoters. These promoters could be Pseudomonas intrinsic promoters like a fusaric acid sensitive promoter, orthogonal like the rhamnose inducible promoter from E. coli or engineered based on the arabinose inducible system (araC prototype). Basic tool for this purpose will be our newly developed plasmid format called the METAbrick which allows for easy exchange and construction of modular parts.

This will involve:

1. Theoretical discussion on the design of the recombineering parts.
2. DNA engineering and parts construction.
3. Screening of recombineering success in P. putida – 2 systems RecTE (Swingle et al., 2010) and lambda Red (Datsenko and Wanner, 2000).
4. Application of the system for the manipulation of P. putida genome.

General

This project will include a concise written report (in English, with critical assessment of the work) and an oral presentation of the work. The project will include synthetic biology and molecular biology techniques along with the usage of biophysical tools.

Contact

Stamatios Damalas (stamatios.damalas@wur.nl) and Rita Volkers (rita.volkers@wur.nl).