

Thesis project:

Stamatios Damalas

Synthetic Biology - Screening of alkane production in *Pseudomonas putida*.

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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 biphasic 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

After confirming the expression of the enzymes needed for alkane production from a newly constructed plasmid, we want to screen for alkane production both in *E. coli* and *P. putida*. Based on the results we will try to select the best conditions for optimal yield, improve the production by using protein engineering or alter the chain length of the produced molecules through metabolic pathway engineering (Schirmer et al., 2010, Choi and Lee, 2013, Akhtar et al., 2013).

Final target is to repurpose *P. putida* from being an alkane degrader to being an alkane producer with “a la carte” ability of hydrocarbon production.

This will involve:

1. Theoretical discussion on the design of the alkane production.
2. Protein engineering for optimal expression yield.
3. DNA engineering and parts construction.
4. Screening of alkane production in *P. putida* and *E. coli*.

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

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AKHTAR, M. K., TURNER, N. J. & JONES, P. R. 2013. Carboxylic acid reductase is a versatile enzyme for the conversion of fatty acids into fuels and chemical commodities. *Proceedings of the National Academy of Sciences*, 110, 87-92.

CHOI, Y. J. & LEE, S. Y. 2013. Microbial production of short-chain alkanes. *Nature*, 502, 571-4.

SCHIRMER, A., RUDE, M. A., LI, X., POPOVA, E. & DEL CARDAYRE, S. B. 2010. Microbial Biosynthesis of Alkanes. *Science*, 329, 559-562.