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Poster Session

Developing Novel Pathways for Butanol Production in E. coli

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In the biofuels market, n-butanol has been gaining attention due to its superior fuel properties when compared with ethanol. Although n-butanol is naturally produced by solventogenic bacteria through ABE fermentation, the low productivities obtained with this bioprocess discouraged its use. Thus, most n-butanol produced nowadays is chemically synthesized via petrochemical routes. One possible alternative is to express non-native pathways in microbial factories.

Our work focuses on the production of butanol through heterologous pathways, previously generated within our group [1] using a (hyper)graph-based algorithm, in *Escherichia coli*. First, we evaluated these pathways according to diverse criteria including the number of biochemical steps required, maximum yield, conservation of carbon atoms and novelty. A novel pathway generated by the algorithm, which is significantly different from the available in solventogenic bacteria, was then introduced in a genome-scale model of *E. coli* (iJO1366) and the production of butanol was optimized *in silico* using Optflux (<http://www.optflux.org>), a software platform for metabolic engineering. Several different environmental conditions and sets of gene knock-outs were evaluated with the aim of redirecting the cell metabolism to solvent production. The biological significance of the predicted phenotypes was analyzed to rationally design a strain that couples growth and butanol production.

Using biomolecular techniques, a novel strain of *E. coli* – able to produce butanol through the selected pathway in the same conditions simulated in *in silico* – is currently being constructed.

[1] F. Liu, P. Vilaça, I. Rocha, and M. Rocha, "Development and application of efficient pathway enumeration algorithms for metabolic engineering applications," *Comput. Methods Programs Biomed.*, Dec. 2014.