OPENFLUX: EFFICIENT MODELLING SOFTWARE FOR 13C-BASED METABOLIC FLUX ANALYSIS

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The quantitative analysis of metabolic fluxes, i.e., in vivo activities of intracellular enzymes and pathways, provides key information on biological systems in systems biology and metabolic engineering. It is based on a comprehensive approach combining (i) tracer cultivation on 13C substrates, (ii) 13C labelling analysis by mass spectrometry and (iii) mathematical modelling for experimental design, data processing, flux calculation and statistics. Whereas the cultivation and the analytical part is fairly advanced, a lack of appropriate modelling software solutions for all modelling aspects in flux studies is limiting the application of metabolic flux analysis.

We have developed OpenFLUX as a user friendly, yet flexible software application for small and large scale 13C metabolic flux analysis. The application is based on the new Elementary Metabolite Unit (EMU) framework significantly enhancing computation speed for flux calculation. From simple notation of metabolic reaction networks defined in a spreadsheet, the OpenFLUX parser automatically generates MATLAB-readable metabolite and isotopomer balances, thus strongly facilitating model creation. The model can be used to perform experimental design, parameter estimation and sensitivity analysis either using the built-in gradient-based search or Monte Carlo algorithms or in user-defined algorithms. Exemplified for a microbial flux study OpenFLUX allowed in a very user-friendly fashion to automatically compile the EMU-based model from an Excel file containing metabolic reactions and carbon transfer mechanisms. It reliably reproduced the published data and optimum flux distributions for the network under study were found quickly (<20 sec).

OpenFLUX is a fast, accurate application to perform steady-state 13C metabolic flux analysis. It will strongly facilitate and enhance the design, calculation and interpretation of metabolic flux studies. By providing the software open source, we hope it will evolve with the rapidly growing field of fluxomics.


COMPARING RESULTS OBTAINED FROM THE PATHWAY ACTIVITY PROFILING (PAPI) ALGORITHM WITH 13C-BASED METABOLIC FLUX ANALYSIS.

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Metabolomics has gained increased popularity in the last 10 years. This popularity comes from its use as a functional genomics tool and its diverse range of potential applications. However, metabolomics data sets are usually complex, difficult to interpret and challenging to correlate. The Pathway Activity Profiling (PAPI) (1) is a method developed in our group to correlate metabolite levels with potential metabolic pathways likely to be active inside the cells. This method uses the number of metabolites detected from each metabolic pathway and their relative abundances in the samples to predict the activity of different pathways. PAPI has been recently published and became very popular. However, PAPI consider that pathways which intermediate are found at lower intracellular levels are down-regulated compared to pathways with intermediates accumulating inside the cells. This assumption has been controversial, and, therefore, we propose to compare the results generated by PAPI with metabolic flux analysis of two different microorganisms: Escherichia coli and Enterococcus faecalis. Based in the fraction of 13C-labelling distribution in the biomass, we estimated the flux through different metabolic pathways from the central carbon metabolism of these bacteria when growing under different environmental conditions. Metabolic flux analysis is the ultimate measurement of metabolic pathway activity, and should answer the question of whether or not the intracellular metabolic intermediates of a pathway accumulate when down-regulated.