In silico analyses of interactions between systems objectives for engineering of biological communities

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Project Goals: Genome-scale models of metabolism have been developed for a wide-range of different organisms with different modes of metabolism and growing in a diverse set of environments. Using Flux Balance analysis (FBA) to assess the metabolic phenotypes of these systems has been very successful. However, as the quantity and quality of in situ data for complex biological systems improve, we have the opportunity to assess the trade-offs among various biological objectives of systems and predict deviations from the maximum growth paradigm that has dominated nearly all FBA analyses. To this end, we are developing a high-performance computing tool for Multi-Objective Flux Analysis (MOFA) of biological systems. This mode of analysis becomes particularly important for studying multi-cellular communities as well as engineering of systems to produce compounds of interests.

Genome-scale models of metabolism have become a standard tool for analyzing metabolic capabilities of biological systems. Examination of the models with Flux Balance Analysis[1] (FBA) has informed us about metabolic activities of individual organisms as well as general truths about universal metabolic characteristics. For FBA, available annotated genomic information about a system is used to develop a system-level reconstruction of the metabolic network. The model is constrained using fundamental physico-chemical principles, experimental observations, and an assumption that the system is at steady state. Linear programming is then used to optimize for a metabolic flux pattern that results in the optimum value of a biological function (typically growth).

While FBA models have proven very useful in identifying factors that are critical for optimum cellular growth (or other biological objectives) under well defined environmental situations, the necessity to optimize only one objective limits the use of this method for analysis of multi-cellular communities or examining the trade-offs among competing cellular objectives.

Although a number of FBA-based methods have been developed to study inter-cellular interactions (*e.g.*, COMETS[2] and OPTCOM[3]), none of them systematically examines the tradeoffs among the system's objectives. This knowledge about how an alteration in activity of a cellular process would affect other important biological processes is particularly crucial for design of metabolic engineering projects that aim to enhance a cellular activity. The results can also be used to inform trait-based models of multi-cellular communities[3]. This type of examination requires development of genome-scale multi-objective flux analysis (MOFA) models that would account for the different metabolic objectives in a system or for each member of a community**.**

Multi-objective optimization (MO) is a critical tool in a number of fields where a decision maker needs to consider tradeoffs between various conflicting objectives. Simulating genome-scale MOFA models of metabolism requires use of high performance parallel computing. LLNL's

extensive computational capabilities allow us to develop tools that would permit such undertakings.

As a test case, we used MOFA to examine the diverse metabolic phenotypes of purple non-sulfur bacterium, *Rhodopseudomonas palustris* (RP). We have developed a highly curated genome-scale model of RP's metabolism and constrained it with a large volume of experimental measurements. While FBA analyses provided us with information about which metabolic pathways were used for carbon fixation, hydrogen production, as well as proton economy of RP for a variety of different nutrient sources, MOFA analyses informed us that under light-anaerobic conditions the observed metabolic behaviors are not in agreement with FBA's central paradigm of cells growing at maximum feasible rates.

Our MOFA analyses show that in light-anaerobic conditions RP grows at slightly lower than theoretical maximum rates. Mapping flux measurements onto the MOFA predicted multi-dimensional Pareto front show that: 1) growth under these conditions is limited by the amount of light the cell absorbs; 2) depending on the nutrient-source, proton metabolism of the cell can effect the rate of growth; 3) RP's metabolism is geared toward optimum carbon efficiency and not maximum rate of growth; 4) hydrogen production by RP results in reduced rate of carbon fixation and cellular growth.

References

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