Metabolic Flux Balance Analysis: MATLAB COBRA Toolbox

Brandon Palomo

Flux Balance Analysis

Flux balance analysis (FBA) is used to calculate the flow of metabolites through a metabolic network to study genome-scale metabolic network reconstructions. *In Silico* modeling of the metabolic network can predict the growth rate of an organism and the production rate of a bio-valuable product. The results can be analyzed to study the phenotype under varying cellular and environmental conditions.

Constraint Based Analysis

FBA is mathematically represented as a numerical matrix $\boldsymbol{S}$ of stoichiometric coefficients for all reactions involved in the model. The stoichiometries impose mass balance constraints on the system at steady state. The flux through all the reactions in the model are represented by vector $\boldsymbol{v}$ and the concentrations of the metabolizes are represented by vector $\boldsymbol{x}$.

$$\boldsymbol{S} \cdot \boldsymbol{v} = \frac{d\boldsymbol{x}}{dt} = \boldsymbol{0}$$

Additional constraints on the system include defining upper and lower bounds for the allowable fluxes of every reaction. To optimize a phenotype, the constraints must be studied with an objective function (e.g. biomass production).

Phenotype Optimization

The optimization of a phenotype using FBA can be determined using the Constraint Based Reconstruction and Analysis (COBRA) Toolbox in MATLAB. Representing biomass production as the objective function Z allows for the prediction of the maximum growth rate by calculating the maximum flux allowed through the biomass reaction. The objective function is calculated as $Z = \boldsymbol{c}^T \cdot \boldsymbol{v}$ where $\boldsymbol{c}$ is a vector of weights. As an example, arginine biosynthesis was coupled to biomass production in a genome-scale model of *E. coli* k-12 W3110 from the Biochemical Genetic and Genomic BiGG Database. Linear programming is used to calculated flux distribution.

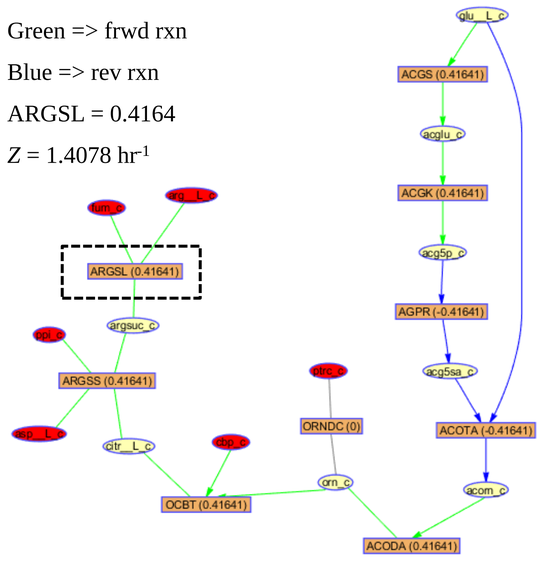
Arginine Biosynthesis Flux

Green => frwd rxn

Blue => rev rxn

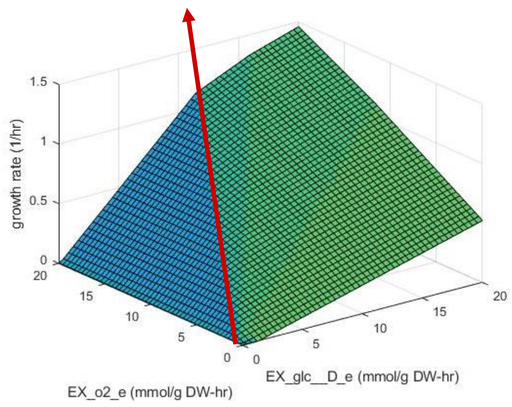
ARGSL = 0.4164

Z = 1.4078 hr^{-1}



Phase Plane Analysis

Line of Optimality



Arginine Biosynthesis Conditions

The biosynthesis of arginine in an *in silico* model of *E. coli* k-12 W3110 was determined under aerobic conditions using an uptake exchange reaction of 18.5 mmol/gDW/hr for glucose and 20 mmol/gDW/hr for oxygen (biologically realistic uptake rates).

Discussion

To determine how substrate uptake rates influence the phenotype of the cell, glucose and oxygen uptake reaction rates were varied simultaneously with respect to the biomass production rate. The results reveal the line of optimality that determines the optimal phenotype for bio-production of arginine without limitation on substrate availability as shown in the Figure 2. In Figure 1, the biomass production rate was determined from FBA to be 1.4078 hr^{-1}, which means the *E.* coli cells are predicted to double every 29.54 minutes.

Future Directions

To improve the yield of arginine, the flux thorough precursor reactions will be improved by evaluation of gene knockout designs and robustness analysis. Additional reactions may be added into the network to simulate synthetic metabolic networks and flux variability analysis will be explored to final alternative optimal solutions.

References

Orth, J.D., Thiele, I. & Palsson, B.O. What is flux balance analysis? Nat. Biotech. 28, 245–248 (2010)