479b Cybernetic Modeling Approach for Analysis and Redesign of Biochemical Pathways

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Unlike stoichiometric models of metabolic networks, cybernetic models incorporate kinetic information and are therefore fully dynamic and fully predictive. They do not overlook the fact that the flux state of the cell is ultimately determined by the levels of pathway enzymes and metabolic intermediates, and that perturbations to these intracellular variables will necessarily influence the rates of substrate uptake and product formation. Furthermore, they describe the coupling between metabolic fluxes and environmental conditions, e.g. substrate levels or dilution rate, in a way that allows the cell model to be easily integrated within a reactor model for purposes of process optimization and control. The ability to describe complex dynamic phenomena such as steady-state multiplicity, oscillatory behavior, unbalanced growth, and futile cycling is an additional advantage of cybernetic models vis-à-vis stoichiometric models.

This presentation will discuss the application of cybernetic models to relevant metabolic engineering problems. Specifically, we show how simulation and analysis of cybernetic models can provide key insights to guide the process of recombinant strain design. These models are capable of predicting not only the effects of gene knockouts, but also targeted overexpression and attenuation of genes. Furthermore, the models can describe the effects of deregulating enzyme activity controls, direct feeding of metabolic precursors, and augmenting the reaction network via gene additions. Sensitivity analysis is applied to study the effects of these various pathway manipulations, which can be leveraged to compute relevant coefficients within the Metabolic Control Analysis (MCA) framework. The ultimate application of the models, however, is to couple dynamic simulations with global search algorithms that attempt to identify a collection of pathway manipulations and/or operational parameters that gives optimal performance in reference to a particular engineering objective. We present computational results along with experimental data aimed at improving the productivity of fermentation products in *E. coli*. It should be noted, however, that the methodology is general, and its application to alternate organisms and/or biotechnological products may also be presented.