PoLiMeR tutorial Metabolic Control Analysis

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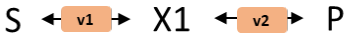
Assignment 1: A minimal model - Principles of metabolic control analysis

For this assignment you will need to construct a simple model, consisting of two reactions with reversible Michaelis-Menten kinetics.



Reversible Michaelis-Menten looks like this:

$$v_1 = \frac{\frac{V_{f\_1}}{K_{MS\_1}} \cdot \left([S] - [X1]/K_{eq\_1}\right)}{1 + [S]/K_{MS\_1} + [X1]/K_{MX1\_1}}$$

$$v_2 = \frac{\frac{V_{f\_2}}{K_{MX1\_2}} \cdot \left([X1] - [P]/K_{eq\_1}\right)}{1 + [X1]/K_{MX1\_2} + [P]/K_{MP\_2}}$$

I gave it the following parameters, but you can play with other values:

[S] = 1 mM; [P] = 0 mM; $[X1]_{initial}$ = 0 mM

V_{f_1} = 10 mM min^{-1}; K_{S_1} = 0.1 mM; K_{X1_1} = 0.5 mM; K_{eq_1} = 25 (dimensionless)

V_{f_2} = 5 mM min^{-1}; K_{X1_2} =1 mM; K_{P_2} = 1 mM; K_{eq_2} = 1 (dimensionless)

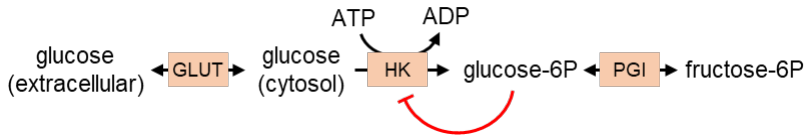
a. Build the model and calculate the steady-state concentration of X1 and the flux through reaction 1 and 2.
b. Calculate the flux control coefficients. Which enzyme exerts the highest control over the steady-state flux?
c. Check your answer under b by plotting the flux first as a function of V_{f_1}, then as a function of V_{f_2}. Are the results in agreement with the flux control coefficients?
d. Calculate the elasticity coefficients. Can you explain why one is positive and the other negative?
e. Which enzyme has the highest absolute elasticity towards metabolite X_1? Is the result in agreement with the flux control coefficients? Does the connectivity theorem hold?

Assignment 2: A minimal model – Control properties can change

a. Vary the maximum velocity of the first enzyme (V_{f_1}). What happens to the flux control coefficients if you increase V_{f_1}? And what if you decrease it? Can you explain this?
b. What happens to the elasticity coefficients if you increase V_{f_1}? Can you explain this?
c. Now reset V_{f_1} to its original value. Then vary, one by one, the K_M of enzyme 1 and 2 towards the metabolite X_1. What is the effect on the elasticity coefficients? And how does this change the flux control coefficients? Can you explain what you see?
d. You will probably find that the changes in the elasticity coefficients are not always easy to explain. The reason is that the change of a parameter (e.g. a K_M) causes changes in the variables (the concentration of X1 in this case). The effect on the elasticity coefficients is therefore the resultant of both. Can you find a way to change the K_M of enzyme 2 without changing the metabolite concentration? Hint: try to find a compensatory change in enzyme 2.
e. Now alter the K_M of enzyme 2 and make the compensatory change. Can you explain the effect of the K_M on the elasticity coefficients? And on the control coefficients?

Assignment 3 – Feedback inhibition in glycolysis

For this assignment, you will use model B of the introductory course. This is a model of the first reactions of glycolysis. It includes the feedback inhibition of hexokinase (HK) by glucose 6-phosphate (G6P), which is found in mammalian cells.



a. Do you remember how to increase or decrease the strength of the feedback inhibition?
b. Vary the inhibition constant (K_i) of hexokinase towards G6P and calculate the flux control coefficients and the elasticity coefficients. In which range of K_i values do you see an effect?
c. Can you explain the effect of the K_i on the control and elasticity coefficients? Hint: it may be helpful to consider this as if it were a two enzyme problem with only one intermediate, i.e. a supply and demand block around G6P. Or around cytosolic glucose (Gluc_c).
d. To study the effect of the feedback strength via the elasticity of HK towards G6P alone, i.e. at constant concentrations of the metabolites, find a compensatory change in one of the parameters of HK. What is the effect on the flux and on the control properties?