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Branched and Cyclic Systems

12.1 Branched Pathways

Branching structures in metabolism are probably one of the most common metabolic patterns. Even a pathway such as glycolysis, often depicted as a straight chain in textbooks is in fact a highly branched pathway.

A linear perturbation analysis of a branched pathway can reveal some interesting potential behavior. Consider the following simple branched pathway:

where J_i are the steady state fluxes. By the law of conservation of mass, at steady state, the fluxes in each limb are governed by the relationship:

$$J_1 - (J_2 + J_3) = 0$$

In terms of control theory, there will be four sets of control coefficients, one concerned with changes in the intermediate, S, and three sets corresponding to each of the individual fluxes.

Let the fraction of flux through J_2 be given by $\alpha = J_2/J_1$ and the fraction

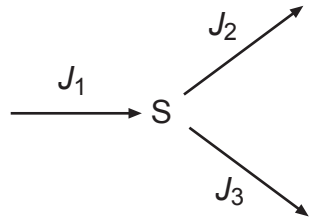

Figure 12.1: Simple branched pathway. This pathway has three different fluxes, J_1, J_2, and J_3 which at steady state are constrained by $J_1 = J_2 + J_3$.

of flux through J_3 be $1-\alpha = J_3/J_1$. The flux control coefficients for step two and three can be derived and shown to be equal to [15]:

$$C_{E_2}^{J_2} = \frac{\varepsilon_1 - \varepsilon_3(1-\alpha)}{\varepsilon_1 - \varepsilon_2\alpha - \varepsilon_3(1-\alpha)} > 0$$

$$C_{E_3}^{J_2} = \frac{\varepsilon_2(1-\alpha)}{\varepsilon_1 - \varepsilon_2\alpha - \varepsilon_3(1-\alpha)} < 0$$

Note that the flux control coefficient, $C_{E_3}^{J_2}$ is negative, indicating that changes in the activity of E_3 decrease the flux in the other limb. To understand the properties of a branched system it is instructive to look at different flux distributions. For example consider the case when the bulk of flux moves down J_3 and only a small amount goes through the upper limb J_2, that is $\alpha \to 0$ and $1-\alpha \to 1$ (See Figure 12.2(b)). Let us examine how the small amount of flux through J_2 is influenced by the two branch limbs, E_2 and E_3.

$$C_{E_2}^{J_2} \to \frac{\varepsilon_1 - \varepsilon_3}{\varepsilon_1 - \varepsilon_3} = 1$$

$$C_{E_3}^{J_2} \to \frac{\varepsilon_2}{\varepsilon_1 - \varepsilon_3}$$

The first thing to note is that E_2 tends to have proportional influence over its own flux. Since J_2 only carries a very small amount of flux, any changes

in E_2 will have little effect on S, hence the flux through E_2 is almost entirely governed by the activity of E_2. Because of the flux summation theorem and the fact that $C_{E_2}^{J_2} = 1$ means that the remaining two coefficients must be equal and opposite in value. Since $C_{E_3}^{J_2}$ is negative, $C_{E_1}^{J_2}$ must be positive. Unlike a linear chain, the values for $C_{E_2}^{J_2}$ and $C_{E_1}^{J_2}$ are not bounded between zero and one and depending on the values of the elasticities it is possible for the control coefficients to greatly exceed one [27, 34]. It is conceivable to arrange the kinetic constants so that every step in the branch has a control coefficient of unity (one of which must be -1). Using the old terminology, we would conclude from this that every step in the pathway is the rate limiting step.

Let us now consider the other extreme, when most of the flux is through J_2, that is $\alpha \rightarrow 1$ and $1 - \alpha \rightarrow 0$ (See Figure 12.2(a)). Under these conditions the control coefficients yield:

$$\begin{aligned} C_{E_2}^{J_2} &\rightarrow \frac{\varepsilon_1}{\varepsilon_1 - \varepsilon_2} \\ C_{E_3}^{J_2} &\rightarrow 0 \end{aligned}$$

In this situation the pathway has effectively become a simple linear chain. The influence of E_3 on J_2 is negligible. Figure 12.2 summarizes the changes in sensitivities at a branch point.

12.2 Futile or Substrate Cycles

Related to branched systems are cyclic pathways. A typical cyclic pathway in shown in Figure 12.3. For cycling to occur both forward and back reactions must operate. It is typical to find that the forward and reverse reactions are chemically distinct. Often one reaction will be driven by ATP while the other by the hydrolysis of phosphate groups. Typical examples in metabolism include the cycle between glucose and glucose-6-phosphate and the cycling between fructose-6-phosphate and fructose 1.6-bisphosphate. Such cycles have often been called futile cycles (or better substrate cycles) because of the expenditure of free energy (as ATP) without any apparent benefit. A number of suggestions have been put forward to ra-

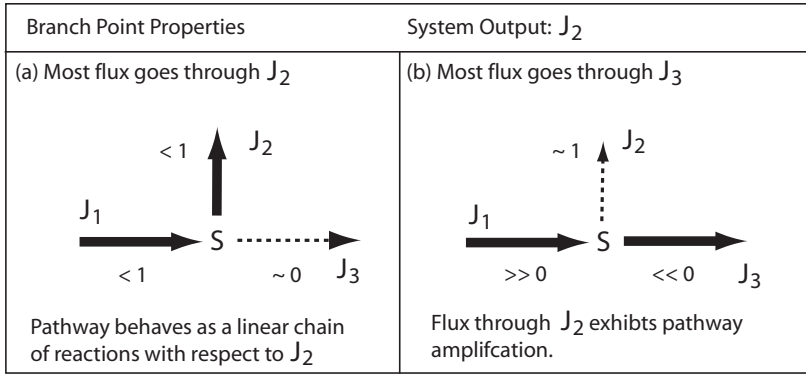

Figure 12.2: The figure shows two flux extremes relative to the flux through branch J_2. In case (a) where most of the flux goes through J_2, the branch reverts functionally to a simple linear sequence of reactions comprised of J1 and J_2. In case (b), where most of the flux goes through J_3, the flux through J_2 now becomes very sensitive to changes in activity at J_1 and J_3. Given the right kinetic settings, the flux control coefficients can become ŚultrasensitiveŠ with values greater than one (less than minus one for activity changes at J_3). The values next to each reaction indicates the flux control coefficient for the flux through J_2 with respect to activity at the reaction.

tionalize this apparent waste of energy. These include heat production, control of flux direction, metabolite buffering and more sensitive control of the net flux through the pathway. We will only consider the later here.

12.2.1 Sensitivity Control

Figure 12.3 shows a typical cyclic pathway embedded in a linear chain. Of interest is the sensitivity of the pathway flux, v_1 or v_4 to changes in v_2. The simplest assumption to make is that when we change v_2 there is no change in back flux, v_3. This could be for a number of reasons, for example v_3 is saturated by its substrate S_2. Figure 12.4 illustrate two situations, a references state in panel a) and a perturbation to v_2 shown in panel b). v_2 increases by 5% (or ten absolute flux units). Assuming that the entire

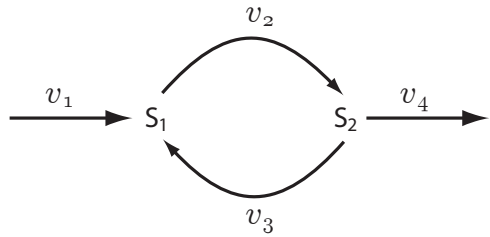

Figure 12.3: Cyclic Pathway.

flux changes appears in output flux v_4 and that v_3 is not changed, the the percentage change in v_4 (or v_1) is 100%, a twenty fold amplification.

This effect can be easily quantified as follows. First we note the flux constraint due to the cycle:

$$v_1 = v_2 - v_3$$

We then assume that a perturbation in v_2 leads to the same change in v_1, that is:

$$\delta v_2 = \delta v_1$$

We can now compute the fractional changes in v_1 and v_2 as:

$$\frac{\delta v_1}{v_1} = \frac{\delta v_2}{v_2}\frac{v_2}{v_1}$$

The degree of amplification is then given by

$$\frac{\delta v_1/v_1}{\delta v_2/v_2} = \frac{v_2}{v_1}$$

Since $v_2 = v_1 + v_3$ then

$$\frac{\delta v_1/v_1}{\delta v_2/v_2} = \frac{v_1 + v_3}{v_1} = 1 + \frac{v_3}{v_1} \tag{12.1}$$

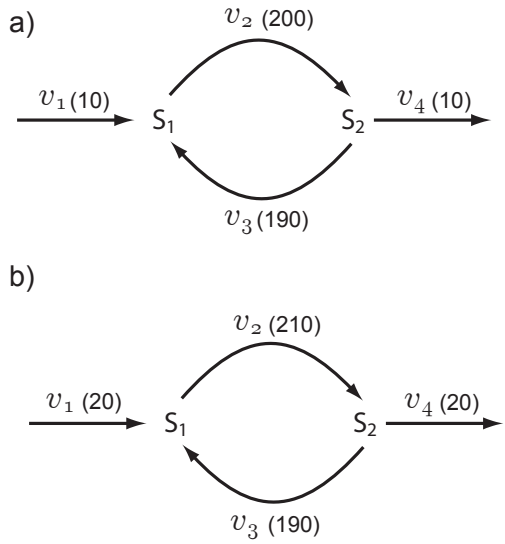

Figure 12.4: Amplification in a substrate cycle. Panel a) Reference state, values refer to fluxes at various points, note that $v_1 = v_2 - v_3$. Panel b) Activation of v_2 by 5% leads to a 100% change in v_1 and v_4. It assumes that v_3 is not activated by any changes in S_2.

This result shows that the higher the cycling rate (v_3) compared to the through flux, the greater the amplification. This equation gives us the maximum degree of amplification possible. In practice, v_3 will not remain unchanged because S_2 rises. In addition S_1 will fall due to high consumption which will reduce v_2 but increase v_1 due to lower product inhibition. The resulting amplification is therefore a more complicated function than the one suggested by equation 12.1 although it does give the maximum possible.

To carry out a more detailed analysis we must turn to metabolic control analysis. We can examine the flux control coefficient for $C_2^{J_1}$:

$$C_2^{J_1} = \frac{\varepsilon_1^1 \varepsilon_2^4 (1 + v_3/v_1)}{D}$$
$$D = \varepsilon_1^1 \varepsilon_2^4 - (1 + v_3/v_1)(\varepsilon_1^1 \varepsilon_2^2 + \varepsilon_2^4 \varepsilon_1^2) + (v_3/v_1)(\varepsilon_1^1 \varepsilon_2^3 + \varepsilon_2^4 \varepsilon_1^3)$$

Let us simplify this equation by assuming that there is little or not product inhibition from S_2 on to v_2 and S_1 on to v_3. This means that $\varepsilon_1^3 = 0$ and $\varepsilon_2^2 = 0$. If we also multiply top and bottom by v_1 and using the relation $v_1 + v_3 = v_2$, then these assumptions simplify the control equation to:

$$C_2^{J_1} = \frac{\varepsilon_1^1 \varepsilon_2^4 v_2}{D}$$
$$D = \varepsilon_1^1 \varepsilon_2^4 v_1 - \varepsilon_2^4 \varepsilon_1^2 v_2 + \varepsilon_1^1 \varepsilon_2^3 v_3$$

To things to note immediately from this equation. There must be product inhibition on the first step, ε_1^1, in order to get any sensitivity. If ε_1^1 is zero then so is $C_2^{J_1}$. This is because all control is now on the first step. This highlights again the danger of using rate laws in models that are product insensitive because the use of such rate laws often give misleading or trivial results of no real interest. The second relatively simple statement to make from the above equation is the important of ε_2^3. This elasticity is the activation of the reverse arm with respect to S_2. The larger this elasticity the smaller the degree of amplification. This is expected because any flux that flows back along the reverse cycle instead of into v_4 reduces the potential amplification factor. To analyze the equation further we can make additional simplifications.

We know that sensitivity increases when the cycling rate increases relative to the main flux, v_1 and v_4. If v_2 and v_3 are much greater than v_1 then we can simplify the equation further to:

$$C_2^{J_1} = \frac{v_2}{\varepsilon_2^3/\varepsilon_2^4 v_3 - \varepsilon_1^2/\varepsilon_1^1 v_2}$$

If the cycling rate is so high that v_2 and v_3 are almost indistinguishable then we can see that maximal sensitivity is achieved when:

$$\frac{\varepsilon_2^3}{\varepsilon_2^4} + \frac{\varepsilon_1^2}{\varepsilon_1^1} \ll 1$$

This tells us that substrate activation of v_4 by S_2 should be stronger than substrate activation of S_2 on v_3 and secondly that product inhibition of S_1 on v_1 must be stronger than substrate activation of S_1 on v_2. If we think about this in a thought experiment, these results are expected.

The requirements for amplification in substrate cycles is fairly complicated and questions remain whether real pathways use this mechanism in vivo.

12.3 Conserved Cycles

Ultrasensitivity, cascades