Metabolic Regulation and Control

Definitions

Metabolic regulation: The process by which the steady-state flow of metabolites through a pathway is maintained.

Metabolic control: The influence exerted on the enzymes of a pathway in response to an external signal in order to alter the flux of metabolites.

Relevant Terms

• ***Flux.*** A term used in metabolic control analysis to indicate the rate of a multi-component system (metabolic pathway). The term "rate" is reserved for individual components (enzyme) of a system.

• ***Regulation.*** Occurs when the system maintains some variable (*e.g.*, temperature or concentration) constant over time despite fluctuations in external conditions (linked to homeostasis).

• ***Control.*** Refers to adjusting the output of a system with time. Control as a verb implies the ability to start, stop or direct something.

• ***Metabolic Control.*** Defined as the power to change the state of metabolism in response to an external signal. It is measurable in terms of the strength of the metabolic response to external factors, without any assumption about the function/purpose/mechanism of the response.

Reasons to control metabolic flow

1. To provide products at the rate they are needed, that is, to balance supply with demand

2. To maintain the steady-state concentrations of intermediates in a pathway within a narrow range (homeostasis)

Why is metabolic homeostasis important?

1. In an open system such as metabolism, the steady state is the state of maximum thermodynamic efficiency.

2. Many intermediates participate in more than one pathway, so that changing their concentrations may disturb a delicate balance.

3. The rate at which a pathway can respond to a control signal slows if large changes in intermediate concentrations are involved.

4. Large changes in intermediate concentrations may have deleterious effects on cellular osmotic properties.

For each step in a metabolic pathway:

J = flux of metabolites = v_f - v_r
v_f = rate of forward reaction
v_r = rate of reverse reaction

If the reaction is at equilibrium, then $J = 0$.

If the reaction is far from equilibrium, $v_f >>> v_r$ and $J \cong v_f$.

The flux throughout a steady-state pathway is constant and is determined by the pathway's rate-determining step (or steps).

Control of flux through a metabolic pathway requires:

1) that the flux vary in response to the organism's metabolic requirements; and
2) that a change in flux be communicated throughout the pathway to maintain a steady state.

Concept of rate-limiting step; regulator enzymes (allosteric) that are non-covalently or covalently regulated.

Key Questions are:

1) Are the regulatory enzymes truly rate-limiting for the pathway?

2) Is there only one rate-limiting step, or are there more than one in the pathway?

3) Does controlling these enzymes really control the flux of metabolites through the pathway or is the function of feedback inhibition really to maintain a steady state?

Metabolic Control Analysis (MCA)

(Discussion taken from Voet/Voet, pp. 619-630)

MCA makes no assumption that only one step is rate-limiting in a metabolic pathway.

MCA defines a **flux control coefficient**, C^J, to measure the sensitivity of flux to a change in enzyme concentration.

$$C^J = \frac{\delta J/J}{\delta\,[E]/[E]} = \frac{\delta \ln J}{\delta \ln [E]} \approx \frac{\Delta J/J}{\Delta\,[E]/[E]}$$

$C^{1.0}$ = doubling [E] doubles the flux through the pathway
(analogous to a reaction that is 1st-order in [S])

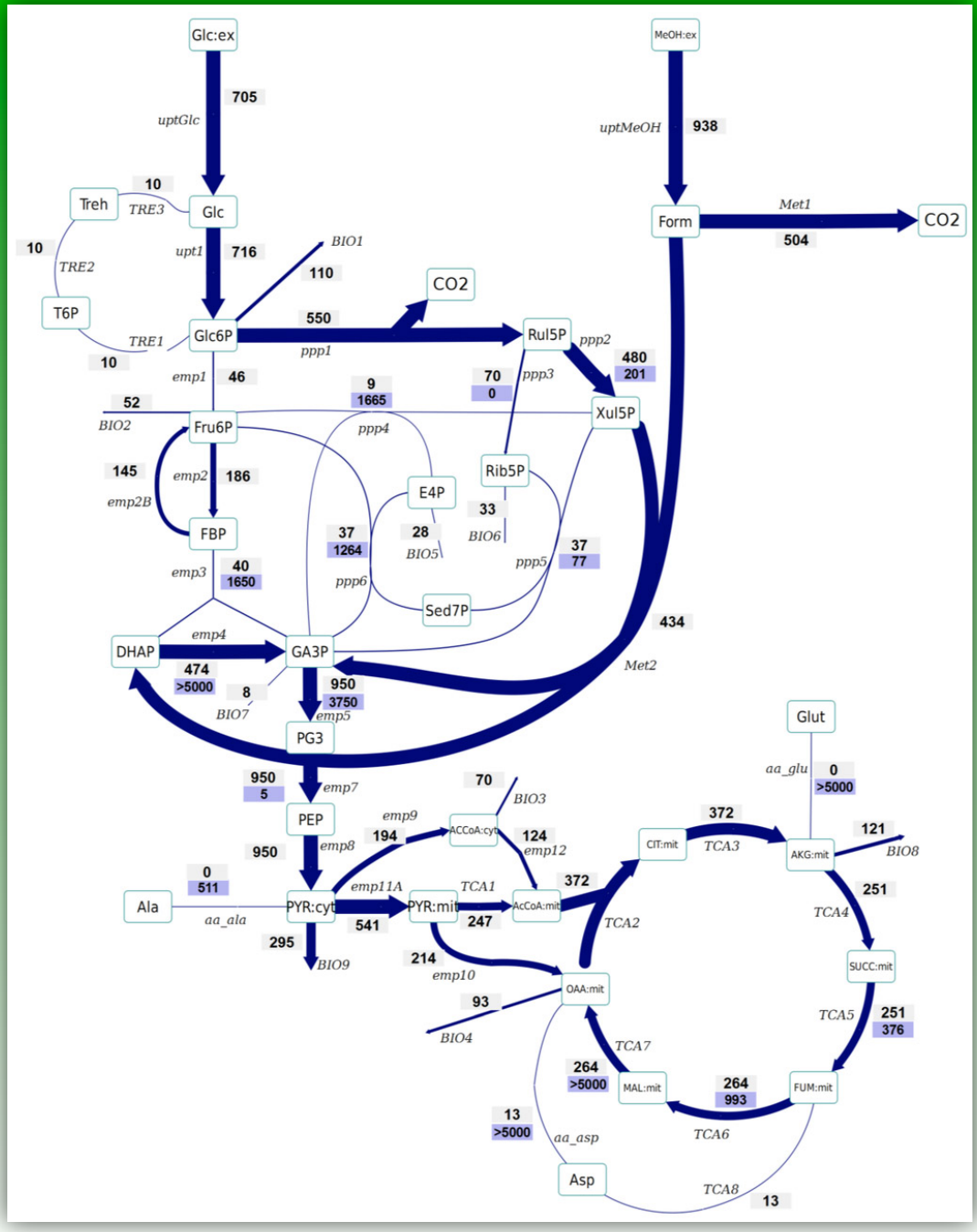
$C^{0.0}$ = the flux is insensitive to [E]
(analogous to a reaction that is zero-order in [S])

J = 0-1; most often has intermediate values (*e.g.*, if a 10% increase in [E] increases the flux by 7.5%, then J = 0.075/0.10 = 0.75)

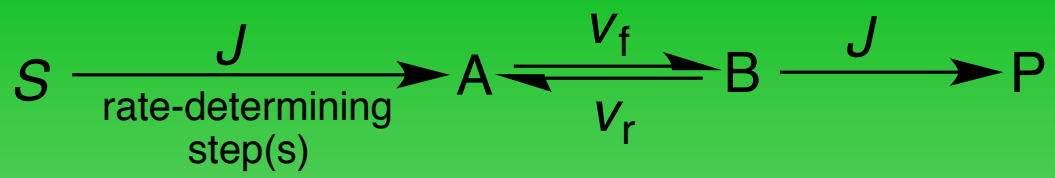
The flux control coefficient for each of the participating enzymes in a metabolic pathway is the fraction of the total control on the pathway exerted by that enzyme.

The sum of all flux control coefficients involved in controlling a metabolic pathway must equal 1. This is the **additivity theorem** of metabolic control. It states that the flux control coefficient of a particular enzyme in a system depends, in part, on the flux control coefficients of the other enzymes in the system, that is, an enzyme's flux control coefficient is a property of the system as a whole.

Flux control in a metabolic system is shared among all of the enzymes in the system, even though most of their flux control coefficients are likely to be small.


Glc:ex
705
uptGlc
MeOH:ex
uptMeOH
938
Treh
10
TRE3
Glc
Form
Met1
CO2
504
10
TRE2
upt1
716
BIO1
110
T6P
CO2
TRE1
Glc6P
550
ppp1
Rul5P
ppp2
480
201
10
emp1
46
9
1665
70
0
ppp3
52
BIO2
Fru6P
ppp4
Xul5P
145
emp2
186
Rib5P
emp2B
E4P
33
FBP
37
1264
28
BIO5
BIO6
37
77
ppp5
emp3
40
1650
ppp6
Sed7P
434
emp4
DHAP
GA3P
Met2
474
>5000
8
BIO7
950
3750
emp5
PG3
Glut
aa_glu
0
>5000
950
5
emp7
70
BIO3
PEP
emp9
194
ACCoA:cyt
124
emp12
950
emp8
372
121
CIT:mit
TCA3
AKG:mit
BIO8
0
511
Ala
aa_ala
PYR:cyt
emp11A
541
PYR:mit
TCA1
AcCoA:mit
247
372
TCA2
251
TCA4
295
BIO9
214
emp10
SUCC:mit
OAA:mit
93
BIO4
TCA5
251
376
TCA7
264
>5000
MAL:mit
264
993
FUM:mit
13
>5000
TCA6
aa_asp
Asp
TCA8
13

Consider the following steady-state pathway:



An increase in J causes an increase in [A], which in turn causes an increase in v_f. The amount of increase in [A] (denoted Δ[A]) that causes v_f to increase the appropriate amount (denoted Δv_f) is determined as follows:

$$\Delta J = \Delta v_f$$

After algebraic manipulation, we get:

$$\frac{\Delta J}{J} = \frac{\Delta v_f}{v_f}\,\frac{v_f}{J} = \frac{\Delta v_f}{v_f}\,\frac{v_f}{(v_f - v_r)}$$

This equation relates the fractional change in flux through the rate-determining step(s) to the fractional change in v_f, the forward rate of the next reaction in the pathway.

The relationship between substrate concentration and the rate of an enzymatic reaction is expressed by the Michaelis-Menten equation:

$$v_f = \frac{V_{max}^{f}\ [A]}{K_M + [A]}$$

When [A] <<< K_M, then:

$$v_f = \frac{V_{max}^{f}\ [A]}{K_M}$$

and

$$\Delta v_f = \frac{V_{max}^{f} \Delta [A]}{K_M}$$

Hence:

$$\frac{\Delta v_f}{v_f} = \frac{\Delta [A]}{[A]}$$

The fractional change in the forward rate is equal to the fractional change in substrate concentration. Upon further substitution, we get:

$$\frac{\Delta J}{J} = \frac{\Delta [A]}{[A]} \frac{v_f}{(v_f - v_r)}$$

This equation relates the fractional change in flux through a metabolic pathway's rate-determining step(s) to the fractional change in substrate concentration necessary to communicate that change to the following reaction steps. The quantity $v_f/(v_f-v_r)$ is a measure of the **sensitivity** of a reaction's fractional change in flux to its fractional change in substrate concentration. It is also a measure of the reversibility of the reaction (how close it is to equilibrium).

Key Insights

1. For an irreversible reaction, v_r approaches 0 (relative to v_f) and $v_f/(v_f-v_r)$ approaches 1. The reaction thus requires a nearly equal fractional increase in substrate concentration in order to respond to a fractional increase in flux.

2. As a reaction approaches equilibrium, v_r approaches v_f and $v_f/(v_f-v_r)$ approaches infinity. The reaction's response to a fractional increase in flux thus requires a much smaller fractional increase in its substrate concentration.

The ability of a reaction to communicate a change in flux increases as the reaction approaches equilibrium.

The ratio $v_f/(v_f-v_r)$ is called the **elasticity coefficient**, ε, in metabolic control analysis. It is the fractional change in the net rate of an enzyme reaction, v, with respect to the fractional change in substrate concentration [A].

$$\varepsilon = \frac{\delta v/v}{\delta\,[A]/[A]} = \frac{\delta \ln v}{\delta \ln [A]} \approx \frac{v_f}{(v_f - v_r)}$$

For an enzyme functioning far from equilibrium: $v_f >> v_r$ and changing substrate concentration has only a small effect on the net rate of the enzyme reaction (ε is close to 1).

For an enzyme functioning close to equilibrium: v_f and v_r are much faster than the overall net rate, ε approaches infinity and only a tiny change in substrate concentration is needed to adjust to a new flux. *Large elasticity coefficients are associated with maintaining homeostasis.*