Properties of the Steady State

Sensitivity Analysis
“Metabolic Control Analysis”

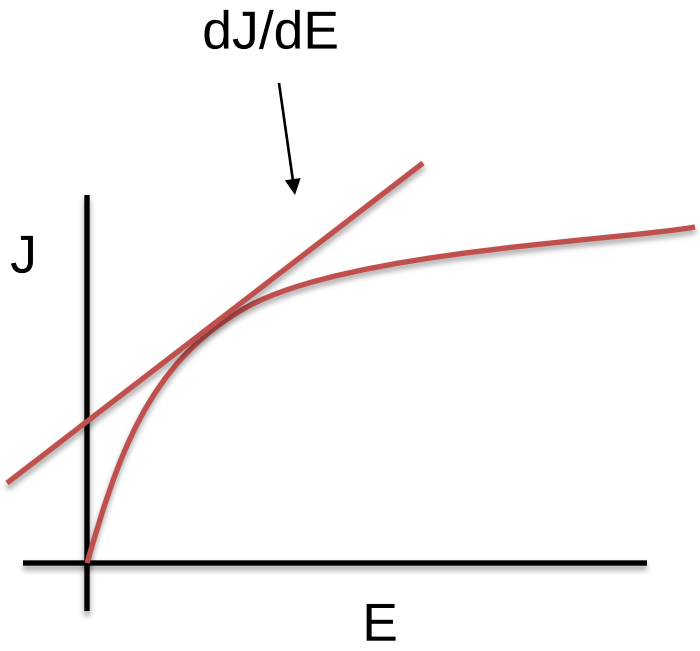
Flux and Concentation Control Coefficients:

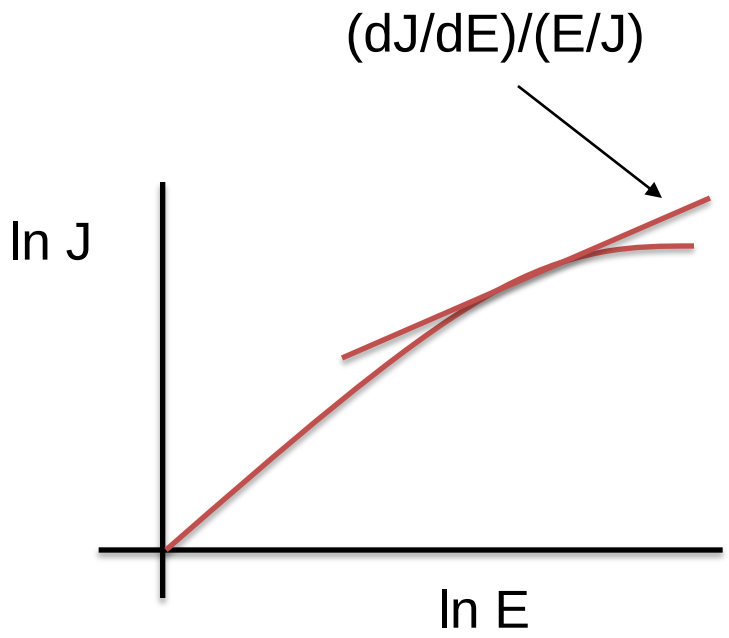
$$C_{E_i}^{J} = \frac{dJ}{dE_i}\frac{E_i}{J} = \frac{d\ln J}{d\ln E_i} \approx J\%/E_i\%$$

$$C_{E_i}^{S_j} = \frac{dS_j J}{dE_i}\frac{E_i}{S_j} = \frac{d\ln S_j}{d\ln E_i} \approx S_j\%/E_i\%$$

Metabolic Control Analysis

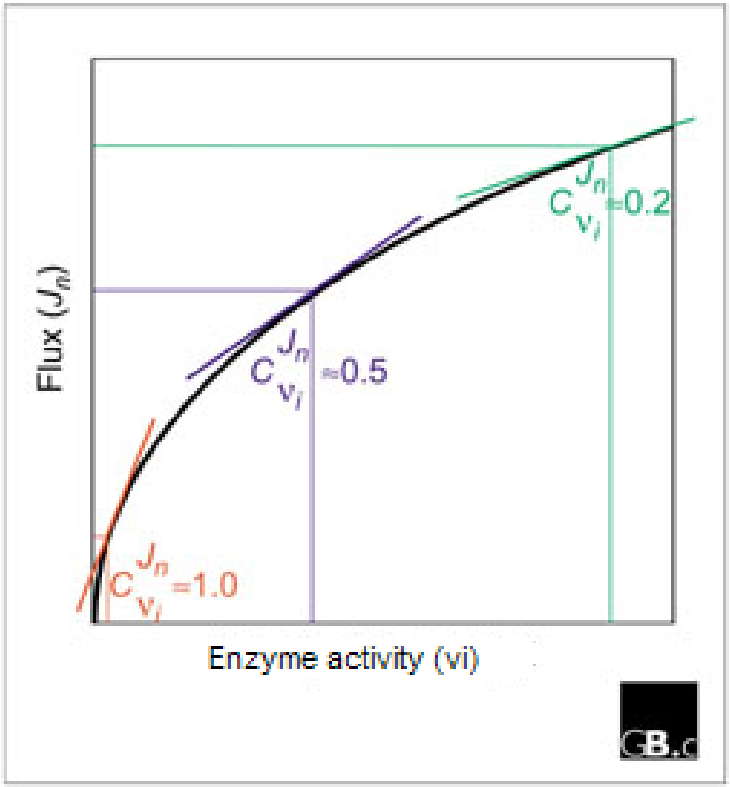
Geometrical interpretation:





Metabolic Control Analysis

Geometrical interpretation:



Flux control coefficients (C) for typical variations in pathway flux (J) measured at step n with a steady-state rate (v) at step *i* of a pathway. The coefficients are equal to the slope of the tangent to the curve (shown) multiplied by the scaling term v_i/J_n. This figure is adapted http://genomebiology.com/2000/1/6/reviews/1031

Metabolic Control Analysis

Some characteristics of flux control coefficients.

In a linear metabolic network, the value of any particular flux control coefficient is bounded between zero and one.

$$0 \leq C_E^J \leq 1$$

This condition applies to a linear chain

Metabolic Control Analysis

How can you measure control coefficients?

1. Changing gene expression and measuring the effect on the system.
2. Using inhibitors to change an enzyme's activity and measuring the effect on the system.
3. Building a computer model and getting the computer to compute the coefficients

Metabolic Control Analysis

What about some real values?

The following information was taken from a paper by S. Thomas et al, Biochemical Journal, 1997, 322, 119-127

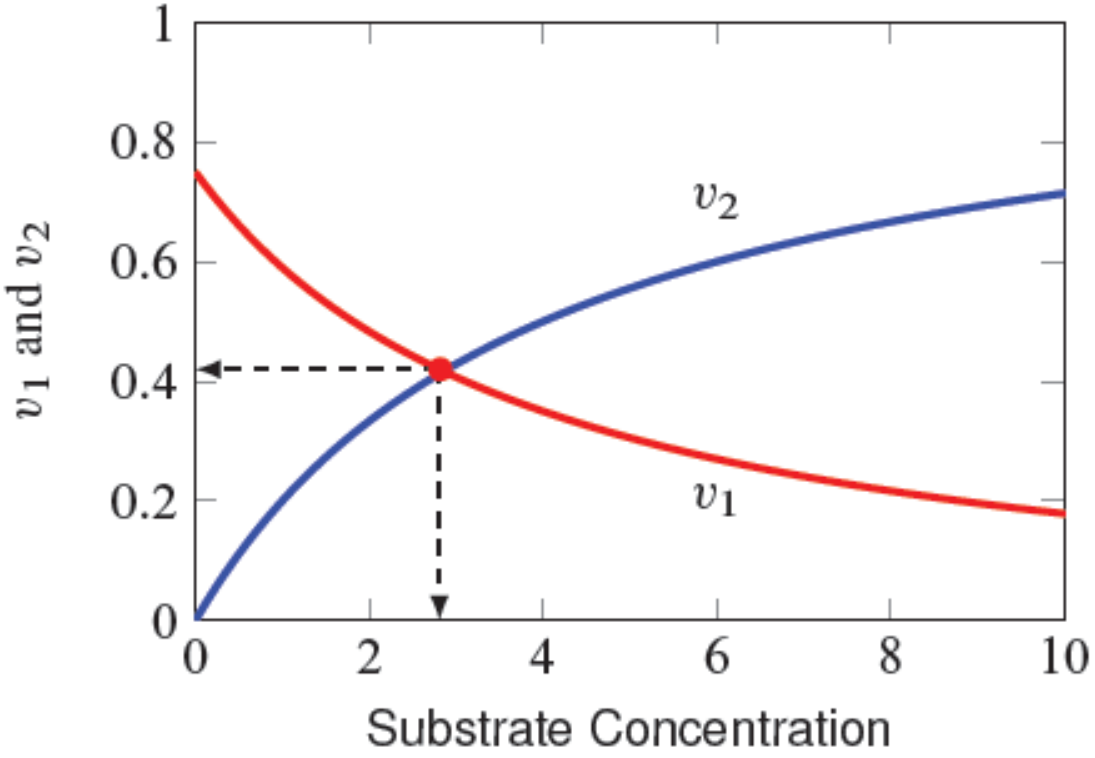
Glycolysis in tuber tissue of potato.

Values computed using a combination of calculation and experimental work.

Enzyme	Flux Control Coefficient
PGM	0.029
PGI	0.139
PFK	0.132
Aldolase	0.0
TPI	0.0
GAPDH/PGK	0.001
enolase	0.005
PK	0.702
Sum	1.008

Metabolic Control Analysis

$$X_o \xrightarrow{v_1} S \xrightarrow{v_2} X_1$$



Metabolic Control Analysis

$$X_o \xrightarrow{v_1} S \xrightarrow{v_2} X_1$$

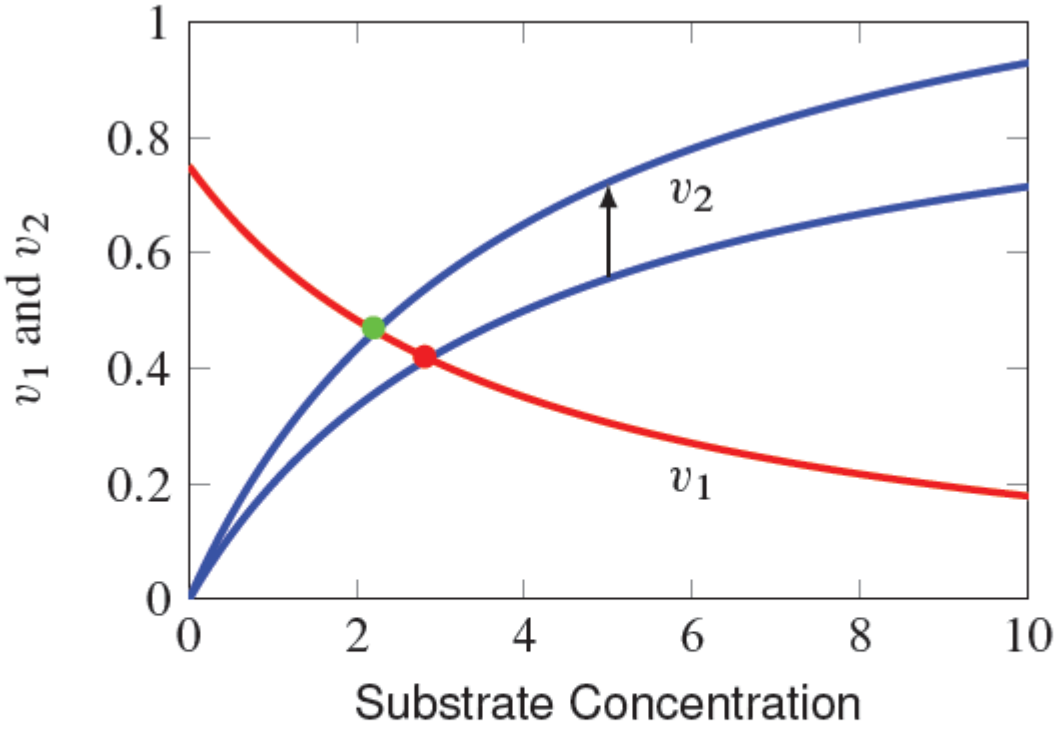


Figure 9.2: v_1 has been increased by 30% by increasing the enzyme activity on v_1. This results in a displacement of the steady state curve to the right, leading to an increase in the steady state concentration of S.

Metabolic Control Analysis

$$X_o \xrightarrow{v_1} S \xrightarrow{v_2} X_1$$

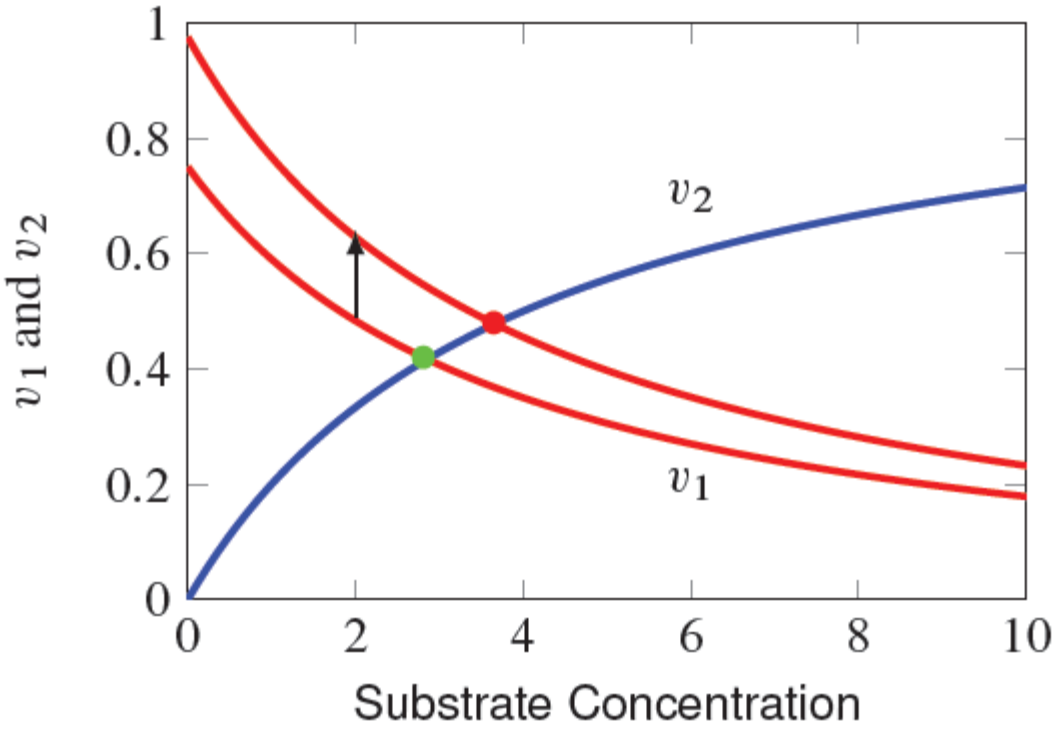


Figure 9.3: v_2 has been increased by 30% by increasing the enzyme activity on v_2. This results in a displacement of the steady state curve to the left, leading to a decrease in the steady state concentration of S.

Metabolic Control Analysis

$$X_o \xrightarrow{v_1} S \xrightarrow{v_2} X_1$$

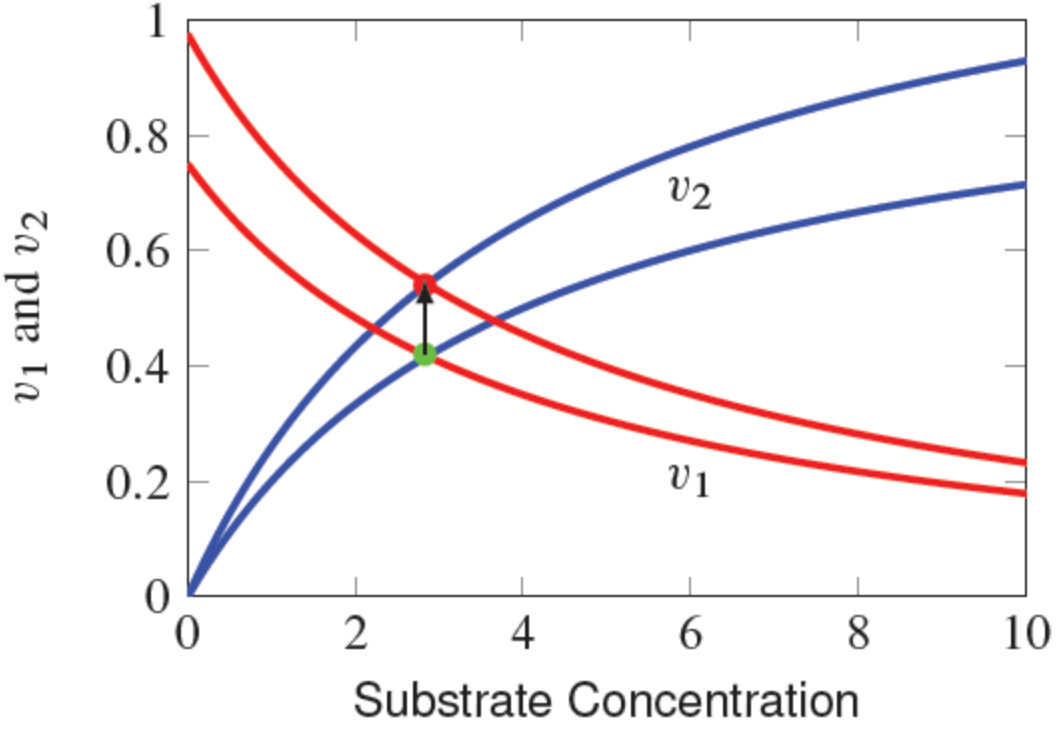


Figure 9.4: In this experiment, both v_1 and v_2 are increased by 30 %. Because both rates are increased by the same amount, the rate of change of S does not change. This means that there is no resulting change to the steady state concentration of S. The net flux through the pathway has however increased by 30 %.

Metabolic Control Analysis

$$X_o \xrightarrow{v_1} S \xrightarrow{v_2} X_1$$

Carry out a simulation to confirm the results of the previous slides.

Metabolic Control Analysis

The Summation Theorems

$$C^J_{E_1} + C^J_{E_2} + C^J_{E_3} \ldots + C^J_{E_n} = 1$$

$$C^S_{E_1} + C^S_{E_2} + C^S_{E_3} \ldots + C^S_{E_n} = 0$$

Metabolic Control Analysis

The Summation Theorems

$$C^J_{E_1} + C^J_{E_2} + C^J_{E_3} \ldots + C^J_{E_n} = 1$$

1. The summation theorem shows that the enzymes of a pathway can share the control of flux.

2. Given that n >> 1, the average value for a control coefficient must be 1/n, ie very small – most mutations are recessive

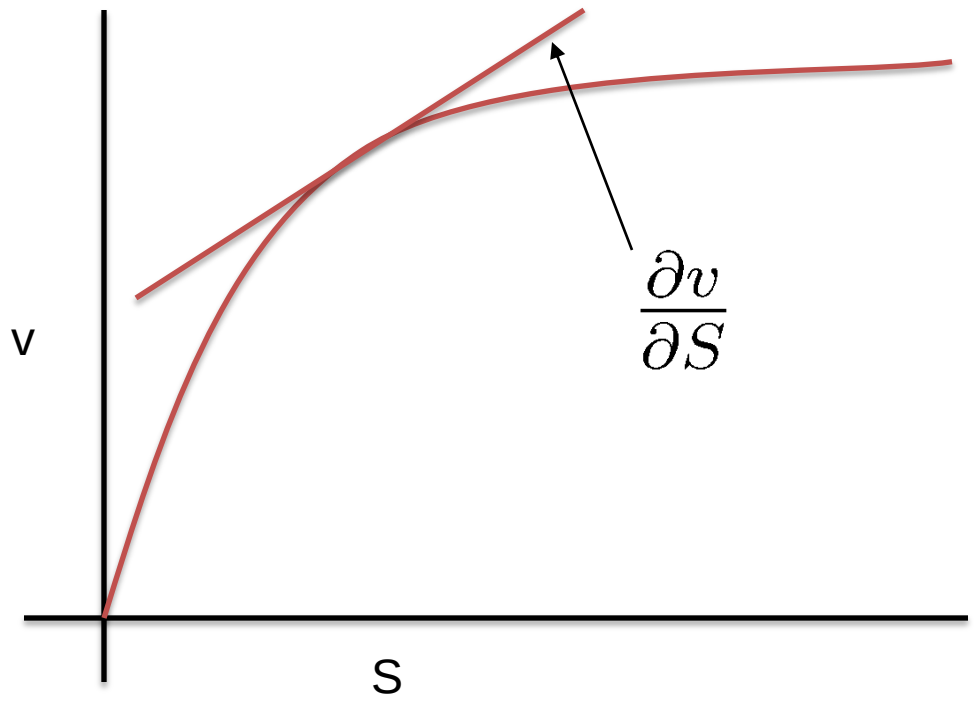
3. Changes in one control coefficient result in changes in other control coefficients. This means the a control coefficient is a **system property** and not an intrinsic property of the enzyme alone.

Metabolic Control Analysis

Carry out a simulation to show that the summation theorem is Valid

Elasticities

Michaelis-Menten Curve for an isolated enzyme

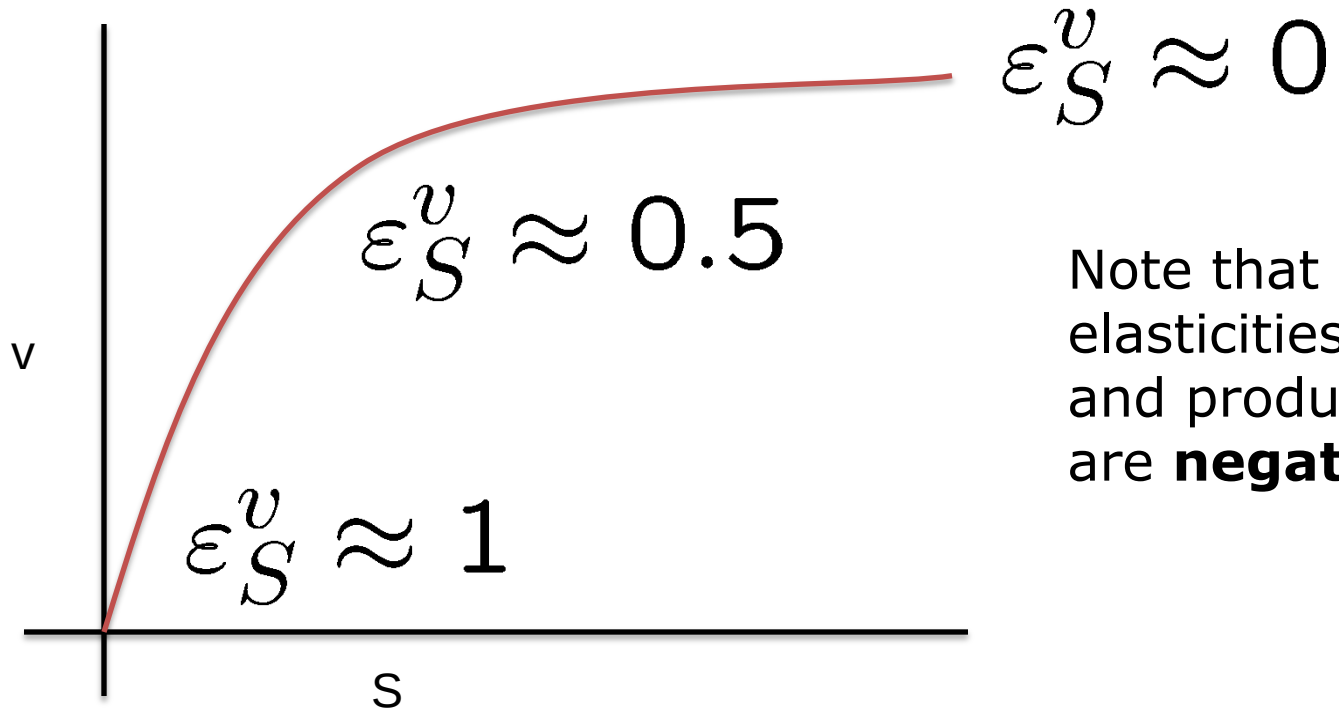


Let us define the elasticity as:

$$\varepsilon_S^v = \frac{\partial v}{\partial S}\frac{S}{v}$$

Elasticities

Michaelis-Menten Curve for an isolated enzyme



Note that substrate elasticities are **positive** and product elasticities are **negative**

Elasticities

Another way of looking at an Elasticity:

$$\frac{\delta v}{v} = \varepsilon_S^v \frac{\delta S}{S}$$

We can use an elasticity to predict the change in the rate of a reaction given a change in the substrate concentration.

Elasticities

Another way of looking at an Elasticity:

$$S_1 \longrightarrow S_2$$

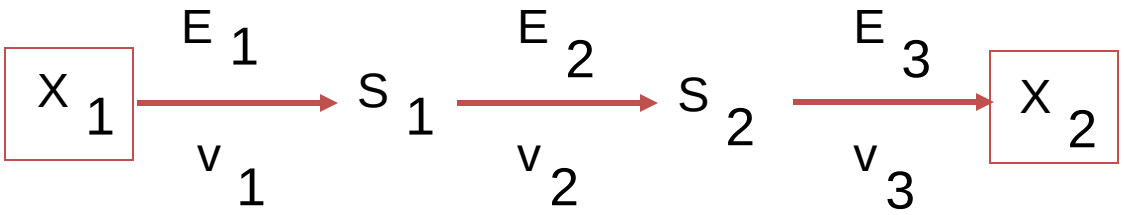
Product inhibition term

$$\frac{\delta v}{v} = \varepsilon^{v}_{S_1} \frac{\delta S_1}{S_1} + \varepsilon^{v}_{S_2} \frac{\delta S_2}{S_2}$$

< 0 !

In general if there are multiple changes happening around an enzyme we can simply sum each contribution using the appropriate elasticity.

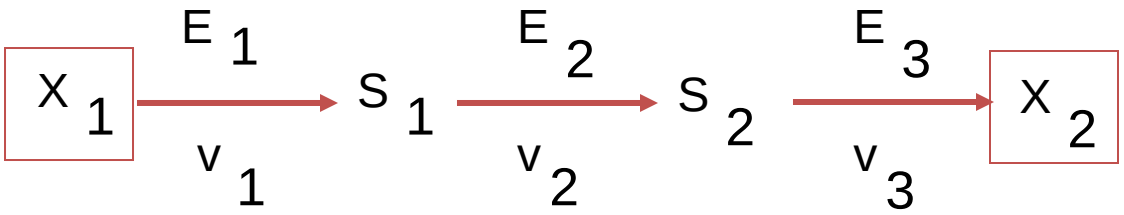
Perturbation Analysis



Increase E3:

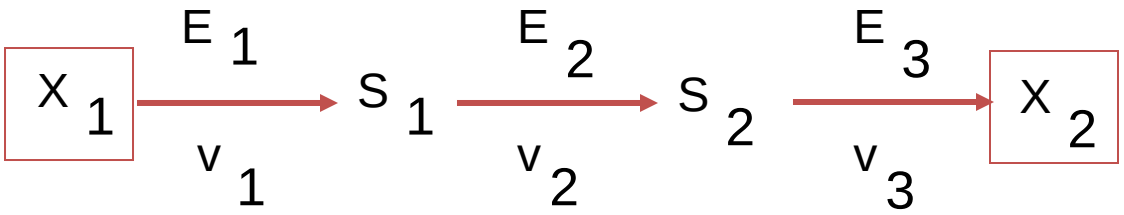
1. v3 Increases
2. S2 Decreases
3. v2 Increases
4. S1 Decreases
5. v1 Increases

Perturbation Analysis



$$\frac{dv_1}{v_1} = \varepsilon_1^1 \frac{dS_1}{S_1}$$

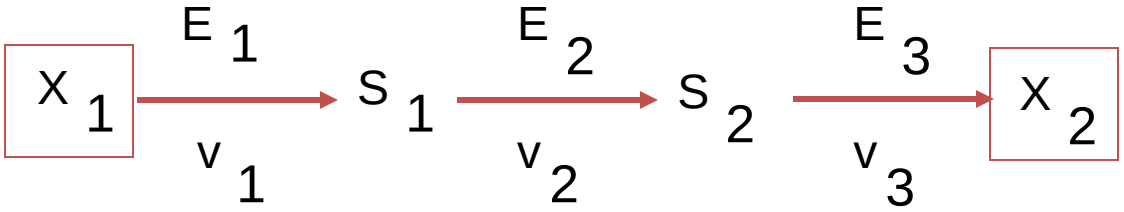
Perturbation Analysis



$$\frac{dv_1}{v_1} = \varepsilon_1^1 \frac{dS_1}{S_1}$$

$$\frac{dv_2}{v_2} = \varepsilon_1^2 \frac{dS_1}{S_1} + \varepsilon_2^2 \frac{dS_2}{S_2}$$

Perturbation Analysis

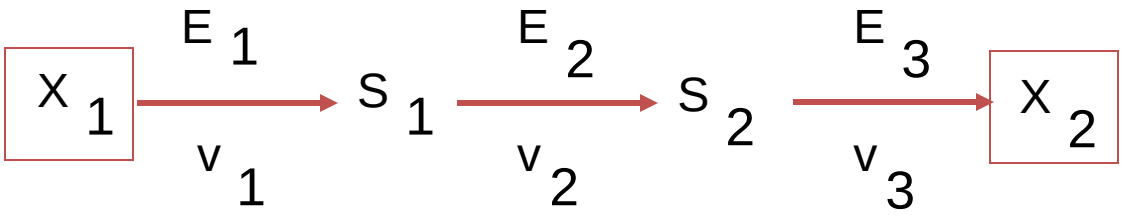


$$\frac{dv_1}{v_1} = \varepsilon_1^1 \frac{dS_1}{S_1}$$

$$\frac{dv_2}{v_2} = \varepsilon_1^2 \frac{dS_1}{S_1} + \varepsilon_2^2 \frac{dS_2}{S_2}$$

$$\frac{dv_3}{v_3} = \varepsilon_2^3 \frac{dS_2}{S_2} + \varepsilon_E^3 \frac{dE_3}{E_3}$$

Metabolic Control Analysis

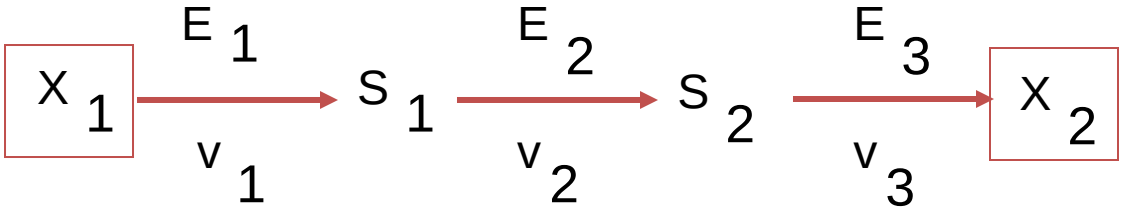


$$C^{J}_{E_3} = \varepsilon^{1}_{1} C^{S_1}_{E_3}$$

$$C^{J}_{E_3} = \varepsilon^{2}_{1} C^{S_1}_{E_3} + \varepsilon^{2}_{2} C^{S_2}_{E_3}$$

$$C^{J}_{E_3} = \varepsilon^{3}_{2} C^{S_2}_{E_3} + 1$$

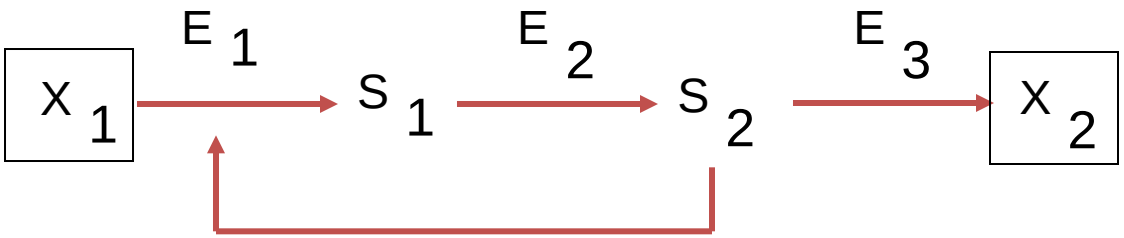
Perturbation Analysis



$$C_{E_3}^{J} = \frac{\varepsilon_1^1 \varepsilon_2^2}{\varepsilon_1^2 \varepsilon_2^3 - \varepsilon_1^1 \varepsilon_2^3 + \varepsilon_1^1 \varepsilon_2^2}$$

$$C_{E_3}^{S_2} = \frac{\varepsilon_1^1 - \varepsilon_1^2}{\varepsilon_1^2 \varepsilon_2^3 - \varepsilon_1^1 \varepsilon_2^3 + \varepsilon_1^1 \varepsilon_2^2}$$

Perturbation Analysis

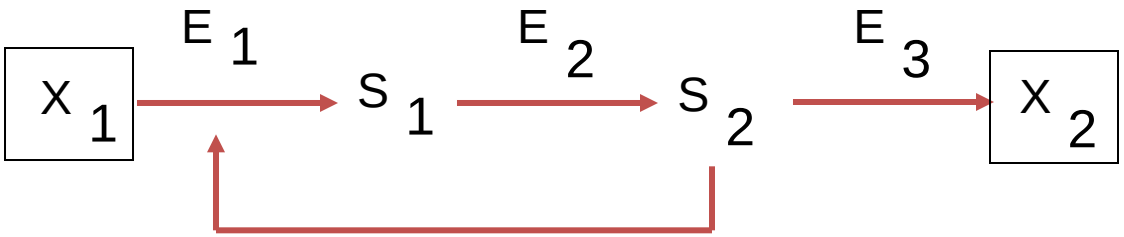


$$C_{E_3}^{J} = \frac{\varepsilon_1^1\varepsilon_2^2 - \varepsilon_2^1\varepsilon_1^2}{\varepsilon_1^2\varepsilon_2^3 - \varepsilon_1^1\varepsilon_2^3 + \varepsilon_1^1\varepsilon_2^2 - \varepsilon_2^1\varepsilon_1^2}$$

$$C_{E_3}^{S_2} = \frac{\varepsilon_1^1 - \varepsilon_1^2}{\varepsilon_1^2\varepsilon_2^3 - \varepsilon_1^1\varepsilon_2^3 + \varepsilon_1^1\varepsilon_2^2 - \varepsilon_2^1\varepsilon_1^2}$$

Extra term

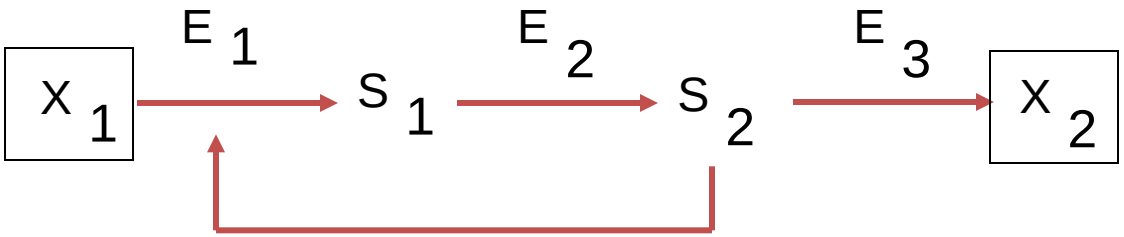
Perturbation Analysis



$$C_{E_3}^{J} = \frac{T_1 - \varepsilon_2^1 \varepsilon_1^2}{T_2 - \varepsilon_2^1 \varepsilon_1^2}$$

$$C_{E_3}^{S_2} = \frac{T_3}{T_2 - \varepsilon_2^1 \varepsilon_1^2}$$

Metabolic Control Analysis

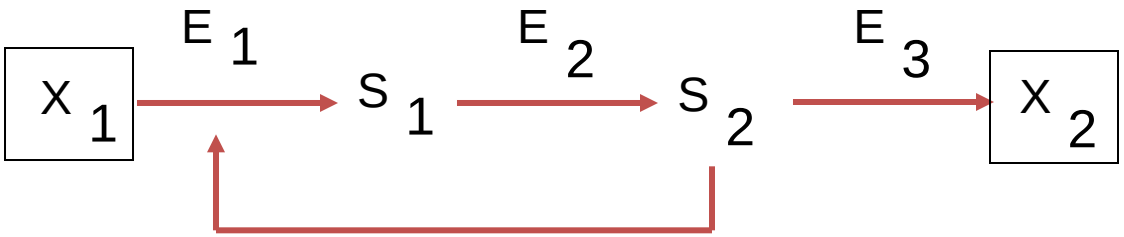


What happens as the feedback gets stronger and stronger?

i.e

The absolute value of ε_2^1 gets larger?

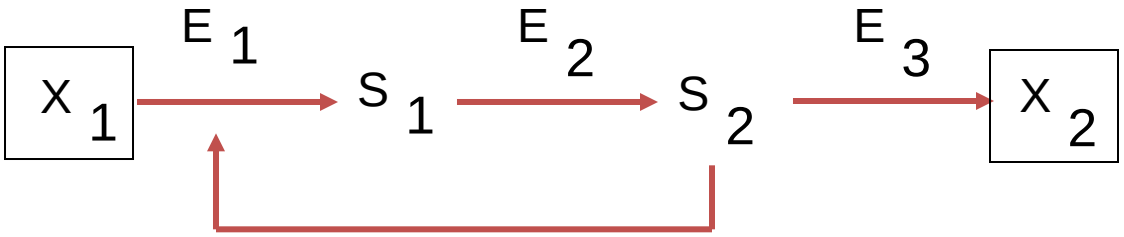
Perturbation Analysis



$$C^{J}_{E_3} \longrightarrow 1$$

$$C^{S_2}_{E_3} \longrightarrow 0$$

Perturbation Analysis



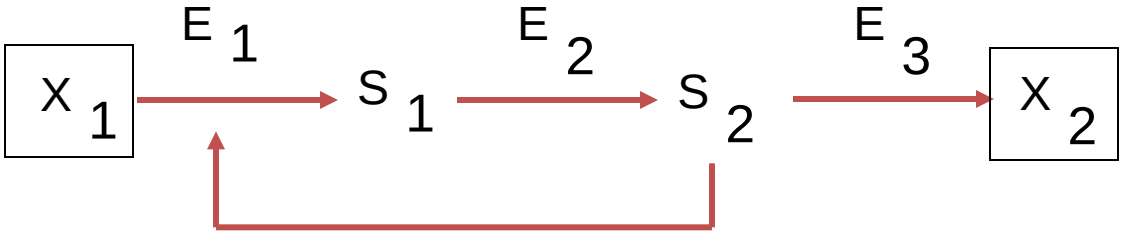
What does this mean?

Feedback has the following consequences:

1. All flux control moves down stream beyond the signal leaving little or no flux control upstream. In fact, the 'controlled' step has very little flux control.

2. The signal molecule is locked into homoeostasis

Perturbation Analysis



What does this mean?

The net effect of this is that feedback control creates a demand controlled network. That is, control over the flux through the pathway is determined largely by the demand for S2.

Important examples is this include:

1. Glycolysis
2. Amino acid biosynthesis