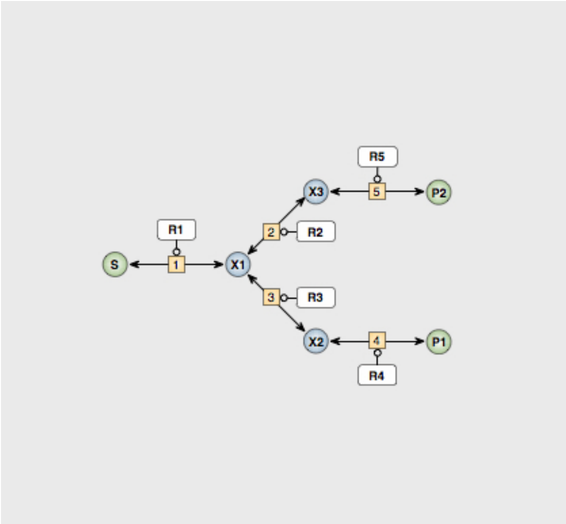
Systems Biology Tutorial 4: Structural analysis of reaction networks

1. Consider the linear branched pathway:



   (a) Construct the stoichiometric matrix N.

$$N = \begin{bmatrix} 1 & -1 & -1 & 0 & 0 \\ 0 & 0 & 1 & -1 & 0 \\ 0 & 1 & 0 & 0 & -1 \end{bmatrix}$$

   where

$$\begin{bmatrix} \dot{x}_1 \\ \dot{x}_2 \\ \dot{x}_3 \end{bmatrix} = N \cdot \begin{bmatrix} v1 \\ v2 \\ v3 \\ v4 \\ v5 \end{bmatrix}$$

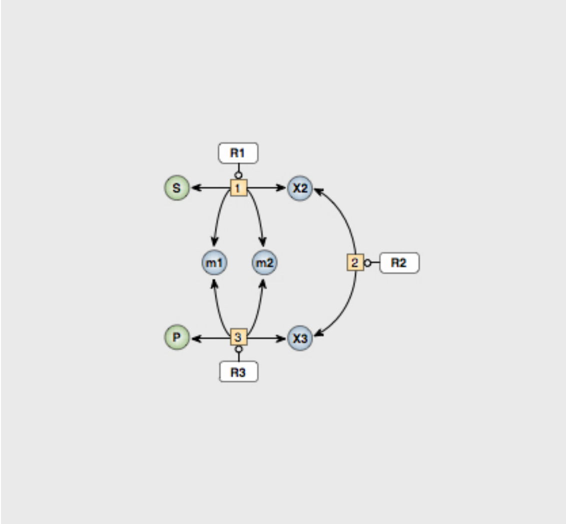
   (b) Are there any dependent metabolites?
   No

   (c) Derive the steady-state flux relations by hand from Nv = 0. How many independent fluxes are there?
   2 independent fluxes. If you choose J1 and J2 the relations are:

$$\begin{aligned} J3 &= J1 - J2 \\ J4 &= J3 = J1 - J2 \\ J5 &= J2 \end{aligned}$$

   (d) Check your answers by running the branch5 model on JWS Online and generating the N, L and K matrices.

2. Consider the linear pathway with a moiety:



(a) Construct the stiochiometric matrix.

$$N = \begin{bmatrix} -1 & 0 & 1 \\ 1 & -1 & 0 \\ 1 & 0 & -1 \\ 0 & 1 & -1 \end{bmatrix}$$

where

$$\begin{bmatrix} \dot{m}_1 \\ \dot{x}_2 \\ \dot{m}_2 \\ \dot{x}_3 \end{bmatrix} = N \cdot \begin{bmatrix} v1 \\ v2 \\ v3 \end{bmatrix}$$

(b) Are there any dependent metabolites?

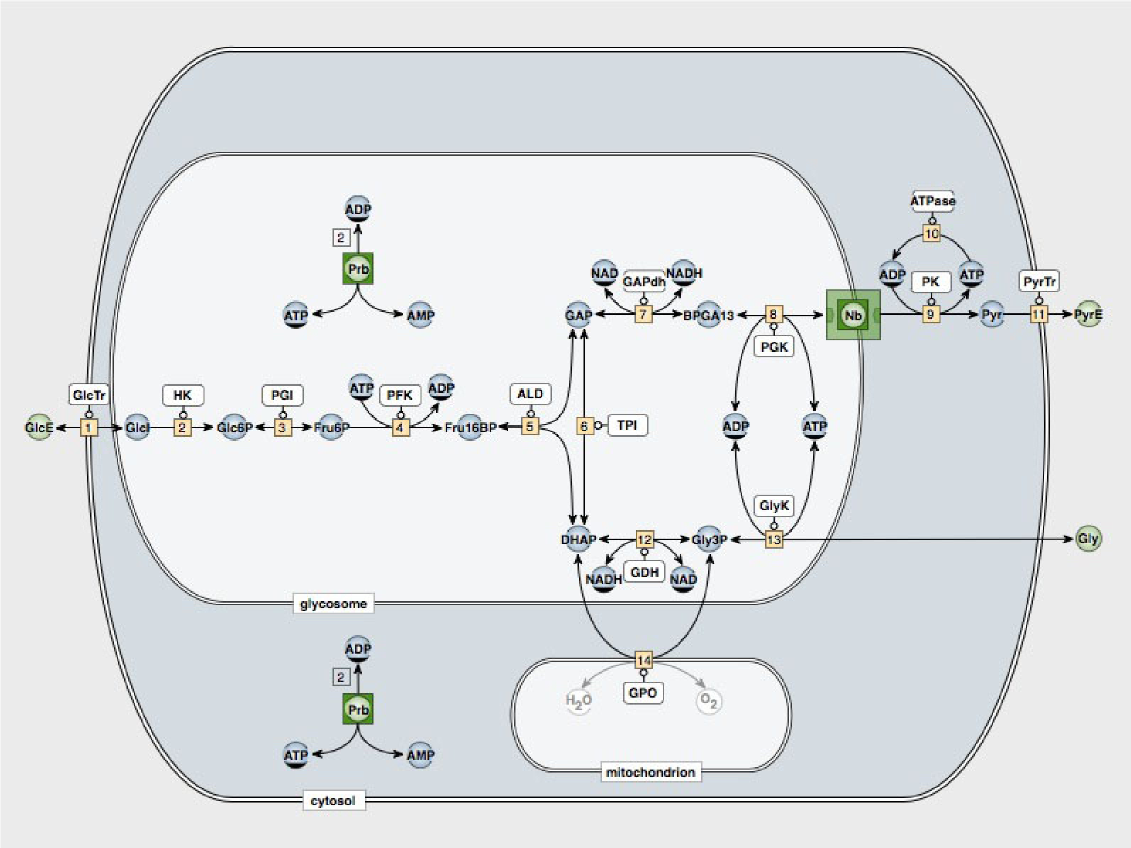
Yes. m1+m2=constant1 and x3+x2+m1=constant2.

(c) Derive the steady-state flux relations by hand from Nv = 0. How many independent fluxes are there?

1 independent flux. J1=J2=J3.

(d) Check your answers by running the lin3moi model on JWS Online and generating the N, L and K matrices.

3. Consider the following model for glycolysis in Trypanosoma brucei:



(a) How many independent fluxes are there?
2

(b) Assuming anaerobic glycolysis: *(J14 = 0, i.e. no flux through vGPO)*

i. What is the flux relation between

A. J12 and J7,
$J12 = J7$

B. J13 and J1?
$J13 = 2 \times J1 - J7$ *in general. Anaerobic: J13 = J1 = J7.*

ii. What is the ratio of PyrE to Gly?
1 to 1

iii. How many moles of ATP are produced per mole of Glc in the glycosome?
net production is 0. v2 consumes 1, v4 consumes 1, v8 produces 1, v13 produces 1.

iv. How many moles of ATP are produced per mole of Glc in the cytosol?
net production is 1

(c) Assuming aerobic glycolysis:

i. Production of what product will increase?
PyrE since reaction v14 will decrease the flux to Gly

ii. What is the maximal flux in J7 related to J1?
$J7 = 2 \times J1$

iii. How many moles of ATP are produced per mole of Glc in the glycosome?
net production is 0. v2 consumes 1, v4 consumes 1, v8 produces 2.

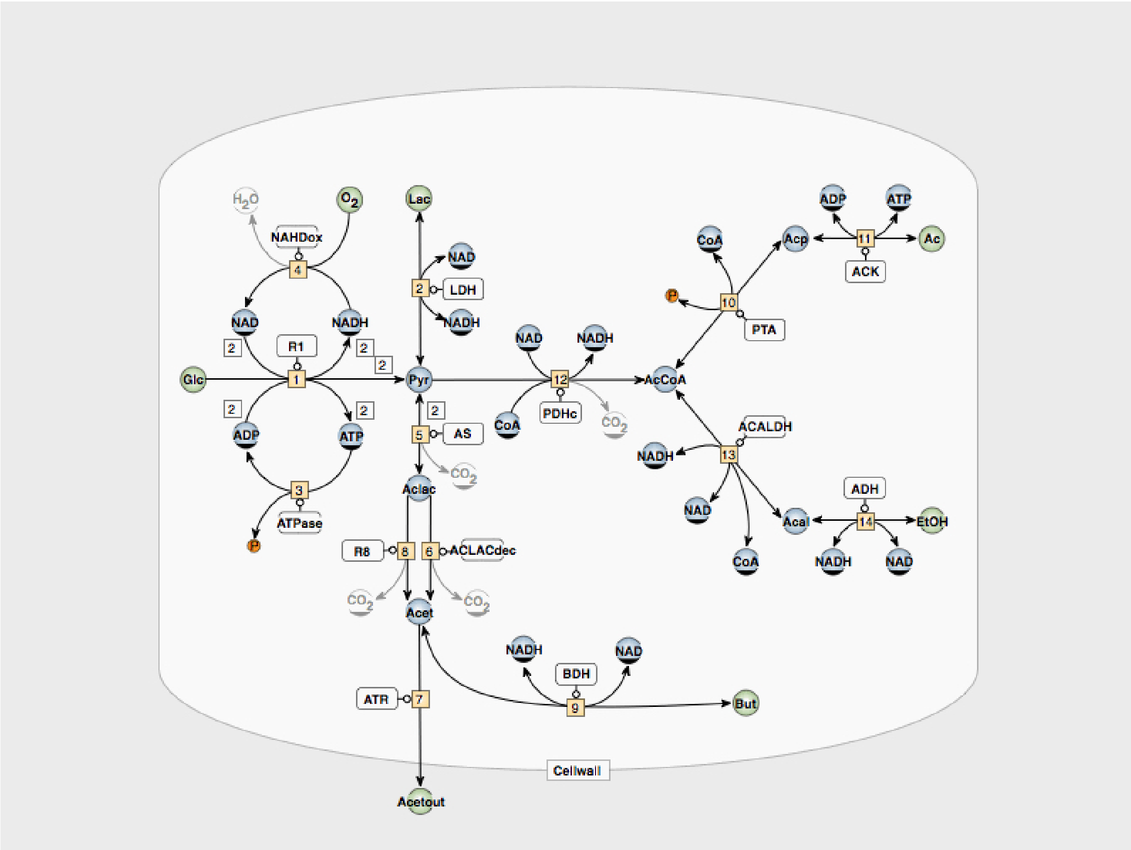
iv. How many moles of ATP are produced per mole of Glc in the cytosol?
net production is 2

(d) Test your answer by running the kerkhovenA model on JWS Online. Adjust the kinetic parameters of GPO to simulate anaerobic / aerobic conditions and plot the relevant rates.

(e) Would glycolysis reach a steady state in the absence of the glycerol branch (anaerobically)?

No, Redox balance will not be maintained i.e. the organism will run out of NADH

4. Consider the following model for glycolysis in Lactococcus lactis:



(a) Consider anaerobic glycolysis:

i. Would steady-state production of lactate be possible for anaerobic glycolysis? If so, what will be the flux relation bewteen J2 and J1 if all available pyruvate is converted tot lactate? How many moles of lactate will be produced per mole of glucose?
yes. $J2 = 2 \times J1$. *2 moles of lactate per mole of glucose.*

ii. How much EtOH will be formed per mole of glucose if all pyruvate were converted to AcCoA?
anaerobic => redox balance has to be mantained in pathway. Up to AcCoA there are 4 NADH and 2 AcCoA. All AcCoA therefore have to be converted to EtOH. Only EtOH will be formed. (2 ETOH per GLC).

(b) Consider aerobic glycolysis:

i. What branch would maximize ATP production?
Reaction 4 oxidises all NADH to NAD; no NADH for EtOH production. All Glc converted to Ac would therefore result in maximum ATP production (4 ATP).

ii. How many moles of product will be formed per mole of glucose if this branch was carrying all the flux?
2 moles of Ac per mole of Glc.

(c) Is the production of butanol redox neutral (anaerobic)?
no. v1 converts 2 NAD to 2 NADH whereas v9 converts only 1 NADH to NAD.

(d) What would be the ratio of Ac to EtOH in the case of anaerobic glycolysis where v12 does not consume NAD (special case in *E. coli* where pyruvate formate lyase catalyses this reaction) ?
anaerobic => redox balance has to be mantained in pathway. Up to AcCoA there are now 2 NADH and 2 AcCoA. 2 AcCoA can therefore split equally between EtOH and Ac. The ratio EtOH:Ac is therefor 1:1