Kinetic FBA

Systems Biology Journal Club

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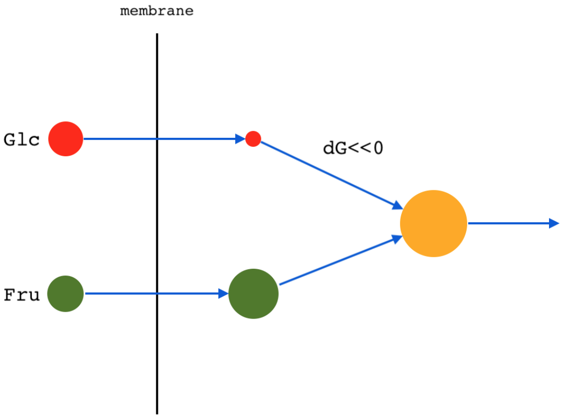
FBA + Kinetic Equations?

Motivation



FBA

$$
\begin{aligned}
\text{maximize} \quad & v_{biomass} = c^T v \\
\text{subject to} \quad & Sv = 0, \\
& l \preceq v \preceq u,
\end{aligned}
\tag{1}
$$

Fluxes Relations

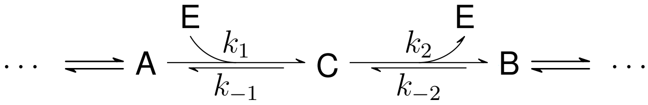
$$Fluxome = f_K(Metabolome, Proteome) \tag{2}$$

Kinetic Equations

$$\mathrm{A}_1 + \mathrm{A}_2 + \cdots + \mathrm{A}_m \underset{k'}{\overset{k}{\rightleftharpoons}} \mathrm{B}_1 + \mathrm{B}_2 + \cdots + \mathrm{B}_n$$

$$v = k[A_1][A_2]\cdots[A_m] - k'[B_1][B_2]\cdots[B_n] \tag{3}$$

Kinetic Equations



$$k_1[A][E] - k_{-1}[C] = v \tag{4}$$

$$k_2[C] - k_{-2}[B][E] = v \tag{5}$$

$$[E] + [C] = \text{const.} = ec \tag{6}$$

Kinetic Equations

$$v = k[A_1][A_2]\cdots[A_m] - k'[B_1][B_2]\cdots[B_n] \tag{7}$$

Non-linear constraint :))

Assumptions

1. Kinetic parameters (k's) are known.
2. Proteome (proteins concentrations, $[E]$'s) is known.
3. All reactions are $1 \to 1$! ($\Rightarrow$ metabolic network would be a graph, rather than a hyper-graph)

Kinetic Simplification

$$k_1[A][E] - k_{-1}[C] = v \tag{8}$$
$$k_2[C] - k_{-2}[B][E] = v \tag{9}$$

$$\Rightarrow k_1 k_2[A][E] - k_{-1}k_2[C] = k_2 v \tag{10}$$
$$k_{-1}k_2[C] - k_{-1}k_{-2}[B][E] = k_{-1}v \tag{11}$$
$$\Rightarrow k_1 k_2[A][E] - k_{-1}k_{-2}[B][E] = (k_{-1} + k_2)v \tag{12}$$
$$\Rightarrow v = \left(\frac{k_1 k_2[E]}{k_{-1} + k_2}\right)[A] - \left(\frac{k_{-1}k_{-2}[E]}{k_{-1} + k_2}\right)[B] \tag{13}$$

(14)

Kinetic Simplification

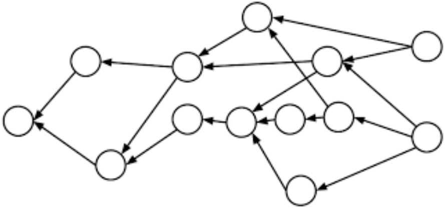
$$\mathrm{A} \underset{k_b}{\overset{k_f}{\rightleftharpoons}} \mathrm{B}$$

$$\Rightarrow v = \left(\frac{k_1 k_2 [E]}{k_{-1} + k_2}\right)[A] - \left(\frac{k_{-1} k_{-2} [E]}{k_{-1} + k_2}\right)[B] \tag{15}$$

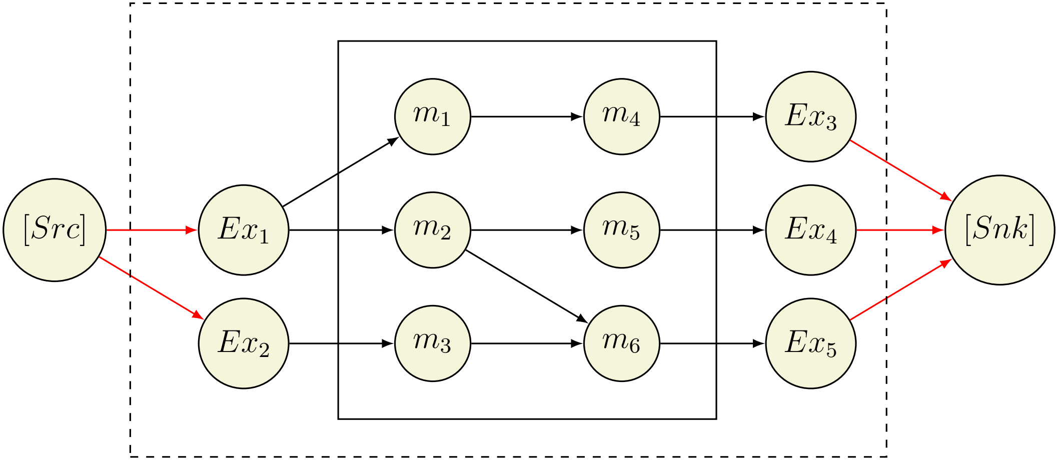
$$:= k_f [A] - k_b [B] \qquad \text{(linear)} \tag{16}$$

Graph Review

#Nodes: m, #Edges (directed): n



Metabolic Network



Graph Parameters

$\mathbf{v}$: fluxes (Fluxome) $\rightarrow$ on edges
$\mathbf{x}$: metabolites concentrations (Metabolome) $\rightarrow$ on nodes
$\mathbf{v}_{in}$: non-exchange fluxes

$$\mathbf{v}_{in} = f_K(Proteome, \mathbf{x}) = f_{K,P}(\mathbf{x}) \tag{17}$$

Graph Equations

#Metabolites: m
#Reactions (exchanges included): n
#Exchanges reactions: l
$\Rightarrow$ Total number of unknowns: $m + n$

$\mathbf{v}_{in} = f_{K,P}(\mathbf{x}) \rightarrow n - l$ equations
$S\mathbf{v} = 0 \rightarrow m$ equations

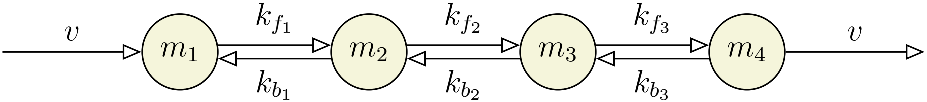
$\Rightarrow l$ degree of freedom remains

Graph Equations

- Simple FBA: n unknowns, m equations
- Kinetic FBA: $n + m$ unknowns, $n + m - l$ equations

$$\Rightarrow \text{advantageous only when } l < n - m$$

Example: Chain Net

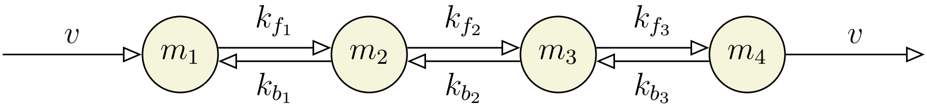


$$n = m + 1, \; l = 2$$

$2m + 1$ unknowns, $2m - 1$ equations

Chain Net

Do kinetic equations help? No! ($l = 2 \nless n - m = 1$)

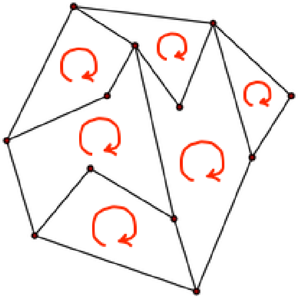


$$v = k_{f_i}[x_i] - k_{b_i}[x_{i+1}] \Rightarrow [x_{i+1}] = \frac{k_{f_i}}{k_{b_i}}[x_i] - \frac{1}{k_{b_i}}v$$

$$\Rightarrow [x_i] = \alpha_i[x_1] - \beta_i v \geq 0 \text{ (merely } [x_1] \geq \max_i\{\frac{\beta_i}{\alpha_i}\}v)$$

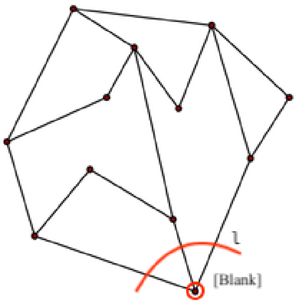
Graph Review

$n - m + 1 =$ number of planar loops



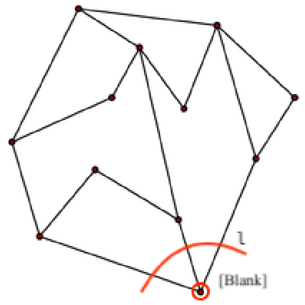
In Metabolic Network

$$\#\text{Nodes} = m+1$$
$$n-(m+1)+1 = n-m = \text{number of (planar) loops}$$
$$l = deg([Blank])$$



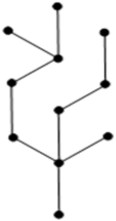
In Metabolic Network

$\Rightarrow$ kinetism(!) is profitable, whenever:
$deg([Blank]) <$ number of (planar) loops



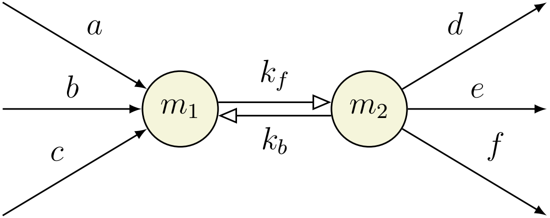
Example: Tree

number of (planar) loops $= 0,\ l \geq 1$



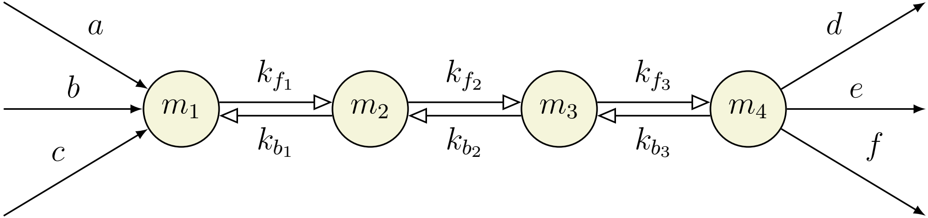
Never happens in a metabolic network :))
(without exchanges, it is blocked)

Useless Equations: Orphan Reaction

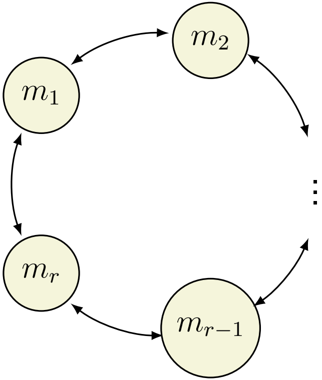


$$v = k_f[x_1] - k_b[x_2] \Rightarrow [x_1] = \frac{k_b}{k_f}[x_2] + \frac{1}{k_f}v \geq 0,\ [x_2] \geq 0$$

Useless Equations: Orphan Chain



Endearing Loops!



$$v = k_{f_i}[x_i] - k_{b_i}[x_{i+1}] \Rightarrow [x_{i+1}] = \frac{k_{f_i}}{k_{b_i}}[x_i] - \frac{1}{k_{b_i}}v$$

Orphan Loop

Clockwise:

$$v = k_{f_i}[x_i] - k_{b_i}[x_{i+1}] \Rightarrow [x_{i+1}] = \frac{k_{f_i}}{k_{b_i}}[x_i] - \frac{1}{k_{b_i}}v$$

$$\Rightarrow [x_i] = \left(\prod_{t=1}^{i-1} \frac{k_{f_t}}{k_{b_t}}\right)[x_1] - \left(\sum_{d=1}^{i-1} \frac{1}{k_{f_d}} \prod_{t=d}^{i-1} \frac{k_{f_t}}{k_{b_t}}\right) v = A_i[x_1] - B_i v \tag{18}$$

Counter-clockwise:

$$v = k_{f_i}[x_i] - k_{b_i}[x_{i+1}] \Rightarrow [x_i] = \frac{k_{b_i}}{k_{f_i}}[x_{i+1}] + \frac{1}{k_{f_i}}v$$

$$\Rightarrow [x_i] = \left(\prod_{t=i}^{r} \frac{k_{b_t}}{k_{f_t}}\right)[x_1] + \left(\sum_{d=i}^{r} \frac{1}{k_{b_d}} \prod_{t=i}^{d} \frac{k_{b_t}}{k_{f_t}}\right) v = C_i[x_1] + D_i v \tag{19}$$

Orphan Loop

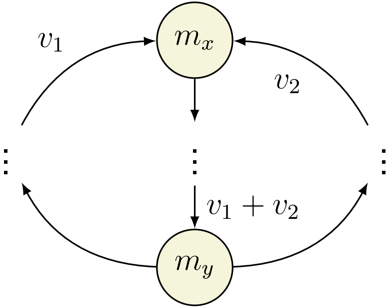
$$[x_i] = A_i[x_1] - B_i v = C_i[x_1] + D_i v$$

$$\Rightarrow [x_1] = \left(\left(\left(\prod_{t=1}^{r} \frac{k_{f_t}}{k_{b_t}} \right) - 1 \right)^{-1} \left(\sum_{d=1}^{r} \frac{1}{k_{f_d}} \prod_{t=d}^{i-1} \frac{k_{f_t}}{k_{b_t}} \right) \right) v = T_1 v$$

$$[x_i] = (A_i T_1 - B_i)v = T_i v$$

one unknown remains (only v)

Orphan Double Loop



$$[x] = A_l[y] - B_l v_1 = A_r[y] - B_r v_2$$
$$[y] = A_c[x] - B_c(v_1 + v_2)$$

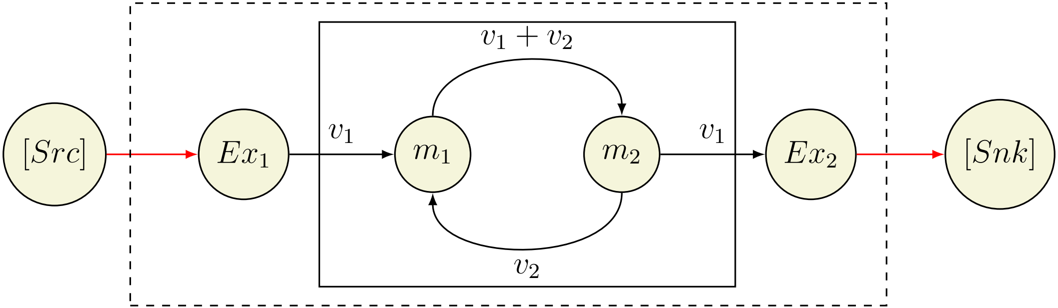
Orphan Double Loop

$$
\begin{aligned}
[x] &= A_l[y] - B_l v_1 \\
[x] &= A_r[y] - B_r v_2 \\
[y] &= A_c[x] - B_c(v_1 + v_2)
\end{aligned}
$$

$$\Rightarrow v_1 = \alpha_1[x],\ v_2 = \alpha_2[x],\ [y] = \beta[x]$$

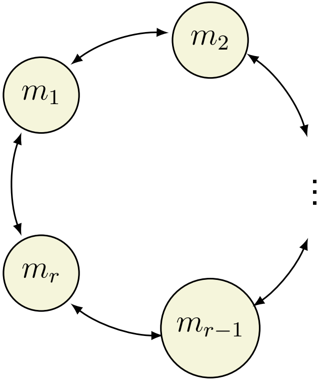
one unknown remains

Loop



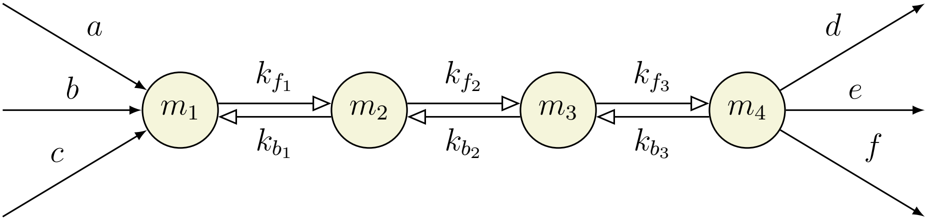
$$[x_i] = \alpha v_1 + \beta v_2$$

Do the Bounds Help?



$$[x_i] = T_i v,\ l_i^x \leq [x_i] \leq u_i^x \Rightarrow \max_i\{\frac{l_i^x}{T_i}\} \leq v \leq \min_i\{\frac{u_i^x}{T_i}\}$$

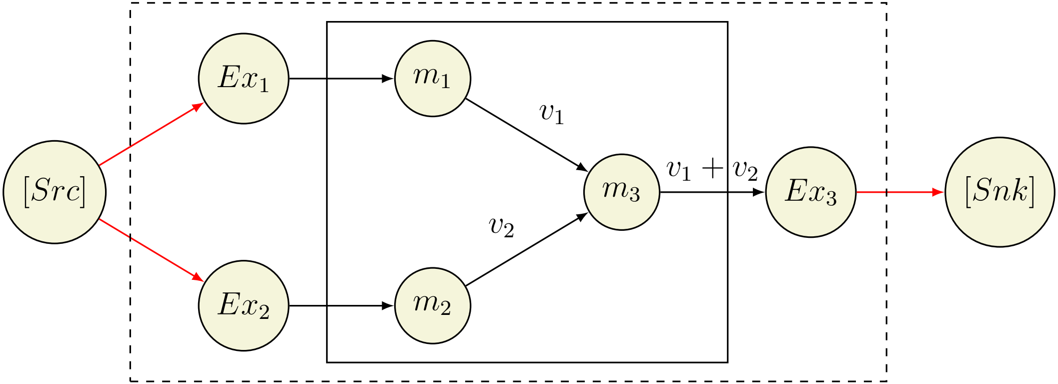
Do the Bounds Help?



$$[x_i] = A_i[x_1] - B_i v,\ \ l_i^x \leq [x_i] \leq u_i^x$$

$$\Rightarrow \max_i\{\frac{A_i l_1 - u_i}{B_i}\} \leq v \leq \min_i\{\frac{A_i u_1 - l_1}{B_i}\}$$

Do the Bounds Help?



$$[Ex_1] = c_1, \ [Ex_2] = c_2$$

Do the Bounds Help?

$$[Ex_1] = c_1 = \alpha_1[x_3] + \beta_1 v_1 \Rightarrow [x_3] = \frac{c_1}{\alpha_1} - \frac{\beta_1}{\alpha_1} v_1$$

$$[Ex_2] = c_2 = \alpha_2[x_3] + \beta_2 v_2 \Rightarrow [x_3] = \frac{c_2}{\alpha_2} - \frac{\beta_2}{\alpha_2} v_2$$

$$\Rightarrow \left(\frac{\beta_1}{\alpha_1}\right) v_1 - \left(\frac{\beta_2}{\alpha_2}\right) v_2 = \left(\frac{c_1}{\alpha_1} - \frac{c_2}{\alpha_2}\right) \tag{20}$$

Do the Bounds Help?

$$\left(\frac{\beta_1}{\alpha_1}\right) v_1 - \left(\frac{\beta_2}{\alpha_2}\right) v_2 = \left(\frac{c_1}{\alpha_1} - \frac{c_2}{\alpha_2}\right)$$

$$v_1 + v_2 \leq u \Rightarrow v_1 \leq u - v_2$$

$$\Rightarrow \left(\frac{\beta_1}{\alpha_1}\right) u - \left(\frac{\beta_1}{\alpha_1} + \frac{\beta_2}{\alpha_2}\right) v_2 \geq \left(\frac{c_1}{\alpha_1} - \frac{c_2}{\alpha_2}\right)$$

$$\Rightarrow v_2 \leq v_2^{max} = \frac{\left(\frac{\beta_1}{\alpha_1}\right) u - \left(\frac{c_1}{\alpha_1} - \frac{c_2}{\alpha_2}\right)}{\left(\frac{\beta_1}{\alpha_1} + \frac{\beta_2}{\alpha_2}\right)} \quad (21)$$

Do the Bounds Help?

$$v_2^{max} = \frac{\left(\frac{\beta_1}{\alpha_1}\right) u - \left(\frac{c_1}{\alpha_1} - \frac{c_2}{\alpha_2}\right)}{\left(\frac{\beta_1}{\alpha_1} + \frac{\beta_2}{\alpha_2}\right)}$$

$$v_2^{max} \approx 0 \Leftrightarrow \left(\frac{\beta_1}{\alpha_1}\right) u \leq \left(\frac{c_1}{\alpha_1} - \frac{c_2}{\alpha_2}\right)$$

$$\alpha_i \approx \frac{k_{b_i}}{k_{f_i}}, \ \beta_i \approx \frac{1}{k_{f_i}}$$

$$\Rightarrow v_2^{max} \approx 0 \Leftrightarrow \frac{u}{k_{b_1}} \leq \left(c_1 \frac{k_{f_1}}{k_{b_1}} - c_2 \frac{k_{f_2}}{k_{b_2}}\right) \Leftrightarrow \frac{u}{c_1} \leq k_{f_1} - \frac{c_2}{c_1} \frac{k_{b_1}}{k_{b_2}} k_{f_2} \tag{22}$$

Do the Bounds Help?

$$\Rightarrow v_2^{max} \approx 0 \Leftrightarrow \frac{u}{k_{b_1}} \leq \left(c_1 \frac{k_{f_1}}{k_{b_1}} - c_2 \frac{k_{f_2}}{k_{b_2}} \right) \Leftrightarrow \frac{u}{c_1} \leq k_{f_1} - \frac{c_2}{c_1} \frac{k_{b_1}}{k_{b_2}} k_{f_2}$$

True since $k_{f_1} >> k_{f_2}$!

$v_2 > 0$ only when:

$$c_1 < \frac{1}{k_{f_1}} \left(u + c_2 \frac{k_{b_1}}{k_{b_2}} k_{f_2} \right) \approx 0! \tag{23}$$

Matrix Formulation

$$\mathbf{v}_{in} = K_E.\mathbf{x}$$

$$S\mathbf{v} = S_{in}\mathbf{v}_{in} + S_{ex}\mathbf{v}_{ex} = S_{in}K_E.\mathbf{x} + S_{ex}\mathbf{v}_{ex} = 0$$

$$S_{in} : \; m \times (n-l), \; S_{ex} : \; m \times l, \; K_E : (n-l) \times m$$

$$\Rightarrow Q := (S_{in}K_E) : \; m \times m$$

$$\Rightarrow Q\mathbf{x} + S_{ex}\mathbf{v}_{ex} = 0$$

Matrix Formulation

$$\Rightarrow Q\mathbf{x} + S_{ex}\mathbf{v}_{ex} = 0$$

Considering the assumed direction for the exchange reactions outward (i.e., A $\longrightarrow$ [Blank]):

$$S_{ex} = P\begin{pmatrix} -I_l \\ 0 \end{pmatrix}$$

$$\Rightarrow Q\mathbf{x} + S_{ex}\mathbf{v}_{ex} = Q\mathbf{x} - \mathbf{v}_{ex}^{(m)} = 0$$

$$\Rightarrow Q\mathbf{x} = \mathbf{v}_{ex}^{(m)}$$

in which $\mathbf{v}_{ex}^{(m)}$ is $\mathbf{v}_{ex}$ elements placed correctly in a $\vec{0}_m$ vector.

(Semi) Kinetic FBA

$$
\begin{aligned}
\text{maximize} \quad & v_{biomass} = c_{met}^T x + c_{ex}^T \mathbf{v}_{ex} \\
\text{subject to} \quad & Q\mathbf{x} = \mathbf{v}_{ex}^{(m)}, \\
& \mathbf{l}_{in} \preceq Q\mathbf{x} \preceq \mathbf{u}_{in}, \\
& (\mathbf{l}_x = Q^{-1}\mathbf{l}_{in} \preceq \mathbf{x} \preceq \mathbf{u}_x = Q^{-1}\mathbf{u}_{in}) \\
& \mathbf{l}_{ex} \preceq \mathbf{v}_{ex} \preceq \mathbf{u}_{ex},
\end{aligned}
\tag{24}
$$

Q Matrix

$$Q_{m\times m} = S_{in}K_E$$

$$q_{i,j} = \mathbf{s}'_i\mathbf{k}_j$$

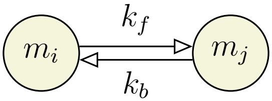
($\mathbf{s}'_i$: i'th row of S_{in}, $\mathbf{k}_j$: j'th column of K_E)

$$q_{i,j} = \sum_{t=1}^{n-l}[\mathbf{s}'_i]_t[\mathbf{k}_j]_t$$

How the concentration of the metabolite j
affects on the production rate of the metabolite i

Q Matrix

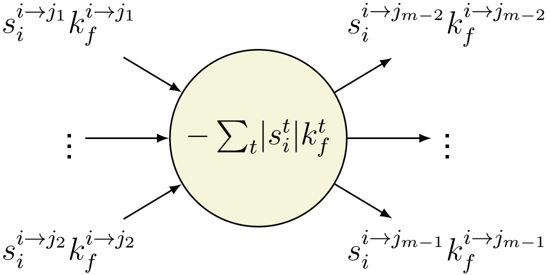
For the networks of $1 \to 1$ reactions:



$$q_{i,j} = \sum_{t=1}^{n-l} [\mathbf{s}'_i]_t [\mathbf{k}_j]_t = s_i^{i \to j} k_j^{i \to j} = (-|s_i^{i \to j}|)(-k_b^{i \to j}) = |s_i^{i \to j}| k_b^{i \to j} = k_b^{i \to j}$$

Q Matrix

$$q_{i,i} = \sum_{t=1}^{n-l} [\mathbf{s}'_i]_t [\mathbf{k}_i]_t = \sum_{j=1}^{m} s_i^{i \to j} k_i^{i \to j} = -\sum_{j=1}^{m} |s_i^{i \to j}| k_f^{i \to j} = -\sum_{j=1}^{m} k_f^{i \to j}$$



Q Matrix

$$q_{i,j} = k_b^{i\to j} = k^{j\to i}$$

$$q_{i,i} = -\sum_j k_f^{i\to j} = -\sum_j k^{i\to j}$$

$$\Rightarrow Q = H - diag(\mathbf{1}^T H), \quad H: \text{ positive hollow matrix}$$

Relaxing Assumptions

?

kFBA

Kinetic parameter estimation (with data from L experimental conditions):

$$
\begin{array}{ll}
\text{minimize} & \sum_l \left(\sum_i w^c_{il}(x_{il} - x^{exp}_{il})^2 + \sum_i w^e_{kl}(x_{kl} - x^{exp}_{kl})^2\right) \\
\text{subject to} & v_{kl} = e_{kl} f_k(x_l;\theta) \quad \forall k \in K, l \in L, \\
& S\mathbf{v}_l = 0 \quad \forall l \in L, \\
& \frac{x_{ATP,l} + 0.5 x_{ADP,l}}{x_{ATP,l} + x_{ADP,l} + x_{AMP,l}} \geq 0.8 \quad \forall l \in L, \\
& 0.001 \leq e_{kl}, x_{il} \leq 10 \quad \forall l \in L, k \in K, i \in I, \\
& 0.35 K^0_{eq,k} \leq K_{eq,l} \leq 2.85 K^0_{eq,k} \quad \forall k \in K
\end{array} \tag{25}
$$

kFBA

$$
\begin{aligned}
\text{minimize} \quad & v_{PTS} \\
\text{subject to} \quad & S\mathbf{v} = 0, \\
& \mathbf{l} \preceq \mathbf{v} \preceq \mathbf{u}, \\
& (l_k - v_k^{(min)})y_k^- + v_k^{(min)} \leq v_k \leq (u_k - v_k^{(max)})y_k^+ + v_k^{(max)}, \\
& \textstyle\sum_k (y_k^+ + y_k^-) \leq n^* \qquad \forall k \in K
\end{aligned} \tag{26}
$$

(n^*: number of violations)

References

Mechanistic analysis of multi-omics datasets to generate kinetic parameters for constraint-based metabolic models; Cameron Cotten and Jennifer L Reed; 2013

Thanks :))