Flux Coupling Analysis, Part I

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(BioSystems, 2010)
Definitions (1)

- A metabolic network (made by “reconstruction”)

- Stoichiometric matrix \((m \times n)\)

\[ S = \begin{bmatrix} 1 & -1 & \cdots \\ 0 & 0 & \cdots \\ 0 & 1 & \cdots \end{bmatrix} \]

- \( \nu \): Flux vector; \( \nu_i \): Flux through reaction \( i \)
Definitions (2)

- In constraint-based modeling of metabolic networks, it is usually assumed that **steady state** condition holds:

  \[ S.v = 0 \]  
  \[ \forall i \in Irr: v_i \geq 0 \]

  (Stoichiometric constraints)  
  (Thermodynamic constraints)
Definitions (3)

An “elementary mode” in a metabolic network can be defined as a steady state flux distribution involving a minimal set of reactions.

\[
\begin{align*}
V^1 &= (1, 0, 0, 1, 1, 0) \\
V^2 &= (1, 0, 1, 1, 0, 1) \\
V^3 &= (1, 1, 1, 0, 0, 0)
\end{align*}
\]
Definitions: Coupled Reactions

- $i \rightarrow j$: for all steady state flux vectors $\nu$, $\nu_i = 0$ implies $\nu_j = 0$

(directionally coupled)

- $4 \rightarrow 5$
- $4 \rightarrow 6$
Definitions: Coupled Reactions

- \( i \leftrightarrow^0 j \): for all steady state flux vectors \( \nu \),
  \( \nu_i = 0 \) implies \( \nu_j = 0 \) and vice versa

(partially coupled)

\[ \nu^1 = (1, 1, 1, 0, 0) \]
\[ \nu^2 = (1, 0, 3, 1, 1) \]
Definitions: Coupled Reactions

• $i \Leftrightarrow j$: for all steady state flux vectors $\nu$, when $\nu_i$ and $\nu_j$ are nonzero, $\nu_i/\nu_j = \text{const.}$ (fully coupled)

• $4 \Leftrightarrow 6$

![Diagram showing coupled reactions with nodes A, B, and C and reactions 1, 2, 4, 6, and 3.]
Definitions: Coupled Reactions

- In metabolic networks, flux coupling is biologically important because functionally related reactions are usually coupled to each other.

- When two fluxes are not coupled, they are "uncoupled".
Flux coupling and EFMs

Theorem 1

Let \( N \) be a metabolic network with flux cone \( C \) and set of elementary modes \( E \). For any two reactions \( i \) and \( j \), the following are equivalent:

- For all \( v \in C \), \( v_i = 0 \) implies \( v_j = 0 \).
- For all \( e \in E \), \( e_i = 0 \) implies \( e_j = 0 \).
Corollary

Let i, j be two non-blocked reactions in a metabolic network N with set of elementary modes E.

- i is directionally coupled to j if and only if for all $e \in E$, $e_i = 0$ implies $e_j = 0$.
- i and j are partially coupled if and only if for all $e \in E$, $e_i = 0$ implies $e_j = 0$ and vice versa.
- i and j are fully coupled if and only if there exists a constant $c \neq 0$ such that for all $e \in E$, $e_j = c \cdot e_i$. 
Definitions: Uncoupled Reactions

- $i \leftrightarrow_j$ M.E.: $i$ and $j$ never appear in the same elementary mode (mutually exclusive)

- $2 \leftrightarrow 4$
Definitions: Uncoupled Reactions

- $i \leftrightarrow^\text{S.C.} j$: $i$ and $j$ are uncoupled, but they are not mutually exclusive
  (sometimes coupled)

- $3 \leftrightarrow^\text{S.C.} 4$
Motivation

- When a metabolic network is “reconstructed”, some reactions might be missing compared to the actual network.

- Do these missing reactions influence the results of flux coupling analysis?
Flux coupling analysis and missing reactions (1)

- $1 \xrightarrow{=} 3$
- $4 \xrightarrow{=} 6$
- $3 \leftrightarrow 4$

\[ \text{S.C.} = 0 \]
Flux coupling analysis and missing reactions (2)

- $1 \iff 3$
- $4 \iff 6$
- $3 \rightarrow 4$
Flux coupling analysis and missing reactions (3)

- Some possible changes in flux coupling due to missing reactions in the network

\[ i \rightarrow j \quad i \leftrightarrow j \]
Flux coupling analysis and missing reactions (4)

- If some reactions are not included in metabolic networks, flux coupling relations in the smaller network may be different from flux coupling relations in actual (complete networks).

- What relations in complete metabolic networks can convert to other relations in an incomplete network?
Arrow from R₁ to R₂: having R₁ in actual network, but having R₂ in an incomplete network

Yellow: Coupling

Green: Uncoupling
Uncoupled reaction pairs might be detected as coupled when networks are incomplete.
Conclusion

- Two reactions might be uncoupled in a complete network, but due to missing reactions (lack of knowledge) they might be detected wrongly as coupled.

- If two reactions are uncoupled in an incomplete network, they are certainly uncoupled in the actual (complete) networks.
Test case 1

- Comparing two versions of *E. coli* network
- Computing the number of changes in coupling relations
Changes in flux coupling and uncoupling relations between two E.coli metabolic models.

<table>
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<th>Change in the coupling type</th>
<th>frequency</th>
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<th>frequency</th>
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<td>F ⇒ U</td>
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<td>D ⇒ U</td>
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<td>U ⇒ D</td>
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</table>
Test case 2

- Correlation between fully-coupled reactions and the co-expression of their corresponding genes (based on an older version of *E. coli* network)

- The analysis was repeated for the new version of *E. coli* network.
Test case 2

- Coupling relations of the “fully-coupled” reactions in the old network were updated.
- Some “fully-coupled” reactions now become directionally-coupled or even uncoupled.
- The pairs that are still fully coupled in the recent version of *E. coli* network, show higher gene expression correlations.