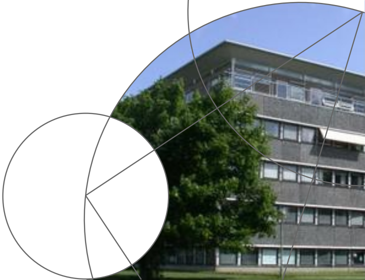
Faculty of Science

Applied algebra in the analysis of biochemical reaction networks

Elisenda Feliu
Department of Mathematical Sciences
University of Copenhagen



Mathematics of Reaction Networks at CPH

- We focus on the development of mathematical theory to analyse mathematical models that arise from studying systems of (bio)chemical reactions.
- My work exploits tools from applied/computational algebraic geometry.

Content

- The big picture
- Mathematical framework
- Some results

The big picture

Biochemical reaction networks

We represent a (bio)chemical reaction with an "arrow" between linear combinations of chemical species

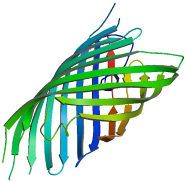
For example, the representation

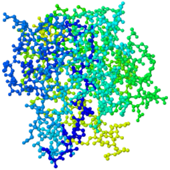
$$A + B \to C$$

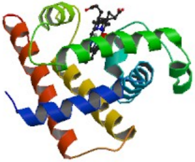
indicates that two species A, B react to form a third species C.

Species: in principle anything (animals, chemical species, healthy or ill people...)

Here: biochemical species, such as proteins





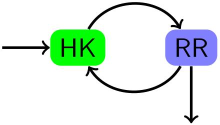


Source: PDB

Example: Two-component systems

Two-component systems are a type of specific biochemical reaction network used by bacteria to transfer cellular signal:

$$\begin{aligned} \mathrm{HK} &\longrightarrow \mathrm{HK_p} \\ \mathrm{HK_p} + \mathrm{RR} &\rightleftharpoons \mathrm{HK} + \mathrm{RR_p} \\ \mathrm{RR_p} &\longrightarrow \mathrm{RR} \end{aligned}$$



HK = histidine kinase; RR = response regulator

The evolution of the concentrations in time is modelled using a system of ordinary differential equations:

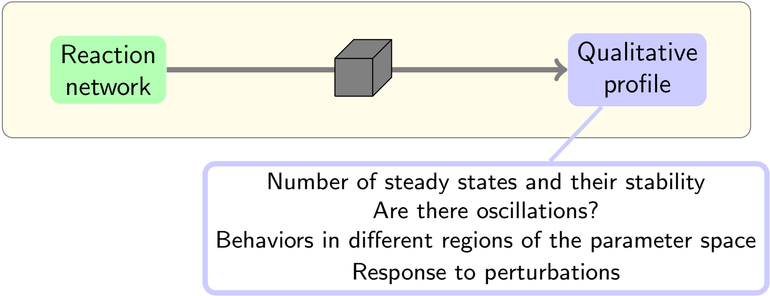
$$\begin{aligned} \dot{x}_1 &= -\kappa_1 x_1 + \kappa_2 x_2 x_3 - \kappa_3 x_1 x_4 \\ \dot{x}_2 &= \kappa_1 x_1 - \kappa_2 x_2 x_3 + \kappa_3 x_1 x_4 \\ \dot{x}_3 &= -\kappa_2 x_2 x_3 + \kappa_3 x_1 x_4 + \kappa_4 x_4 \\ \dot{x}_4 &= \kappa_2 x_2 x_3 - \kappa_3 x_1 x_4 - \kappa_4 x_4 \end{aligned}$$

where
$x_1 = [\mathrm{HK}]$, $x_2 = [\mathrm{HK}_p]$,
$x_3 = [\mathrm{RR}]$, $x_4 = [\mathrm{RR}_p]$.

Here $\kappa_1, \kappa_2, \kappa_3, \kappa_4$ are parameters, generally unknown.

Qualitative profiling

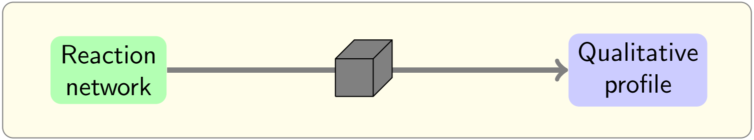
(Dream) Goal:



No numerical simulations, no parameter sampling.

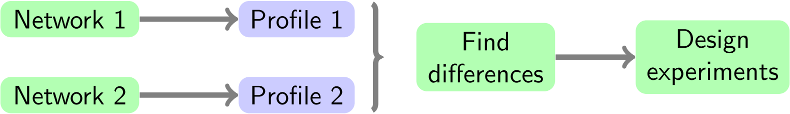
Qualitative profiling

(Dream) Goal:



Relevance:

1. Overview of possible dynamical behaviours → synthetic biology.
2. Assess the validity of the model or mechanism:



Advantage: there is no need to know the parameters.

Mathematical framework

The mathematics of reaction networks

- Chemical reaction network theory (Feinberg, Horn, Jackson, 70ies).
  - Number of steady states and dynamics around steady states.
  - Relation between network structure and dynamical properties.
  - Celebrated result (deficiency zero): all networks in a certain class have a unique steady state, which is asymptotically stable (conjectured globally stable)

- Recently: Growing community of mathematicians with different background and focus on biochemical networks.
  - Provide (computational) strategies to determine network's dynamics.
  - Use theory from applied algebra, polyhedral geometry, dynamical systems, stochastic processes...

Example

$$X_1 \xrightarrow{\kappa_1} X_2$$
$$2X_2 \xrightarrow{\kappa_2} 2X_1$$
$$X_1 + X_2 \xrightarrow{\kappa_3} 2X_2$$

ODE system with $\kappa_1, \kappa_2, \kappa_3 > 0$:

$$\dot{x}_1 = -\kappa_1 x_1 + 2\kappa_2 x_2^2 - \kappa_3 x_1 x_2,$$
$$\dot{x}_2 = \kappa_1 x_1 - 2\kappa_2 x_2^2 + \kappa_3 x_1 x_2.$$

$$\begin{pmatrix} \dot{x}_1 \\ \dot{x}_2 \end{pmatrix} = \begin{pmatrix} -1 & 2 & -1 \\ 1 & -2 & 1 \end{pmatrix} \begin{pmatrix} \kappa_1 x_1 \\ \kappa_2 x_2^2 \\ \kappa_3 x_1 x_2 \end{pmatrix}.$$

Reaction networks

A reaction network over a set of species $\mathcal{X} = \{X_1, \dots, X_n\}$ consists of a finite set of reactions of the form

$$r_j\colon \sum_{i=1}^{n} b_{ij} X_i \to \sum_{i=1}^{n} c_{ij} X_i, \qquad b_{ij}, c_{ij} \in \mathbb{Z}_{\geq 0}$$

Dynamical system:

$$\dot{x} = N v_\kappa(x).$$

- N is the stoichiometric matrix, where entry (i,j) is the net production of species i in reaction j: $N_{ij} = (c_{ij} - b_{ij})$.
- x_i= concentration of species X_i.
- Mass-action: $v_{\kappa,j}(x) = \kappa_j x_1^{b_{1j}} \cdot \ldots \cdot x_n^{b_{nj}}$.
  $\kappa_j > 0$, reaction rate constants.
- $\mathbb{R}^n_{>0}$ and $\mathbb{R}^n_{\geq 0}$ are forward invariant.

Stoichiometric compatibility classes

$$\dot{x}_1 = -\kappa_1 x_1 + 2\kappa_2 x_2^2 - \kappa_3 x_1 x_2, \qquad \dot{x}_2 = \kappa_1 x_1 - 2\kappa_2 x_2^2 + \kappa_3 x_1 x_2.$$

$x_1 + x_2$ is constant along trajectories as

$$\dot{x}_1 + \dot{x}_2 = 0$$

Hence, along any trajectory

$$x_1 + x_2 = c$$

with c depending on the initial condition.

In general: with

$$\dot{x} = Nv(x),$$

any vector ω in the left kernel of N, that is, such that

$$\omega^T N = 0$$

gives $\omega^T \dot{x} = 0$ as above. Hence,

$\omega^T x$ is constant along trajectories

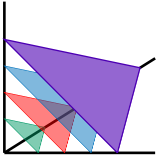
Stoichiometric compatibility classes

W: matrix whose rows form a basis of the left kernel of N, $\ker(N^T) = \mathrm{Im}(N)^{\perp}$

Stoichiometric compatibility classes:

$$Wx = c, \qquad x \in \mathbb{R}^n_{\geq 0}$$

with c vector of total amounts.



Every trajectory is confined to one of these classes, depending on the initial condition.

Steady states

The steady states or equilibrium points of the ODE system are given as the solutions to

$$Nv_\kappa(x) = 0.$$

In particular: we are concerned with positive solutions in each stoichiometric compatibility class:

$$x \in \mathbb{R}^n_{>0} \qquad \text{such that} \qquad Nv_\kappa(x) = 0 \qquad \text{and} \qquad Wx = c$$

Two types of parameters, κ, c, treated as unknown.

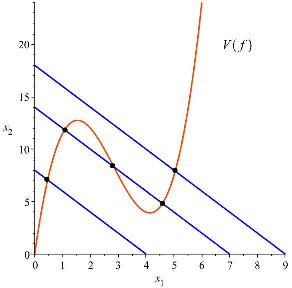
$$C_{\kappa,c} = \{x \in \mathbb{R}^n_{>0} |\ Nv_\kappa(x) = 0, \quad Wx = c\}.$$

Steady states and stoichiometric compatibility classes

In the example,

$$C_{\kappa,c} = \left\{ x \in \mathbb{R}^2_{>0} \;\middle|\; \begin{array}{r} -\kappa_1 x_1 + 2\kappa_2 x_2^2 - \kappa_3 x_1 x_2 = 0, \\ \kappa_1 x_1 - 2\kappa_2 x_2^2 + \kappa_3 x_1 x_2 = 0 \\ \kappa_1 x_1 - 2\kappa_2 x_2^2 + \kappa_3 x_1 x_2 = 0 \\ x_1 + x_2 = c \end{array} \right\}.$$

These sets are parameterised by $\kappa = (\kappa_1, \dots, \kappa_m)$ and $c = (c_1, \dots, c_d)$.

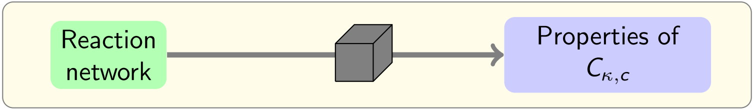


Expected: each of these sets contains a finite number of points.

The number of elements might depend on the parameters.

The number of positive steady states is linked to cell decision-making.

$C_{\kappa,c} = \{x \in \mathbb{R}^n_{>0} | \; Nv_\kappa(x) = 0, \quad Wx = c\}.$



(**Multistationarity**) Is there a choice of reaction rate constants κ and total amounts c such that

$$\#C_{\kappa,c} \geq 2 \quad ? \qquad\qquad \text{(essentially solved)}$$

(**Parameter regions**) For which choices of parameters $\#C_{\kappa,c} = M$?

(**Bistability**) Is there a choice of κ and c such that $C_{\kappa,c}$ has two asymptotically stable positive steady states?

(**Oscillations**) Is there a choice of κ and c such that there is a periodic solution?

Hopf bifurcation: Is there a choice of parameters and steady state such that the Jacobian of the ODE system has a single complex-conjugate pair of eigenvalues that crosses the imaginary axis, while all other eigenvalues remain with negative real parts?

Such a bifurcation generates nearby oscillations.

Our friends today

A simplified signaling mechanism involving a hybrid histidine kinase:

$$\mathrm{HK}_{00} \xrightarrow{\kappa_1} \mathrm{HK}_{\mathrm{p}0} \xrightarrow{\kappa_2} \mathrm{HK}_{0\mathrm{p}} \xrightarrow{\kappa_3} \mathrm{HK}_{\mathrm{pp}}$$
$$\mathrm{HK}_{0\mathrm{p}} + \mathrm{RR} \xrightarrow{\kappa_4} \mathrm{HK}_{00} + \mathrm{RR}_{\mathrm{p}}$$
$$\mathrm{HK}_{\mathrm{pp}} + \mathrm{RR} \xrightarrow{\kappa_5} \mathrm{HK}_{\mathrm{p}0} + \mathrm{RR}_{\mathrm{p}}$$
$$\mathrm{RR}_{\mathrm{p}} \xrightarrow{\kappa_6} \mathrm{RR}$$

The **model model**: 2-site phosphorylation cycle, where phosphorylation is sequential and distributive

$$A + K \rightleftharpoons AK \xrightarrow{k_1} A_p + K \rightleftharpoons A_pK \xrightarrow{k_2} A_{pp} + K$$
$$A_{pp} + F \rightleftharpoons A_{pp}F \xrightarrow{k_4} A_p + F \rightleftharpoons A_pF \xrightarrow{k_3} A + F$$

Both admit 1, 2 or 3 positive steady states.

Key technique I: Parametrizations

A positive parametrization of the (positive) steady states is an injective function

$$\phi\colon \mathbb{R}^d_{>0} \to \mathbb{R}^n_{>0} \qquad \xi \mapsto \phi(\xi)$$

such that the image is exactly the set of positive steady states.

In the example in the intro (two-component system):

$$\phi(x_2,x_4) = \left(\tfrac{\kappa_4}{\kappa_1}x_4, x_2, \tfrac{(\kappa_1+\kappa_3x_4)\kappa_4x_4}{\kappa_1\kappa_2x_2}, x_4\right)$$

$$X_1 \xrightarrow{\kappa_1} X_2$$
$$X_2 + X_3 \xrightarrow{\kappa_2} X_1 + X_4$$
$$X_1 + X_4 \xrightarrow{\kappa_3} X_2 + X_3$$
$$X_4 \xrightarrow{\kappa_4} X_3$$

- In practice it often works: Choose a variable for each conservation law, and solve (e.g. in Maple) the system of steady state equations for these variables.
- From d (reactant-)non-interacting species.

$$X_1 \xrightarrow{\kappa_1} X_2 \qquad X_2 + X_3 \xrightarrow{\kappa_2} X_1 + X_4$$
$$X_4 \xrightarrow{\kappa_4} X_3 \qquad X_1 + X_4 \xrightarrow{\kappa_3} X_2 + X_3$$

Key technique II: Model reduction

We seek results relating qualitative properties of two networks F and G:

(Provided ...) if F has property X for some choice of parameter values, then so does G.

Property X can be:

X1 Having at least ℓ positive/asymptoticaly stable/unstable (...) steady states.

X2 Having a periodic solution.

Many such operations are known

(At least: Craciun and Feinberg, Conradi et al., Joshi and Shiu, Feliu and Wiuf, Banaji and Pantea...)

- Make a reaction reversible

$$F: S + E \longrightarrow S^* + E \qquad G: S + E \rightleftharpoons S^* + E$$

Joshi, Shiu (for X1), Banaji (for X2)

- Add intermediates

$$F: S + E \longrightarrow S^* + E \qquad G: S + E \rightleftharpoons X \longrightarrow S^* + E$$

Feliu, Wiuf (for X1), Banaji (for X2)

2-site

For example, this network admits 3 positive steady states

$$A + K \longrightarrow AK \longrightarrow A_p + K \longrightarrow A_{pp} + K$$

$$A_{pp} + F \longrightarrow A_p + F \longrightarrow A + F$$

and from here we conclude that the double phosphorylation network also does:

$$A + K \rightleftharpoons AK \longrightarrow A_p + K \rightleftharpoons A_pK \longrightarrow A_{pp} + K$$

$$A_{pp} + F \rightleftharpoons A_{pp}F \longrightarrow A_p + F \rightleftharpoons A_pF \longrightarrow A + F$$

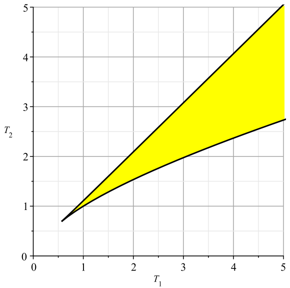
Some results

- Identifying parameter regions for multistationarity
- Hopf bifurcation in signaling cascades
- (Symbolically proving bistability)

Parameter regions for multistationarity

Goal:

find a full or partial description of the parameter that give multistationarity, or a given number of steady states.



- Generic tool: Cylindrical Algebraic Decomposition can theoretically answer this question, but it is impractical.
- Can we find computationally feasible approaches by using the specific structure of systems arising from reaction networks?

The answer might be too complex...

$$\mathrm{HK}_{00} \xrightarrow{\kappa_1} \mathrm{HK}_{\mathrm{p}0} \xrightarrow{\kappa_2} \mathrm{HK}_{0\mathrm{p}} \xrightarrow{\kappa_3} \mathrm{HK}_{\mathrm{pp}}$$
$$\mathrm{HK}_{0\mathrm{p}} + \mathrm{RR} \xrightarrow{\kappa_4} \mathrm{HK}_{00} + \mathrm{RR}_{\mathrm{p}}$$
$$\mathrm{HK}_{\mathrm{pp}} + \mathrm{RR} \xrightarrow{\kappa_5} \mathrm{HK}_{\mathrm{p}0} + \mathrm{RR}_{\mathrm{p}}$$
$$\mathrm{RR}_{\mathrm{p}} \xrightarrow{\kappa_6} \mathrm{RR}$$

The network has three positive steady states if and only if

$$a_2 > 0 \qquad 9a_0a_3 + a_1a_2 < 0$$
$$27a_0^2a_3^2 + 18a_0a_1a_2a_3 - 4a_0a_2^3 + 4a_1^3a_3 - a_1^2a_2^2 < 0 \qquad -6a_0a_2 + 2a_1^2 > 0,$$

where

$$\begin{aligned}
a_0 &= (\kappa_1 + \kappa_2)\kappa_4\kappa_5\kappa_6 > 0 \\
a_1 &= (\kappa_1(c_1\kappa_2\kappa_4 + \kappa_2\kappa_6 + \kappa_3\kappa_6) - c_2(\kappa_1 + \kappa_2)\kappa_4\kappa_6)\kappa_5 \\
a_2 &= (\kappa_1\kappa_2\kappa_3(c_1\kappa_5 + \kappa_6) - c_2\kappa_1(\kappa_2 + \kappa_3)\kappa_5\kappa_6) \\
a_3 &= -c_2\kappa_1\kappa_2\kappa_3\kappa_6 < 0.
\end{aligned}$$

Kothamanchu, Feliu, Cardelli, Soyer (2015)

Partial answer

We say a reaction rate constant κ enables multistationarity if can we find a vector of total amounts c such that there are at least two positive steady states.

What values of κ enable multistationarity?

Theorem. Consider a network such that . . . (some technical conditions). Fix κ. There exists a (computable) polynomial $p(x)$ such that

(A) **Uniqueness.** If

$$\operatorname{sign}(p(x)) = + \quad \text{for } \textbf{all} \text{ positive } x,$$

then there is exactly one positive steady state in each class.

(B) **Multistationarity.** If

$$\operatorname{sign}(p(x^*)) = - \quad \text{for } \textbf{some} \text{ positive } x^*,$$

then there are at least two positive steady states in the class of x^*.

Extra info: The result is based on Brouwer degree theory. The polynomial $p(x)$ is the determinant of the Jacobian of the system of equations evaluated at a parameterisation of the steady state variety. The technical conditions are no boundary steady states and dissipativity.

Conradi C, Feliu E, Mincheva M, Wiuf C (2017) Identifying parameter regions for multistationarity. PloS Computational Biology.

Two-component system

If $\text{sign}(p(x)) = +$ for all positive x, **one steady state** in each class.

If $\text{sign}(p(x^*)) = -$ for one positive x^*, then there is **multistationarity** in the class of x^*.

$$\begin{aligned} \mathrm{HK} &\to \mathrm{HK_p} \\ \mathrm{HK_p} + \mathrm{RR} &\to \mathrm{HK} + \mathrm{RR_p} \\ \mathrm{RR_p} &\to \mathrm{RR} \end{aligned}$$

$$p(x) = \kappa_1\kappa_2 x_2 + \kappa_2\kappa_3 x_3 + \kappa_1\kappa_3.$$

$p(x) > 0$ for all positive x and κ

$\Rightarrow$ One positive steady state in each class for all κ.

Example: Hybrid two-component system

If $\mathrm{sign}(p(x)) = +$ for all positive x, **one steady state** in each class.

If $\mathrm{sign}(p(x^*)) = -$ for one positive x^*, then there is **multistationarity** in the class of x^*.

$$
\begin{gathered}
\mathrm{HK_{00}} \xrightarrow{\kappa_1} \mathrm{HK_{p0}} \xrightarrow{\kappa_2} \mathrm{HK_{0p}} \xrightarrow{\kappa_3} \mathrm{HK_{pp}} \\
\mathrm{HK_{0p}} + \mathrm{RR} \xrightarrow{\kappa_4} \mathrm{HK_{00}} + \mathrm{RR_p} \\
\mathrm{HK_{pp}} + \mathrm{RR} \xrightarrow{\kappa_5} \mathrm{HK_{p0}} + \mathrm{RR_p} \\
\mathrm{RR_p} \xrightarrow{\kappa_6} \mathrm{RR}
\end{gathered}
$$

$$
\begin{aligned}
p(x) = {} & \kappa_1\kappa_2\kappa_3\kappa_6 + (\kappa_1+\kappa_2)\kappa_4\kappa_5\kappa_6 x_5^2 \\
& + \kappa_2\kappa_4\kappa_5^2\left(\frac{\kappa_1}{\kappa_3} - 1\right) x_4 x_5^2 + 2\kappa_1\kappa_2\kappa_4\kappa_5 x_4 x_5 \\
& + (\kappa_2+\kappa_3)\kappa_1\kappa_5\kappa_6 x_5 + \kappa_1\kappa_2\kappa_3\kappa_5 x_4
\end{aligned}
$$

- If $\kappa_1 \geq \kappa_3$: sign$= +$ for all $x_4, x_5 > 0$. There exists a unique positive steady state in each class.
- If $\kappa_1 < \kappa_3$, let $x_i = T$ and T be arbitrarily large. Then sign$= -$. There is multistationarity.

κ **enables multistationarity** $\Leftrightarrow$ $\kappa_1 < \kappa_3$

Critical steps

- Checking the technical conditions
- Finding $p(x)$ involves finding a positive parameterisation of the steady states.
- **Signs: If $p(x)$ has a negative coefficient, can we guarantee that $p(x)$ is negative for some positive x?**
- Computational complexity.

There exist algorithmic (sufficient) criteria to check
all the steps required to apply the theorem.

Signs and the Newton polytope

If $p(x)$ has a negative coefficient, is $p(x)$ negative for some $x > 0$?

$$x^2 - 2xy + y^2 = (x-y)^2 \geq 0$$

Multivariate polynomial

$$f(x) = \sum_{v \in \mathbb{N}^n} \alpha_v x^v,$$

where $x^v = x_1^{v_1} \cdots x_n^{v_n}$ and $\alpha_v \in \mathbb{R}$, for which only a finite number are non-zero.

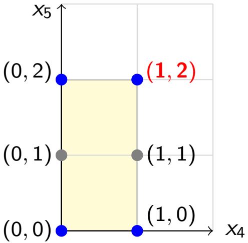
The Newton polytope of $f(x)$, $\mathcal{N}(f)$, is the convex hull of the exponents $v \in \mathbb{N}^n$ for which $\alpha_v \neq 0$.

Proposition: For every vertex v of $\mathcal{N}(f)$, there exists $x^* \in \mathbb{R}^n_{>0}$ such that

$$\operatorname{sign}(f(x^*)) = \operatorname{sign}(\alpha_v).$$

Signs and the Newton polytope

$$p(x) = \kappa_1\kappa_2\kappa_3\kappa_6 + (\kappa_1 + \kappa_2)\kappa_4\kappa_5\kappa_6 x_5^2 + \kappa_2\kappa_4\kappa_5^2\left(\frac{\kappa_1}{\kappa_3} - 1\right) x_4 x_5^2$$
$$+ 2\kappa_1\kappa_2\kappa_4\kappa_5 x_4 x_5 + (\kappa_2 + \kappa_3)\kappa_1\kappa_5\kappa_6 x_5 + \kappa_1\kappa_2\kappa_3\kappa_5 x_4$$



Signs and the Newton polytope

The **model model**: 2-site phosphorylation cycle

$$A + K \rightleftharpoons AK \xrightarrow{k_1} A_p + K \rightleftharpoons A_pK \xrightarrow{k_2} A_{pp} + K$$

$$A_{pp} + F \rightleftharpoons A_{pp}F \xrightarrow{k_4} A_p + F \rightleftharpoons A_pF \xrightarrow{k_3} A + F$$

K_1, K_2, K_3, K_4 Michaelis-Menten constants.

$$\begin{aligned}
p(x) = {} & K_2^2 K_4 k_1^2 k_2 (k_1 k_4 - k_2 k_3) x_1^4 x_3^2 + K_1 K_2^2 K_4 k_1^2 k_3 k_2^2 x_1^4 x_3 \\
& + K_1 K_2 K_3 k_1 k_3 k_4 (k_1 k_4 - k_2 k_3) x_1^3 x_2^2 x_3 + K_2^2 K_3 k_1^2 k_4 (k_1 k_4 - k_2 k_3) x_1^3 x_2 x_3^2 \\
& + 2\, K_1 K_2 K_3 K_4 k_1^2 k_3 k_2 k_4 x_1^3 x_2 x_3 + K_1 K_2 K_3 k_1 k_3 k_4 (k_1 k_4 - k_2 k_3) x_1^2 x_2^3 x_3 \\
& + (K_1^2 K_2 K_3 k_1 k_3^2 k_4 (k_2 + k_4) x_1^2 x_2^3 + K_1 K_2 K_3 k_1 k_3 k_4 (k_1 k_4 - k_2 k_3) x_1^2 x_2^2 x_3^2 \\
& + K_1 K_2 K_3 k_1 k_3 k_4 ((K_2 + K_3) k_1 k_4 - (K_1 + K_4) k_2 k_3) x_1^2 x_2^2 x_3 \\
& + K_1^2 K_2 K_3 K_4 k_1 k_2 k_3^2 k_4 x_1^2 x_2^2 + K_1^2 K_3^2 k_3^2 k_4^2 (k_1 + k_3) x_1 x_2^4 + 2 K_1^2 K_2 K_3 k_1 k_3^2 k_4^2 x_1 x_2^3 x_3 \\
& + K_1^2 K_2 K_3^2 k_1 k_3^2 k_4^2 x_1 x_2^3 + K_1^2 K_3^2 k_3^3 k_4^2 x_2^4 x_3 + K_1^3 K_3^2 k_3^3 k_4^2 x_2^4
\end{aligned}$$

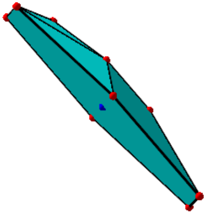
$$b_1(\kappa) = k_1 k_4 - k_2 k_3, \qquad b_2(\kappa) = k_1 k_4 (K_2 + K_3) - k_2 k_3 (K_1 + K_4)$$

$$b_1(\kappa) = k_1 k_4 - k_2 k_3, \qquad b_2(\kappa) = k_1 k_4 (K_2 + K_3) - k_2 k_3 (K_1 + K_4)$$

- One steady state ($p(x) > 0$): $b_1(\kappa) \geq 0$ and $b_2(\kappa) \geq 0$
- Multistationarity: $b_1(\kappa) < 0$
  ($b_1(\kappa)$ is the coefficient of a vertex of the Newton polytope, so $p(x) < 0$ is possible)

What happens when $b_2(\kappa) < 0$ and $b_1(\kappa) \geq 0$?

This coefficient is not of a vertex of the Newton polytope.



$$b_1(\kappa) = k_1 k_4 - k_2 k_3, \qquad b_2(\kappa) = k_1 k_4 (K_2 + K_3) - k_2 k_3 (K_1 + K_4)$$

- One steady state: $b_1(\kappa) \geq 0$ and $b_2(\kappa) \geq 0$
- Multistationarity: $b_1(\kappa) < 0$
  ($b_1(\kappa)$ is the coefficient of a vertex of the Newton polytope)

What happens when $b_2(\kappa) < 0$ and $b_1(\kappa) \geq 0$?

It depends on how large $b_2(\kappa)$ is with respect to the other coefficients, so the exponents are not enough

$$x^2 - 2xy + y^2 = (x-y)^2 \geq 0$$

but

$$x^2 - 3xy + y^2 = (x-y)^2 - xy < 0, \qquad \text{whenever } x = y$$

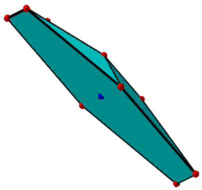
Newton polytope again

Given a face τ of the Newton polytope, let $f_\tau(x)$ be the restriction of $f(x)$ to the monomials supported in the face.

Then for any $x^* \in \mathbb{R}^n_{>0}$ there exists $y^* \in \mathbb{R}^n_{>0}$ such that

$$\mathrm{sign}(f(y^*)) = \mathrm{sign}(f_\tau(x^*)).$$

Restrict the polynomial to the face



$$\begin{aligned} p(x) = {} & K_2^2K_4k_1^2k_2(k_1k_4 - k_2k_3)x_1^4x_3^2 + K_1K_2^2K_4k_1^2k_3k_2^2x_1^4x_3 \\ & + K_2^2K_3k_1^2k_4(k_1k_4 - k_2k_3)x_1^3x_2x_3^2 \\ & + 2\,K_1K_2K_3K_4k_1^2k_3k_2k_4x_1^3x_2x_3 + K_1^3K_3^2k_3^3k_4^2x_2^4 \\ & + K_1K_2K_3k_1k_3k_4((K_2+K_3)k_1k_4 - (K_1+K_4)k_2k_3)x_1^2x_2^2x_3 \\ & + K_1K_2K_3k_1k_3k_4(k_1k_4 - k_2k_3)x_1^2x_2^2x_3^2 \\ & + K_1^2K_2K_3K_4k_1k_2k_3^2k_4x_1^2x_2^2 + 2K_1^2K_2K_3k_1k_3^2k_4^2x_1x_2^3x_3. \\ & + K_1^2K_2K_3^2k_1k_3^2k_4^2x_1x_2^3 + K_1^2K_3^2k_3^3k_4^2x_2^4x_3 \end{aligned}$$

Circuit numbers

Multivariate polynomial in n variables

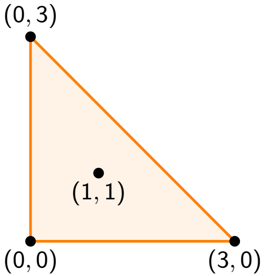
$$p(x) = -\beta x^c + \sum_{i=0}^{m} \alpha_i x^{v_i}$$

$v_0, \ldots, v_m$ define an m-dimensional simplex Δ in $\mathbb{R}^n$.
c is in the relative interior of Δ.
$\alpha_i, \beta > 0$.

Theorem: Let $\lambda_0, \ldots, \lambda_m$ be the convex coordinates of c in $v_0, \ldots, v_m$. Then, $p(x) \geq 0$ for all positive x if and only if

$$\beta \leq \prod_{i=0}^{m} \left(\frac{\alpha_i}{\lambda_i}\right)^{\lambda_i}$$

Iliman, de Wolff (2016)



$p(x, y) = \alpha_0 + \alpha_1 x^3 + \alpha_2 y^3 - \beta xy$, $\beta > 0$

$(1, 1) = \frac{1}{3}(0, 0) + \frac{1}{3}(3, 0) + \frac{1}{3}(0, 3)$.

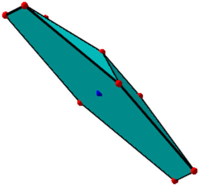
p nonnegative for all $x, y > 0$ if and only if

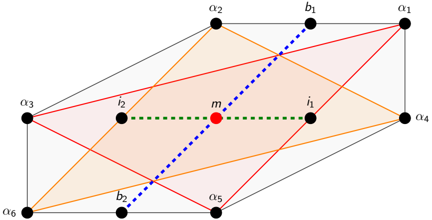
$$\beta \leq 3(\alpha_0\alpha_1\alpha_2)^{1/3}$$

Back to our example

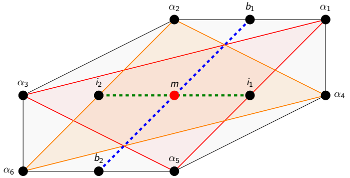
$$p(x) = K_2^2 K_4 k_1^2 k_2 (k_1 k_4 - k_2 k_3) x_1^4 x_3^2 + K_1 K_2^2 K_4 k_1^2 k_3 k_2^2 x_1^4 x_3 + K_2^2 K_3 k_1^2 k_4 (k_1 k_4 - k_2 k_3) x_1^3 x_2 x_3^2$$
$$+ 2 K_1 K_2 K_3 K_4 k_1^2 k_3 k_2 k_4 x_1^3 x_2 x_3 + K_1^3 K_3^2 k_3^3 k_4^2 x_2^4 + K_1 K_2 K_3 k_1 k_3 k_4 (k_1 k_4 - k_2 k_3) x_1^2 x_2^2 x_3^2$$
$$+ K_1 K_2 K_3 k_1 k_3 k_4 ((K_2 + K_3) k_1 k_4 - (K_1 + K_4) k_2 k_3) x_1^2 x_2^2 x_3 + K_1^2 K_2 K_3 K_4 k_1 k_2 k_3^2 k_4 x_1^2 x_2^2$$
$$+ 2 K_1^2 K_2 K_3 k_1 k_3^2 k_4^2 x_1 x_2^3 x_3 + K_1^2 K_2 K_3^2 k_1 k_3^2 k_4^2 x_1 x_2^3 + K_1^2 K_3^2 k_3^3 k_4^2 x_2^4 x_3$$

We assume $b_1(\kappa) \geq 0$ and $b_2(\kappa) < 0$





Partial results



- Assume $b_1(\kappa) \geq 0$ and $b_2(\kappa) < 0$. Then if

$$-b_2(\kappa) \leq 3(\alpha_{a_1}\alpha_{a_3}\alpha_{a_5})^{\frac{1}{3}} + 3(\alpha_{a_2}\alpha_{a_4}\alpha_{a_6})^{\frac{1}{3}} + 2(\alpha_{b_1}\alpha_{b_2})^{\frac{1}{2}} + 2(\alpha_{i_1}\alpha_{i_2})^{\frac{1}{2}},$$

then $p(x) > 0$ for all positive x, and hence multistationarity is not enabled.

- For any $K_2, K_3, K_4, k_1, k_2, k_3, k_4$, multistationarity is enabled for K_1 large enough and is also enabled for K_4 large enough

Feliu, Kaihnsa, de Wolff, Yürück (2020), arXiv

Reaction rate constants enabling multistationarity for the 2-site phosphorylation cycle

We have proper inclusions (with set difference of non-zero measure)

$$\begin{aligned}\{\kappa \mid b_1(\kappa) < 0\} &\subseteq \{\kappa \mid \kappa \text{ enables multistationarity }\} \\ &\subseteq \{\kappa \mid b_1(\kappa) < 0\} \cup \{\kappa \mid b_1(\kappa) \geq 0,\ b_2(\kappa) < 0\}\end{aligned}$$

We have an explicit parametrization of the boundary between the κ's that enable multistationarity and those that do not.

- Both the mono and multi regions are connected (in parameter space)
- The multi region is open, and the mono region is closed.

Feliu, Kaihnsa, de Wolff, Yürück (2020), arXiv

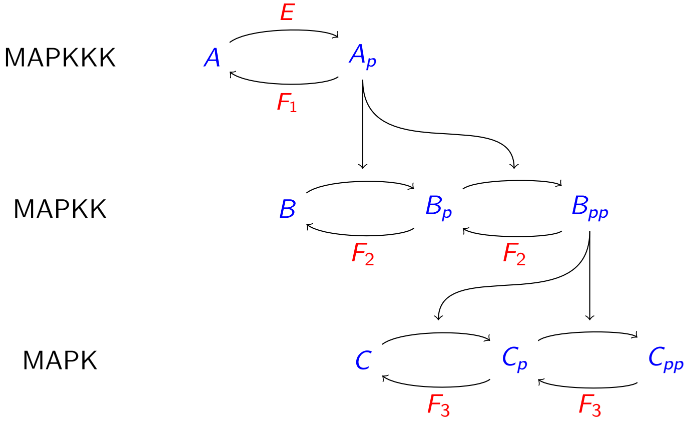
Understanding the signs a multivariate polynomial can attain

is a recurrent problem in this field.

- Deciding on stability
- Deciding on oscillations

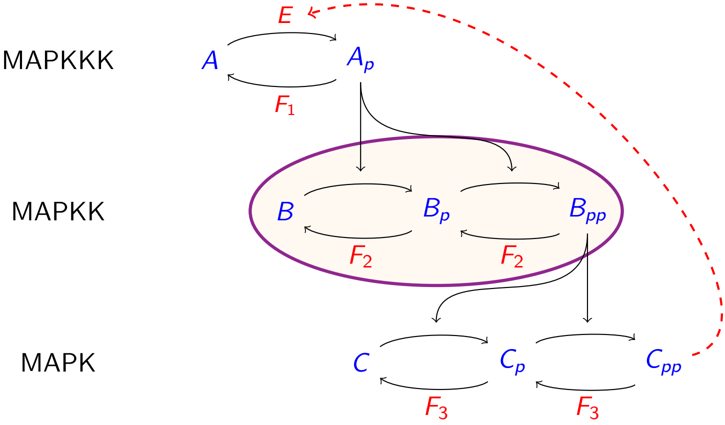
Oscillations in the MAPK cascade

MAPK cascade



Huang, Ferrell model, ‘99

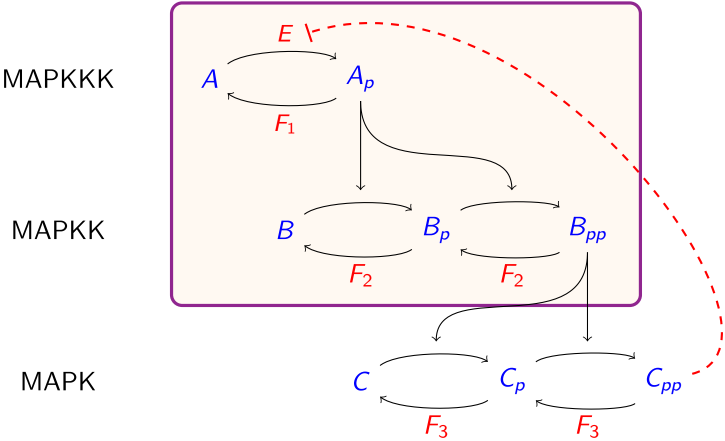
MAPK cascade. Bistability



Huang, Ferrell model, ‘99

Markevich, Hoeck, Kholodenko, ‘04

MAPK cascade. Oscillations



Suggest: Single-stage bistability is necessary for the oscillatory behavior

Kholodenko, '00

Qiao, Nachbar, Kevrekidis, Shvartsman, '07

Double-phosphorylation cycle

Does the double-phosphorylation cycle admit oscillations?

$$\begin{tikzcd} B \arrow[r, bend left, "E"] & B_p \arrow[l, bend left, "F"] \arrow[r, bend left, "E"] & B_{pp} \arrow[l, bend left, "F"] \end{tikzcd}$$

How do we decide this? Standard approach is to find a Hopf bifurcation.

- A Hopf bifurcation in a parameter (μ) dependent ODE $\dot{x} = f(x)$, arises at μ^* when a pair of eigenvalues of the Jacobian evaluated at a curve of steady states, $J_f(x^*(\mu))$, crosses the imaginary axes, and the steady state goes from stable to unstable.
  In this case a periodic solution arises after μ^*.
- In our setting: This means that there exists a choice of parameters κ, and a corresponding steady state x^* such that the Jacobian has a pair of imaginary eigenvalues.

Double-phosphorylation cycle

Does the double-phosphorylation cycle admit oscillations?

$$\begin{tikzcd} B \arrow[r, bend left, "E"] & B_p \arrow[l, bend left, "F"] \arrow[r, bend left, "E"] & B_{pp} \arrow[l, bend left, "F"] \end{tikzcd}$$

- Several (failed) attempts by different groups to show the existence of Hopf bifurcations
- When F acts processively, the network has Hopf bifurcations (Conradi, Mincheva, Shiu '19)

The origin of oscillations in the MAPK cascade

Our results suggest

The double-phosphorylation cycle does not admit Hopf bifurcations

$$\begin{tikzcd} B \arrow[r, bend left, "E"] & B_p \arrow[l, bend left, "F"] \arrow[r, bend left, "E"] & B_{pp} \arrow[l, bend left, "F"] \end{tikzcd}$$

Conradi, Feliu, Mincheva

A single-phosphorylation cascade admits oscillations

$$\begin{tikzcd} A \arrow[r, bend left, "E"] & A_p \arrow[l, bend left, "F_1"] \arrow[d] & \\ B \arrow[r, bend left] & B_p \arrow[l, bend left, "F_2"] & \end{tikzcd}$$

Torres, Feliu

Hurwitz

When does a real polynomial have a pair of imaginary roots?

$$p(z) = \alpha_0 z^n + \alpha_1 z^{n-1} + \cdots + \alpha_{n-1} z + \alpha_n, \qquad \alpha_0 > 0,$$

$$H = \begin{bmatrix} \alpha_1 & \alpha_3 & \alpha_5 & \dots & \dots & 0 \\ \alpha_0 & \alpha_2 & \alpha_4 & \alpha_6 & \dots & 0 \\ 0 & \alpha_1 & \alpha_3 & \alpha_5 & \dots & 0 \\ 0 & \alpha_0 & \alpha_2 & \alpha_4 & \dots & \\ \vdots & \vdots & \vdots & \vdots & \vdots & \alpha_n \end{bmatrix} \qquad H_i = i\text{-th principal minor}$$

- (Liu): $p(z)$ has a simple pair of imaginary roots and the rest of the roots have negative real part, if and only if

$$H_1 > 0, \dots, H_{n-2} > 0, \qquad H_{n-1} = 0, \qquad \alpha_n > 0.$$

- We apply the criterion to the characteristic polynomial of the Jacobian of $Nv_\kappa(x)$ evaluated at a parametrisation of the steady states, after removing $d = n - \mathrm{Rank}(N)$ zero roots:

$$\mathrm{ch}(\lambda) = \lambda^d \big(a_0 \lambda^s + a_1 \lambda^{s-1} + \cdots + a_{s-1}\lambda + a_s \big)$$

Example: enzymatic transfer of calcium ions

$X_1 =$ cytosolic calcium Ca^{++},
$X_2 = \mathrm{Ca}^{++}$ in the endoplasmic reticulum,
$X_3 =$ enzyme catalyzing the transport

$$
\begin{aligned}
0 &\underset{\kappa_2}{\overset{\kappa_1}{\rightleftharpoons}} X_1 \\
X_1 + X_2 &\xrightarrow{\kappa_3} 2X_1 \\
X_1 + X_3 &\underset{\kappa_5}{\overset{\kappa_4}{\rightleftharpoons}} X_4 \xrightarrow{\kappa_6} X_2 + X_3
\end{aligned}
$$

$$
\begin{aligned}
\dot{x}_1 &= \kappa_1 - \kappa_2 x_1 + \kappa_3 x_1 x_2 - \kappa_4 x_1 x_3 + \kappa_5 x_4 \\
\dot{x}_2 &= -\kappa_3 x_1 x_2 + \kappa_6 x_4 \\
\dot{x}_3 &= -\kappa_4 x_1 x_3 + \kappa_5 x_4 + \kappa_6 x_4 \\
\dot{x}_4 &= \kappa_4 x_1 x_3 - \kappa_5 x_4 - \kappa_6 x_4
\end{aligned}
$$

The set of steady states is parametrized by x_4

$$
x_1 = \frac{\kappa_1}{\kappa_2}, \qquad x_2 = \frac{\kappa_2\kappa_6 x_4}{\kappa_1\kappa_3}, \qquad x_3 = \frac{\kappa_2(\kappa_5+\kappa_6) x_4}{\kappa_1\kappa_4}
$$

Gatermann, Eiswirth, Sensse, '05

$$0 \underset{\kappa_2}{\overset{\kappa_1}{\rightleftharpoons}} X_1$$
$$X_1 + X_2 \xrightarrow{\kappa_3} 2X_1$$
$$X_1 + X_3 \underset{\kappa_5}{\overset{\kappa_4}{\rightleftharpoons}} X_4 \xrightarrow{\kappa_6} X_2 + X_3$$

$X_1 =$ cytosolic calcium Ca^{++},
$X_2 = \mathrm{Ca}^{++}$ in the endoplasmic reticulum
$X_3 =$ enzyme catalyzing the transport

The Hurwitz determinants of the characteristic polynomial of the Jacobian of the system evaluated at this parametrization are $(a_1(\kappa), \dots, a_4(\kappa) > 0)$

$$H_1 = a_1(\kappa)(\kappa_2^2\kappa_5 x_4 + \kappa_1^2\kappa_3 + \kappa_1^2\kappa_4 + \kappa_1\kappa_2^2 + \kappa_1\kappa_2\kappa_5 + \kappa_1\kappa_2\kappa_6)$$
$$H_2 = a_2(\kappa)(\kappa_2^4\kappa_5(\kappa_3\kappa_5 + \kappa_3\kappa_6 - \kappa_4\kappa_6)x_4^2 + a_5(\kappa)x_4 + a_3(\kappa))$$
$$\alpha_3 = a_4(\kappa)(\kappa_1\kappa_3(\kappa_1\kappa_4 + \kappa_2\kappa_5 + \kappa_2\kappa_6)$$

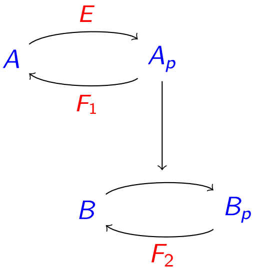
$H_2 = 0$ for some steady state x_4, and hence there is a pair of imaginary eigenvalues if and only if $(\kappa_3\kappa_5 + \kappa_3\kappa_6 - \kappa_4\kappa_6) < 0$, or equivalently

$$\kappa_3 < \frac{\kappa_6\kappa_4}{\kappa_5+\kappa_6}.$$

By taking $T = x_3 + x_4$ as the bifurcation parameter, we confirm that there is a Hopf bifurcation for almost all parameter values (extra condition is satisfied).

- This approach has been used for several reaction networks, eg. Weber et al, Shiu et al. ...
- High computational complexity: The polynomials are HUGE. Often not computable.
- If $H_{n-1}(x)$ has both positive and negative terms, can we guarantee $H_{n-1}(x) = 0$ while the other H's are positive?
- Theoretically, quantifier elimination techniques or optimization methods should solve the sign problem, but are impractical.
- The use of convex coordinates for mass-action type kinetics simplifies slightly the computational cost.

A single-phosphorylation cascade



Full model

$$
\begin{aligned}
A + E &\rightleftharpoons X_1 \longrightarrow A_p + E\\
A_p + F_1 &\rightleftharpoons X_2 \longrightarrow A + F_1\\
B + A_p &\rightleftharpoons Y_1 \longrightarrow B_p + A_p\\
B_p + F_2 &\rightleftharpoons Y_2 \longrightarrow B + F_2
\end{aligned}
$$

Reduced model

$$
\begin{aligned}
A + E &\longrightarrow X_1 \longrightarrow A_p + E\\
A_p + F_1 &\longrightarrow X_2 \longrightarrow A + F_1\\
B + A_p &\longrightarrow Y_1 \longrightarrow B_p + A_p\\
B_p + F_2 &\longrightarrow B + F_2
\end{aligned}
$$

A single-phosphorylation cascade

For the reduced network:

- $H_1, H_2, H_3 > 0$, $\alpha_5 > 0$ for all parameter choices.
- H_4 has 37,235 terms in x and κ with both negative and positive coefficients.
- We set some x and κ equal to one, and reduce to a polynomial that has 9,642 terms, still with negative and positive coefficients.
- We find the Newton polytope. Most vertices have positive coefficients, but 3 have negative coefficient.
- We can find values of x and κ for which $H_4 = 0$.

> The reduced model has a Hopf bifurcation with bifurcation parameter the total amount of A. The Hopf bifurcation is supercritical leading to stable periodic solutions, which can be lifted to the full network for some parameter values.

Work in progress: find the oscillations numerically.

Problem: parameters arising from the Newton polytope are either very large or very small.

(**Torres**, Feliu '20)

Double-phosphorylation cycle

$$\begin{tikzcd}
B \arrow[r, bend left, "E"] & B_p \arrow[l, bend left, "F"] \arrow[r, bend left, "E"] & B_{pp} \arrow[l, bend left, "F"]
\end{tikzcd}$$

Full system

$$B + E \rightleftharpoons X_1 \longrightarrow B_p + E \rightleftharpoons X_2 \longrightarrow B_{pp} + E$$
$$B_{pp} + F \rightleftharpoons Y_2 \longrightarrow B_p + F \rightleftharpoons Y_1 \longrightarrow B + F$$

Reduced systems: irreversible reactions and keep two intermediates. For example

$$B + E \longrightarrow X_1 \longrightarrow B_p + E \longrightarrow B_{pp} + E$$
$$B_{pp} + F \longrightarrow Y_2 \longrightarrow B_p + F \longrightarrow B + F$$

$$B+E \rightleftharpoons X_1 \longrightarrow B_p+E \rightleftharpoons X_2 \longrightarrow B_{pp}+E$$
$$B_{pp}+F \rightleftharpoons Y_2 \longrightarrow B_p+F \rightleftharpoons Y_1 \longrightarrow B+F$$

- H_3 is not a sum of positive terms only with the pair X_1, Y_2.
- $H_3 = 0$ implies $\alpha_4 < 0$, meaning we find a pair of symmetric real eigenvalues.
- We extract the conditions on the parameters under which $\alpha_4 > 0$:

  $(h_6 - h_1)(h_2 + h_5 + h_7)h_3 h_4 + (h_7 - h_2)(h_1 + h_3 + h_6)h_4 h_5 + (h_6 h_7 - h_1 h_2)h_3 h_5.$

  Then, we impose the conditions on H_3 using symbolic software, and confirm that it becomes strictly positive.
- No reduced network with two intermediates admits a Hopf bifurcation.
- The same analysis extends to any choice of three intermediates.
- Conjecture (Feliu): The double-phosphorylation cycle does not admit Hopf bifurcations.
- The same strategy should work, but memory problems!
  Work in progress: find strategies to be able to perform the computations.

Conradi, Feliu, Mincheva '19

Stability

- If there is one steady state, is it asymptotically stable?
- If there are several steady states, which ones are asymptotically stable and which ones are unstable?
- If there are three, are two asymptotically stable and one unstable?

Routh-Hurwitz

Given a real polynomial

$$p(z) = \alpha_0 z^n + \alpha_1 z^{n-1} + \cdots + \alpha_{n-1} z + \alpha_n, \qquad \alpha_0 > 0,$$

how many roots have positive real part and how many have negative real part?

$$H = \begin{bmatrix} \alpha_1 & \alpha_3 & \alpha_5 & \dots & \dots & 0 \\ \alpha_0 & \alpha_2 & \alpha_4 & \alpha_6 & \dots & 0 \\ 0 & \alpha_1 & \alpha_3 & \alpha_5 & \dots & 0 \\ 0 & \alpha_0 & \alpha_2 & \alpha_4 & \dots & \\ \vdots & \vdots & \vdots & \vdots & \vdots & \alpha_n \end{bmatrix}.$$

Criterion:

- If all leading principal minors of H have positive sign, then all roots of the polynomial $p(z)$ have negative real part.
- If not, if none is zero, then the number of roots with positive real part can be determined.

Routh-Hurwitz for reaction networks

- Apply the criterion to the characteristic polynomial of the Jacobian of $Nv_\kappa(x)$ evaluated at a parametrisation of the steady states, after removing $d = n - \text{Rank}(N)$ zero roots:

$$\mathrm{ch}(\lambda) = \lambda^d \big(a_0 \lambda^s + a_1 \lambda^{s-1} + \cdots + a_{s-1}\lambda + a_s \big)$$

  If all roots of the right product have negative real part, then the steady state is asymptotically stable (relative to its class).

- We obtain a collection of polynomials/rational functions: if all have only positive coefficients, then ALL steady states are asymptotically stable.

One positive steady state in each class, which is asymptotically stable:

Two-component system

$$\begin{aligned} \mathrm{HK} &\longrightarrow \mathrm{HK_p} \\ \mathrm{HK_p} + \mathrm{RR} &\rightleftharpoons \mathrm{HK} + \mathrm{RR_p} \\ \mathrm{RR_p} &\longrightarrow \mathrm{RR} \end{aligned}$$

Two substrate enzyme catalysis

$$\begin{aligned} E + S_1 &\rightleftharpoons ES_1 & E + S_2 &\rightleftharpoons ES_2 \\ S_2 + ES_1 &\rightleftharpoons ES_1S_2 & S_1 + ES_2 &\rightleftharpoons ES_1S_2 \\ ES_1S_2 &\rightleftharpoons E + P \end{aligned}$$

Routh-Hurwitz for reaction networks

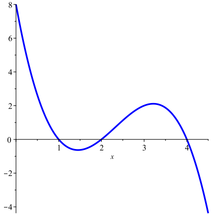
- If not all polynomials have positive coefficients, one should proceed to analyse the Newton polytope. Problem: The polynomials are HUGE. Often not computable.

For small networks we often have

- All Hurwitz determinants are positive except maybe for the last one, which is $a_s H_{s-1}$. Then, the steady state is asymptotically stable if $a_s > 0$ and unstable if $a_s < 0$.
- It is possible to reduce the equations defining $C_{\kappa,c}$ to one polynomial equation $q_{\kappa,c}(t) = 0$, where $t = x_i$ and the x_j are positive rational functions of t.
- For a steady state x^*

$$\mathrm{sign}(a_s(x^*)) = \mathrm{sign}(q'_{\kappa,c}(t(x^*))) \cdot \epsilon, \quad \epsilon \in \{-1, 1\}$$

- "The stability of the steady states alternates with t".



Torres, Feliu (2019), arXiv, also Torres PhD thesis

Hybrid histidine kinase

$$\mathrm{HK}_{00} \xrightarrow{\kappa_1} \mathrm{HK}_{\mathrm{p}0} \xrightarrow{\kappa_2} \mathrm{HK}_{0\mathrm{p}} \xrightarrow{\kappa_3} \mathrm{HK}_{\mathrm{pp}}$$
$$\mathrm{HK}_{0\mathrm{p}} + \mathrm{RR} \xrightarrow{\kappa_4} \mathrm{HK}_{00} + \mathrm{RR}_{\mathrm{p}}$$
$$\mathrm{HK}_{\mathrm{pp}} + \mathrm{RR} \xrightarrow{\kappa_5} \mathrm{HK}_{\mathrm{p}0} + \mathrm{RR}_{\mathrm{p}}$$
$$\mathrm{RR}_p \xrightarrow{\kappa_6} \mathrm{RR}$$

Multi $\Leftrightarrow \kappa_1 < \kappa_3$

Gene transcription network

$$X_1 \longrightarrow X_1 + P_1 \qquad P_1 \longrightarrow 0$$
$$X_2 \longrightarrow X_2 + P_2 \qquad P_2 \longrightarrow 0$$
$$X_2 + P_1 \rightleftharpoons X_2P_1 \qquad 2P_2 \rightleftharpoons P_2P_2$$
$$X_1 + P_2P_2 \rightleftharpoons X_1P_2P_2$$

Multi for all κ

For both networks $q_{\kappa,c}(t)$ has degree 3.

> Whenever the network has three steady states, two are asymptotically stable and one is unstable.

- Combined with results on lifting bistability (like removal of intermediates or making a reaction irreversible), we can assert bistability for many realistic networks.

Lifting bistability

There exist regions in the parameter space for which the following networks have two asymptotically stable steady states and one unstable steady state.

2-site phosphorylation cycle

$$A + K \rightleftharpoons AK \longrightarrow A_p + K \rightleftharpoons A_pK \longrightarrow A_{pp} + K$$
$$A_{pp} + F \rightleftharpoons A_{pp}F \longrightarrow A_p + F \rightleftharpoons A_pF \longrightarrow A + F$$

Phosphorylation of two substrates

$$A + K \rightleftharpoons AK \longrightarrow A_p + K \qquad A_p + F \rightleftharpoons A_pF \longrightarrow A + F$$
$$B + K \rightleftharpoons BK \longrightarrow B_p + K \qquad B_p + F \rightleftharpoons B_pF \longrightarrow B + F$$

Allosteric kinase

$$\mathrm{K_r} + \mathrm{S} \rightleftharpoons \mathrm{K_rS} \longrightarrow \mathrm{K_r} + \mathrm{S_p} \qquad \mathrm{K_r} \rightleftharpoons \mathrm{K_t}$$
$$\mathrm{K_t} + \mathrm{S} \rightleftharpoons \mathrm{K_tS} \longrightarrow \mathrm{K_t} + \mathrm{S_p} \qquad \mathrm{K_rS} \rightleftharpoons \mathrm{K_tS} \qquad \mathrm{S_p} \longrightarrow \mathrm{S}$$

... and many more!

Thank you for your attention