Inverse dynamical analysis

a methodology for probing dynamics in gene regulation and metabolism

James Lu
Inverse Problems Group
Johann Radon Institute for Computational and Applied Mathematics (RICAM)
Linz, Austria

Rainer Machné
Theoretical Biochemistry Group (TBI)
University of Vienna
Vienna, Austria

In collaboration with: **Heinz Engl** (RICAM), **Peter Schuster** (TBI, Vienna)

BIRD, Berlin, March 12-14 2007

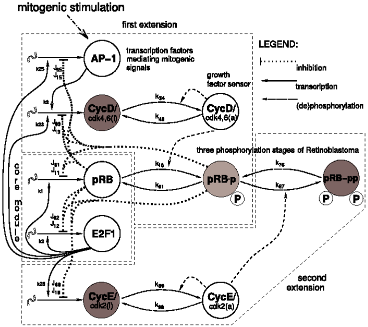
Outline

- Gene regulation models: G_1/S module, GATA networks
  - Auto-regulatory feedback loops
- The Forward Problem
  - Dynamics and Bifurcations
- Inverse Bifurcation Analysis
  - Numerical Results
  - Biological interpretation
- Inverse Eigenvalue Analysis
  - Evolution of the *bifurcation phenotype*

Outline

- Gene regulation models: G_1/S module, GATA networks
  - Auto-regulatory feedback loops
- The Forward Problem
  - Dynamics and Bifurcations
- Inverse Bifurcation Analysis
  - Numerical Results
  - Biological interpretation
- Inverse Eigenvalue Analysis
  - Evolution of the *bifurcation phenotype*

The G_1/S Module of Mammalian Cell Cycle



- What can be said about the core regulatory mechanism of the bifurcation points?
  - Inverse Bifurcation and Eigenvalue Analysis

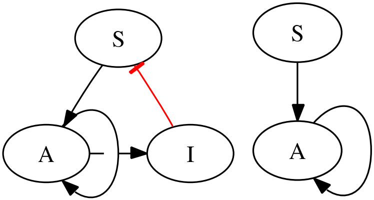
M. Swat, A. Kel, H. Herzel, *Bioinformatics* (2004)

from Bio to Math and back

The Forward Problem: bottom-up modeling

interaction graph $\longrightarrow$ reactions $\longrightarrow \frac{dx}{dt} = f(x, \alpha) \longrightarrow x(t) \longrightarrow$ bifurcations

interaction graph



reaction network

$$S_{\text{nuclear}} \longrightarrow S_{\text{nuclear}} + A_{\text{RNA}}$$
$$A_{\text{RNA}} \longrightarrow A_{\text{mRNA}}$$
$$A_{\text{mRNA}} \longrightarrow A_{\text{mRNA}} + A_{\text{protein}}$$
$$A_{\text{protein}} \longleftrightarrow A_{\text{nuclear}}$$
$$A_{\text{nuclear}} \longrightarrow A_{\text{nuclear}} + A_{\text{RNA}}$$
$$A_x \longrightarrow$$

compiling decades of diploma and PhD students' wet lab work

1. stoichiometry: analysis
2. chemical kinetics

from Bio to Math and back

The Forward Problem:

interaction graph $\longrightarrow$ reactions $\longrightarrow \frac{dx}{dt} = f(x, \alpha) \longrightarrow x(t) \longrightarrow$ bifurcations

equations

$$
\begin{aligned}
\frac{drA}{dt} &= N \cdot \left(V_a^b + V_a^A \cdot \left(\frac{A}{\left(K_a^A + A\right)} \right)^{hA} + V_a^S \cdot \left(\frac{S}{\left(K_a^S + S\right)} \right) \right) - (kex_a + \Phi_r A) \cdot rA \\
\frac{dmA}{dt} &= kex_a \cdot rA \cdot c1/c2 - \Phi_m A \cdot mA \\
\frac{dAc}{dt} &= ktl_a \cdot mA + kex_A \cdot A \cdot c1/c2 - (kim_A + \Phi_{Ac}) \cdot Ac \\
\frac{dA}{dt} &= kim_A \cdot Ac - (kex_A + \Phi_A) \cdot A
\end{aligned}
$$

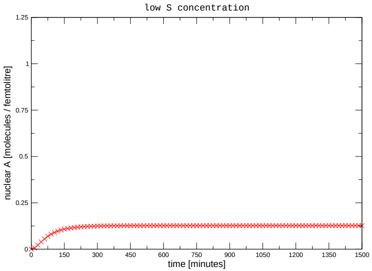
dynamical systems theory:
ODEs, PDEs, Markov process, Boolean nw., Bayesian nw., Petri nets, ...

from Bio to Math and back

The Forward Problem:

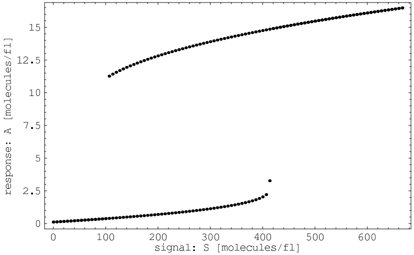
interaction graph $\longrightarrow$ reactions $\longrightarrow \frac{dx}{dt} = f(x, \alpha) \longrightarrow x(t) \longrightarrow$ bifurcations

time courses: $x(t)$



still rarely availabe from wet labs
(steady-state paradigm)

stability and bifurcation analysis

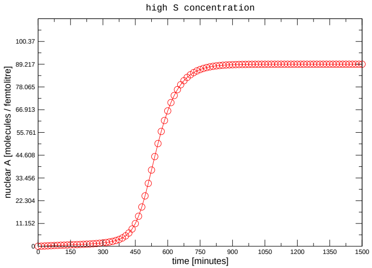


from Bio to Math and back

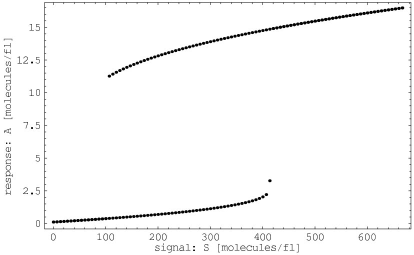
The Forward Problem:

$$\text{interaction graph} \rightarrow \text{reactions} \rightarrow \frac{dx}{dt} = f(x, \alpha) \rightarrow x(t) \rightarrow \text{bifurcations}$$

time courses: $x(t)$



stability and bifurcation analysis



still rarely availabe from wet labs

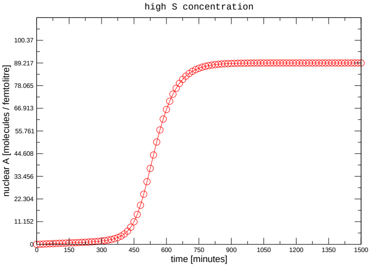
(steady-state paradigm)

from Bio to Math and back : *bifurcation phenotypes*

The Forward Problem:

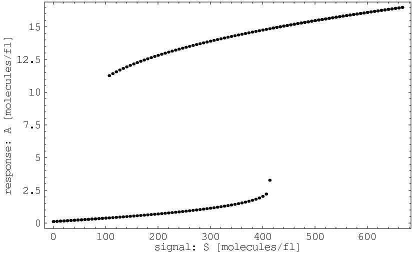
interaction graph $\to$ reactions $\to$ $\frac{dx}{dt} = f(x, \alpha) \to x(t) \to$ bifurcations

time courses: $x(t)$



still rarely availabe from wet labs *(steady-state paradigm)*

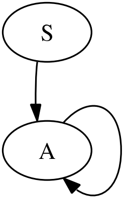
stability and bifurcation analysis
w.r.t. physiological signals

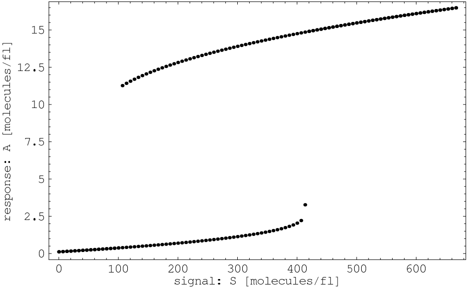


$\approx$ signal/response curves from titration experiments

Feedback and Bifurcations I: hysteresis/bistability

2 saddle-node bifurcations : signal switches, memory effects





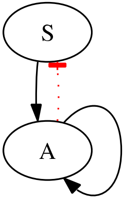
conjecture: *multistability requires positive feedback*

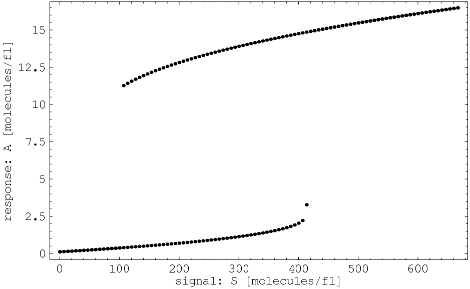
- S: e.g. low nutrient concentration or infection
- S should really decrease significantly to switch-off A
  - $\Rightarrow$ **avoid oscillating deficiency or infection**
- Irreversibility w.r.t. S: terminal differentiation!

Thomas R et al. *Eur J Biochem.* 1976, Thomas R and Kaufman M. *Chaos* 2001, Soulé C. *C R Biol.* 2006

Feedback and Bifurcations I: hysteresis/bistability

2 saddle-node bifurcations : signal switches, memory effects





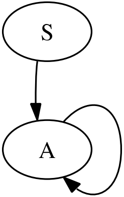
conjecture: *multistability requires positive feedback*

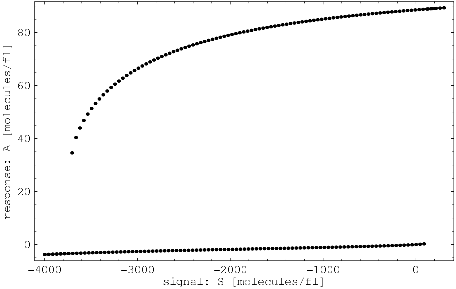
- S: e.g. low nutrient concentration or infection
- S should really decrease significantly to switch-off A
  - $\Rightarrow$ **avoid oscillating deficiency or infection**
- Irreversibility w.r.t. S: terminal differentiation!

Thomas R et al. *Eur J Biochem.* 1976, Thomas R and Kaufman M. *Chaos* 2001, Soulé C. *C R Biol.* 2006

Feedback and Bifurcations I: hysteresis/bistability

2 saddle-node bifurcations : signal switches, memory effects





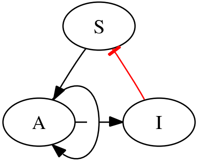
conjecture: *multistability requires positive feedback*

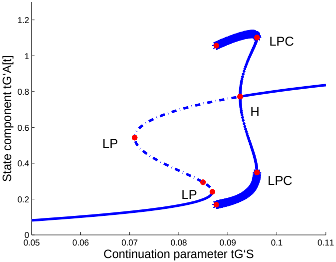
- S: e.g. low nutrient concentration or infection
- S should really decrease significantly to switch-off A
  - $\Rightarrow$ **avoid oscillating deficiency or infection**
- Irreversibility w.r.t. S: terminal differentiation!

Thomas R et al. *Eur J Biochem.* 1976, Thomas R and Kaufman M. *Chaos* 2001, Soulé C. *C R Biol.* 2006

Feedback and Bifurcations II: oscillations

Hopf bifurcations : oscillatory phenomena





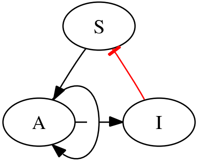
conjecture: *oscillation requires negative feedback and delay*

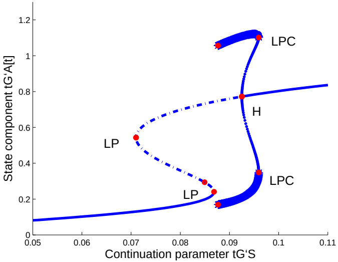
- Life is an intrinsically rhythmic phenomenon:
  - metabolic oscillations, nanovibrations
    - yeast respiration cycles with genome-wide effects, e.g. cell cycle gating!
  - cell cycle
  - circadian clock
  - lunar and solar cycle

Hunding A. *Biophys Struct Mech.* 1974, Klevecz et al. *PNAS* 2004

Feedback and Bifurcations II: oscillations

Hopf bifurcations : oscillatory phenomena



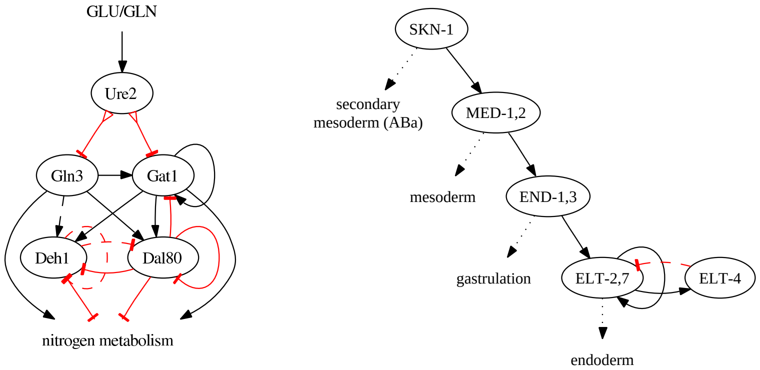


conjecture: *oscillation requires negative feedback and delay*

- Life is an intrinsically rhythmic phenomenon:
  - metabolic oscillations, nanovibrations
    - yeast respiration cycles with genome-wide effects, e.g. cell cycle gating!
  - cell cycle
  - circadian clock
  - lunar and solar cycle

Hunding A. *Biophys Struct Mech.* 1974, Klevecz et al. *PNAS* 2004

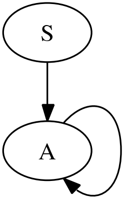
S. cerevisiae and *C. elegans* GATA networks

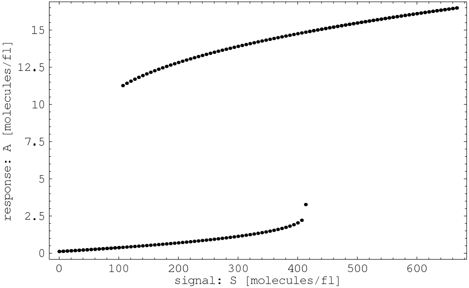


- common origin in auto-activator?
  ⇒ coherent functional story up to mammals
- different duplication and diversification events
- generating cascades by mutation of bindings sites and domains
- generating competitive inhibitors by loss of activating domain

Cooper TG. *FEMS Microbiol Rev.* 2002, Maduro MF, Rothman JH. *Dev Biol.* 2002

Dosage effects: hypersensitivity / haploinsufficiency

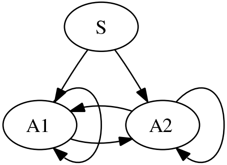


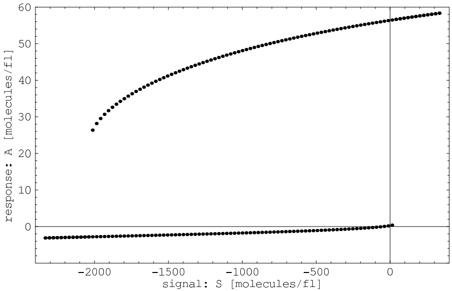


- gene duplication: $N: 1 \longrightarrow 2$
  - **hypersensitivity** or even **irreversibility** wrt to S
- related to **hypersensitive immune response**?
  - IL4 $\longrightarrow$ STAT6 $\longrightarrow$ GATA-3 $\longrightarrow$ GATA-3 $\longrightarrow$ T_{h2} activation
- **haploinsufficiency** $N: 2 \longrightarrow 1$
  - haploinsufficiency diseases known for both GATA-2 (confirmed auto-activator) and GATA-6

Machné R, Lu J, Müller S, Endler L, Widder S, Flamm C, Engl H, Schuster PK, Murray DB et al. *in preparation* 2007

Dosage effects: hypersensitivity / haploinsufficiency



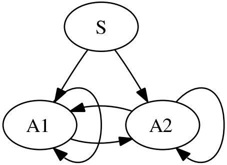


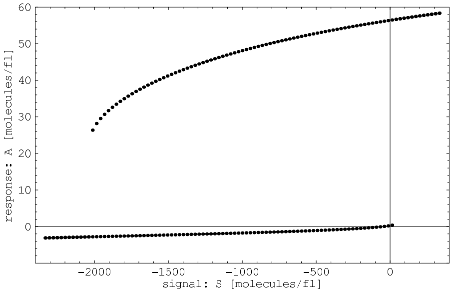
- gene duplication: $N : 1 \rightarrow 2$
  - **hypersensitivity** or even **irreversibility** wrt to S
- related to **hypersensitive immune response**?
  - IL4 → STAT6 → GATA-3 → GATA-3 → T_{h2} activation
- **haploinsufficiency** $N : 2 \rightarrow 1$
  - haploinsufficiency diseases known for both GATA-2 (confirmed auto-activator) and GATA-6

Machné R, Lu J, Müller S, Endler L, Widder S, Flamm C, Engl H, Schuster PK, Murray DB et al. *in preparation* 2007

Susumu Ohno. *Evolution by gene duplication.* 1970

Dosage effects: hypersensitivity / haploinsufficiency

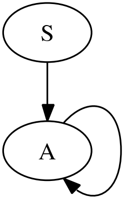


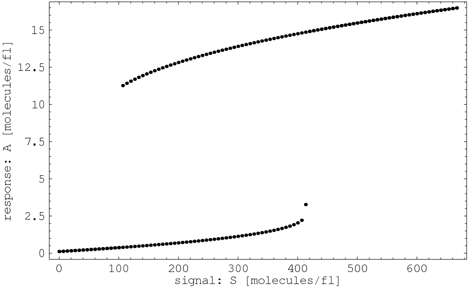


- gene duplication: $N : 1 \rightarrow 2$
  - **hypersensitivity** or even **irreversibility** wrt to S
- related to **hypersensitive immune response**?
  - IL4 $\rightarrow$ STAT6 $\rightarrow$ GATA-3 $\rightarrow$ GATA-3 $\rightarrow$ T_{h2} activation
- **haploinsufficiency** $N : 2 \rightarrow 1$
  - haploinsufficiency diseases known for both GATA-2 (confirmed auto-activator) and GATA-6

Machné R, Lu J, Müller S, Endler L, Widder S, Flamm C, Engl H, Schuster PK, Murray DB et al. *in preparation* 2007
Höfer T et al. *PNAS* 2002

Dosage effects: hypersensitivity / haploinsufficiency





- gene duplication: $N : 1 \rightarrow 2$
  - **hypersensitivity** or even **irreversibility** wrt to S
- related to **hypersensitive immune response**?
  - IL4 $\rightarrow$ STAT6 $\rightarrow$ GATA-3 $\rightarrow$ GATA-3 $\rightarrow$ T_{h2} activation
- **haploinsufficiency** $N : 2 \rightarrow 1$
  - haploinsufficiency diseases known for both GATA-2 (confirmed auto-activator) and GATA-6

Machné R, Lu J, Müller S, Endler L, Widder S, Flamm C, Engl H, Schuster PK, Murray DB et al. *in preparation* 2007
Rodrigues NP et al. *Blood* 2005, Grass JA et al. *Mol Cell Biol.* 2006

Inverse dynamical analysis

- In modelling gene regulation systems, one would like to:
  - probe the possibility for the model to exhibit bistability or oscillations
  - characterize parameter variations that can give rise to different qualitative dynamics
- Given a plausible model:
  - identify mechanisms in model that can give rise to various *bifurcation phenotypes*: verify or falsify experimentally
  - design for desired dynamical characteristics
- Methods:
  - inverse eigenvalue analysis
  - inverse bifurcation analysis
  - *sparsity-promoting* regularization

Inverse dynamical analysis

- In modelling gene regulation systems, one would like to:
  - probe the possibility for the model to exhibit bistability or oscillations
  - characterize parameter variations that can give rise to different qualitative dynamics
- Given a plausible model:
  - identify mechanisms in model that can give rise to various *bifurcation phenotypes*: verify or falsify experimentally
  - design for desired dynamical characteristics
- Methods:
  - inverse eigenvalue analysis
  - inverse bifurcation analysis
  - *sparsity-promoting* regularization

Inverse dynamical analysis

- In modelling gene regulation systems, one would like to:
  - probe the possibility for the model to exhibit bistability or oscillations
  - characterize parameter variations that can give rise to different qualitative dynamics
- Given a plausible model:
  - identify mechanisms in model that can give rise to various *bifurcation phenotypes*: verify or falsify experimentally
  - design for desired dynamical characteristics
- Methods:
  - inverse eigenvalue analysis
  - inverse bifurcation analysis
  - *sparsity-promoting* regularization

Inverse problems

- Forward problem: $\frac{dx}{dt} = f(x, \alpha) \rightarrow x(t) \rightarrow$ bifurcations

$$F(\alpha) = x$$

- Inverse problem: $\frac{dx}{dt} = f(x, \alpha) \leftarrow x(t) \leftarrow$ bifurcations
  - typically *ill-posed* (in the sense of Hadamard)
  - non-uniqueness;
  - instability of inversion
- Variational regularization: add penalty term

$$\min_{\alpha} \|F(\alpha) - x\| + \mu \mathcal{R}(\alpha)$$

- While stabilizing ill-posed problems, regularization brings bias to the solution
- For biological systems, we usually want the solution to be *sparse*, i.e., having as few non-zeros in the solution as possible

Inverse problems

- Forward problem: $\frac{dx}{dt} = f(x, \alpha) \rightarrow x(t) \rightarrow$ bifurcations

$$F(\alpha) = x$$

- Inverse problem: $\frac{dx}{dt} = f(x, \alpha) \leftarrow x(t) \leftarrow$ bifurcations
  - typically *ill-posed* (in the sense of Hadamard)
  - non-uniqueness;
  - instability of inversion
- Variational regularization: add penalty term

$$\min_{\alpha} \|F(\alpha) - x\| + \mu \mathcal{R}(\alpha)$$

- While stabilizing ill-posed problems, regularization brings bias to the solution
- For biological systems, we usually want the solution to be *sparse*, i.e., having as few non-zeros in the solution as possible

Inverse problems

- Forward problem: $\frac{dx}{dt} = f(x, \alpha) \rightarrow x(t) \rightarrow$ bifurcations

$$F(\alpha) = x$$

- Inverse problem: $\frac{dx}{dt} = f(x, \alpha) \leftarrow x(t) \leftarrow$ bifurcations
  - typically *ill-posed* (in the sense of Hadamard)
  - non-uniqueness;
  - instability of inversion
- Variational regularization: add penalty term

$$\min_{\alpha} \|F(\alpha) - x\| + \mu \mathcal{R}(\alpha)$$

- While stabilizing ill-posed problems, regularization brings bias to the solution
- For biological systems, we usually want the solution to be *sparse*, i.e., having as few non-zeros in the solution as possible

Inverse problems

- Forward problem: $\frac{dx}{dt} = f(x, \alpha) \rightarrow x(t) \rightarrow$ bifurcations

$$F(\alpha) = x$$

- Inverse problem: $\frac{dx}{dt} = f(x, \alpha) \leftarrow x(t) \leftarrow$ bifurcations
  - typically *ill-posed* (in the sense of Hadamard)
  - non-uniqueness;
  - instability of inversion
- Variational regularization: add penalty term

$$\min_{\alpha} \|F(\alpha) - x\| + \mu\mathcal{R}(\alpha)$$

- While stabilizing ill-posed problems, regularization brings bias to the solution
- For biological systems, we usually want the solution to be *sparse*, i.e., having as few non-zeros in the solution as possible

Inverse problems

- Forward problem: $\frac{dx}{dt} = f(x, \alpha) \to x(t) \to$ bifurcations

$$F(\alpha) = x$$

- Inverse problem: $\frac{dx}{dt} = f(x, \alpha) \leftarrow x(t) \leftarrow$ bifurcations
  - typically *ill-posed* (in the sense of Hadamard)
  - non-uniqueness;
  - instability of inversion
- Variational regularization: add penalty term

$$\min_{\alpha} \|F(\alpha) - x\| + \mu \mathcal{R}(\alpha)$$

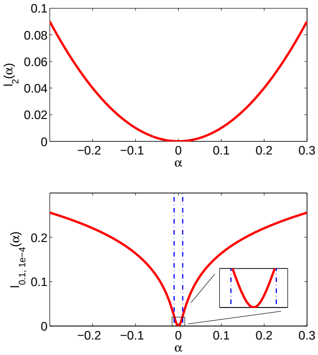
- While stabilizing ill-posed problems, regularization brings bias to the solution
- For biological systems, we usually want the solution to be *sparse*, i.e., having as few non-zeros in the solution as possible

Sparsity-promoting regularization: l_p, $p \leq 1$

- Consider (smoothed) functionals $\mathbb{R}^n \to \mathbb{R}$:
  $l_{p,\epsilon}(\alpha) = \sum_i (\alpha_i^2 + \epsilon)^{p/2}$
- Convex only within the box $\{\alpha : |\alpha_i| < \sqrt{\epsilon},\ 0 < i \leq n\}$

Sparsity-promoting regularization: l_p, $p \leq 1$

- Consider (smoothed) functionals $\mathbb{R}^n \to \mathbb{R}$:
  $l_{p,\epsilon}(\alpha) = \sum_i (\alpha_i^2 + \epsilon)^{p/2}$
- Convex only within the box $\{\alpha : |\alpha_i| < \sqrt{\epsilon},\ 0 < i \leq n\}$

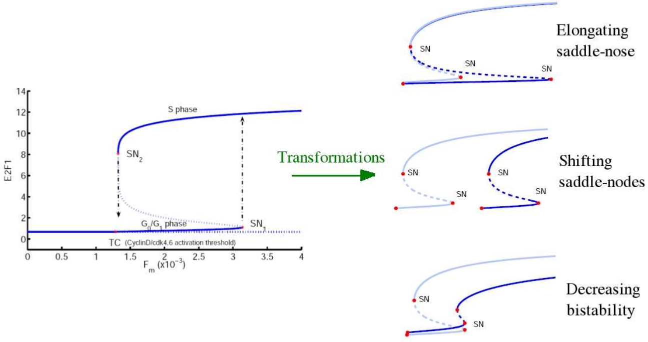


Inverse bifurcation: mammalian G_1/S transition

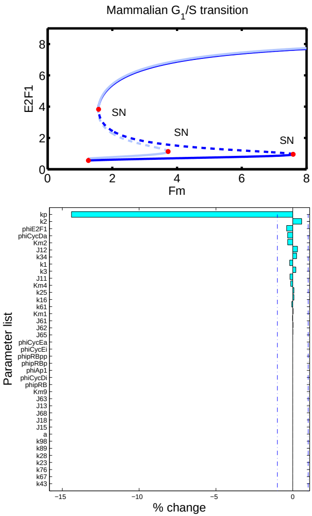
- Map: bifurcation phenotypes $\rightarrow$ parameter sets
- Consider the following 3 modes of geometric transformations of the nominal bifurcation diagram:

Inverse bifurcation: mammalian G_1/S transition

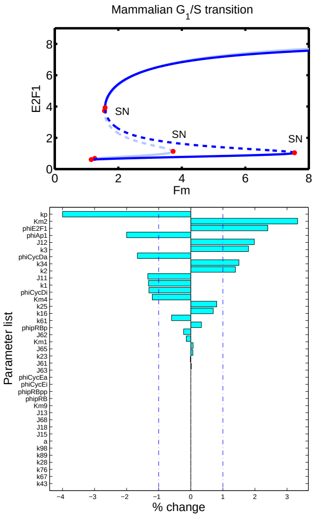
- Map: bifurcation phenotypes $\rightarrow$ parameter sets
- Consider the following 3 modes of geometric transformations of the nominal bifurcation diagram:



Inverse bifurcation: effect of sparsity-promiting penalty



(a) $l_{0.1,10^{-4}}$ regularization



(b) l_2 regularization

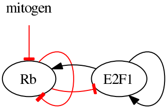
Inverse bifurcation: identified module

Table: Result of hierarchical algorithm with $p = 0.1, \epsilon = 10^{-4}$

Modification Case	Level $j = 1$	Level $j = 2$	Level $j = 3$
Elongating SN_1 nose	$k_p \downarrow$ 14.3%	$k_{34} \uparrow$ 31.7% $K_{m2} \uparrow$ 6.4%	$\phi_{\text{AP-1}} \downarrow$ 20.9% $\phi_{\text{E2F1}} \uparrow$ 7.3%
Moving $SN_{1,2}$ to right	$K_{m4} \uparrow$ 269.3%	$J_{11} \uparrow$ 191.7% $k_p \uparrow$ 17.3%	$k_2 \downarrow$ 39.9% $\phi_{\text{E2F1}} \downarrow$ 11.7% $K_{m2} \downarrow$ 10.3%
Decreasing bistabiliy	$J_{11} \uparrow$ 128.5% $k_p \uparrow$ 33.8%	$k_1 \uparrow$ 169.1% $K_{m2} \downarrow$ 21.7% $J_{12} \downarrow$ 20.1%	$k_2 \downarrow$ 43.7% $\phi_{\text{E2F1}} \downarrow$ 28.3%

Inverse bifurcation: identified module

$$\begin{aligned}
\frac{d}{dt}[\text{pRB}] &= k_1 \frac{[\text{E2F1}]}{K_{m1} + [\text{E2F1}]} \frac{J_{11}}{J_{11} + [\text{pRB}]} \frac{J_{61}}{J_{61} + [\text{pRB}_p]} \\
&- k_{16}[\text{pRB}][\text{CycD}_a] + k_{61}[\text{pRB}_p] - \phi_{\text{pRB}}[\text{pRB}], \\
\frac{d}{dt}[\text{E2F1}] &= k_p + k_2 \frac{a^2 + [\text{E2F1}]^2}{K_{m2}{}^2 + [\text{E2F1}]^2} \frac{J_{12}}{J_{12} + [\text{pRB}]} \frac{J_{62}}{J_{62} + [\text{pRB}_p]} \\
&- \phi_{\text{E2F1}}[\text{E2F1}]
\end{aligned}$$



$$\begin{aligned}
\frac{d}{dt}[\text{CycD}_i] &= -k_{34}[\text{CycD}_i] \frac{[\text{CycD}_a]}{K_{m4} + [\text{CycD}_a]} + \cdots \\
\frac{d}{dt}[\text{CycD}_a] &= k_{34}[\text{CycD}_i] \frac{[\text{CycD}_a]}{K_{m4} + [\text{CycD}_a]} + \cdots
\end{aligned}$$

Possible interprations: G_1/S module

1. Qualitative model - arbitrary interpretations!
2. Modification 1 + 2 : move bistability
   - move SN_1 or SN_1/SN_2 : insensitivity to mitogen
     $\longrightarrow$ E2F1/pRB $\leftrightarrow$ p53/Mdm2 involved in cell-cycle arrest
   - $\downarrow k_p$: decreasing basal transcription of E2F1
     $\longrightarrow$ induction of differentiation markers in squamous cancer cell line (Wong et al. *Oncogene* 2005)
3. Modification 3 : decrease bistability
   - loss of threshold : continuous response to mitogen
     $\longrightarrow$ increase stochasticity of response
   - $\uparrow k_p, \uparrow J11$ etc. : weaken E2F1 self-control
     $\longrightarrow$ yeast *swi4* knock-out ($\approx$ E2F1) shows increased stochasticity in cell cycle (Ubersax JA *Mol Syst Biol* 2006)
4. Compare *Mod. 2, level 2* with *Mod. 3, level 1*
   $\longrightarrow$ dynamic properties of complex systems unintuitive!
5. Only one step in model-experiment loop!
6. Apply to GATA evolution: based on realistic data, only relative changes in evolutionary duplication scenarios!

Possible interprations: G_1/S module

1. Qualitative model - arbitrary interpretations!
2. Modification 1 + 2 : move bistability
   - move SN_1 or SN_1/SN_2 : insensitivity to mitogen
     $\rightarrow$ E2F1/pRB $\leftrightarrow$ p53/Mdm2 involved in cell-cycle arrest
   - $\downarrow k_p$: decreasing basal transcription of E2F1
     $\rightarrow$ induction of differentiation markers in squamous cancer cell line (Wong et al. *Oncogene* 2005)
3. Modification 3 : decrease bistability
   - loss of threshold : continuous response to mitogen
     $\rightarrow$ increase stochasticity of response
   - $\uparrow k_p, \uparrow J11$ etc. : weaken E2F1 self-control
     $\rightarrow$ yeast *swi4* knock-out ($\approx$ E2F1) shows increased stochasticity in cell cycle (Ubersax JA *Mol Syst Biol* 2006)
4. Compare *Mod. 2, level 2* with *Mod. 3, level 1*
   $\rightarrow$ dynamic properties of complex systems unintuitive!
5. Only one step in model-experiment loop!
6. Apply to GATA evolution: based on realistic data, only relative changes in evolutionary duplication scenarios!

Possible interprations: G_1/S module

1. Qualitative model - arbitrary interpretations!
2. Modification 1 + 2 : move bistability
   - move SN_1 or SN_1/SN_2 : insensitivity to mitogen
     $\longrightarrow$ E2F1/pRB $\leftrightarrow$ p53/Mdm2 involved in cell-cycle arrest
   - $\downarrow k_p$: decreasing basal transcription of E2F1
     $\longrightarrow$ induction of differentiation markers in squamous cancer cell line (Wong et al. *Oncogene* 2005)
3. Modification 3 : decrease bistability
   - loss of threshold : continuous response to mitogen
     $\longrightarrow$ increase stochasticity of response
   - $\uparrow k_p, \uparrow J11$ etc. : weaken E2F1 self-control
     $\longrightarrow$ yeast *swi4* knock-out ($\approx$ E2F1) shows increased stochasticity in cell cycle (Ubersax JA *Mol Syst Biol* 2006)
4. Compare *Mod. 2, level 2* with *Mod. 3, level 1*
   $\longrightarrow$ dynamic properties of complex systems unintuitive!
5. Only one step in model-experiment loop!
6. Apply to GATA evolution: based on realistic data, only relative changes in evolutionary duplication scenarios!

Possible interprations: G_1/S module

1. Qualitative model - arbitrary interpretations!
2. Modification 1 + 2 : move bistability
   - move SN_1 or SN_1/SN_2 : insensitivity to mitogen
     $\longrightarrow$ E2F1/pRB $\leftrightarrow$ p53/Mdm2 involved in cell-cycle arrest
   - $\downarrow k_p$: decreasing basal transcription of E2F1
     $\longrightarrow$ induction of differentiation markers in squamous cancer cell line (Wong et al. *Oncogene* 2005)
3. Modification 3 : decrease bistability
   - loss of threshold : continuous response to mitogen
     $\longrightarrow$ increase stochasticity of response
   - $\uparrow k_p$, $\uparrow J11$ etc. : weaken E2F1 self-control
     $\longrightarrow$ yeast *swi4* knock-out ($\approx$ E2F1) shows increased stochasticity in cell cycle (Ubersax JA *Mol Syst Biol* 2006)
4. Compare *Mod. 2, level 2* with *Mod. 3, level 1*
   $\longrightarrow$ dynamic properties of complex systems unintuitive!
5. Only one step in model-experiment loop!
6. Apply to GATA evolution: based on realistic data, only relative changes in evolutionary duplication scenarios!

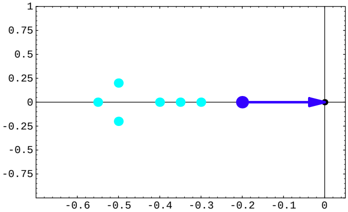
Possible interprations: G_1/S module

1. Qualitative model - arbitrary interpretations!
2. Modification 1 + 2 : move bistability
   - move SN_1 or SN_1/SN_2 : insensitivity to mitogen
     $\rightarrow$ E2F1/pRB $\leftrightarrow$ p53/Mdm2 involved in cell-cycle arrest
   - $\downarrow k_p$: decreasing basal transcription of E2F1
     $\rightarrow$ induction of differentiation markers in squamous cancer cell line (Wong et al. *Oncogene* 2005)
3. Modification 3 : decrease bistability
   - loss of threshold : continuous response to mitogen
     $\rightarrow$ increase stochasticity of response
   - $\uparrow k_p, \uparrow J11$ etc. : weaken E2F1 self-control
     $\rightarrow$ yeast *swi4* knock-out ($\approx$ E2F1) shows increased stochasticity in cell cycle (Ubersax JA *Mol Syst Biol* 2006)
4. Compare *Mod. 2, level 2* with *Mod. 3, level 1*
   $\rightarrow$ dynamic properties of complex systems unintuitive!
5. Only one step in model-experiment loop!
6. Apply to GATA evolution: based on realistic data, only relative changes in evolutionary duplication scenarios!

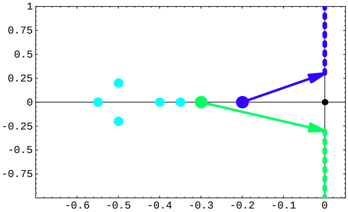
Inverse eigenvalue problems: ODE setting

Definition (IEP for Saddle-Node and Hopf bifurcations)

Denote scalars $\lambda_{SN} = \{0\}$ or $\lambda_H = \{\pm\omega i\}$. With equilibrium condition $f(x,\alpha) = 0$, determine parameter values α such that $\sigma(\frac{df}{dx}) \supset \lambda_{SN,H}$.



(c) IEA for Saddle-Node



(d) IEA for Hopf

Inverse eigenvalue problems: ODE setting

- **Hybrid** solution algorithm:
  - Lift-and-Project (LP)
  - Quasi-Newton (QN)
- Least square formulations with regularization:

$$\text{LSIEP1} \quad : \quad J(\alpha) = \sum_i |\lambda_i(\alpha) - \lambda_i^d|^2 + \mu\|\boldsymbol{\alpha} - \boldsymbol{\alpha}^*\|_{l_p},$$

$$\text{LSIEP2} \quad : \quad J(\alpha) = \|A(\alpha) - Q\tilde{T}(\{\lambda_i^d\})Q^H\|_{\mathcal{F}}^2 + \mu\|\boldsymbol{\alpha} - \boldsymbol{\alpha}^*\|_{l_p}.$$

Inverse eigenvalue problems: ODE setting

- **Hybrid** solution algorithm:
  - Lift-and-Project (LP)
  - Quasi-Newton (QN)
- Least square formulations with regularization:

$$\text{LSIEP1} \quad : \quad J(\alpha) = \sum_i |\lambda_i(\alpha) - \lambda_i^d|^2 + \mu\|\boldsymbol{\alpha} - \boldsymbol{\alpha}^*\|_{l_p},$$

$$\text{LSIEP2} \quad : \quad J(\alpha) = \|A(\alpha) - Q\tilde{T}(\{\lambda_i^d\})Q^H\|_{\mathcal{F}}^2 + \mu\|\boldsymbol{\alpha} - \boldsymbol{\alpha}^*\|_{l_p}.$$

Inverse eigenvalue problems: ODE setting

- **Hybrid** solution algorithm:
  - Lift-and-Project (LP)
  - Quasi-Newton (QN)
- Least square formulations with regularization:

$$\text{LSIEP1} \quad : \quad J(\alpha) = \sum_i |\lambda_i(\alpha) - \lambda_i^d|^2 + \mu\|\boldsymbol{\alpha} - \boldsymbol{\alpha}^*\|_{l_p},$$

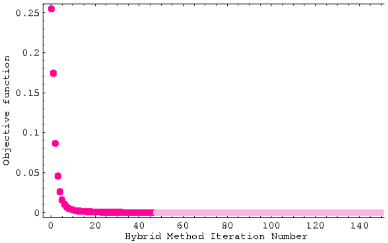
$$\text{LSIEP2} \quad : \quad J(\alpha) = \|A(\alpha) - Q\tilde{T}(\{\lambda_i^d\})Q^H\|_{\mathcal{F}}^2 + \mu\|\boldsymbol{\alpha} - \boldsymbol{\alpha}^*\|_{l_p}.$$

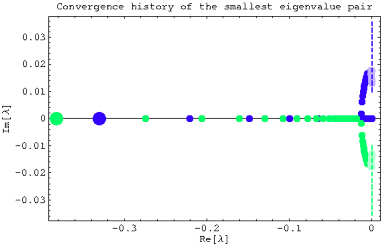
Inverse eigenvalue problems: ODE setting

- **Hybrid** solution algorithm:
  - Lift-and-Project (LP)
  - Quasi-Newton (QN)
- Least square formulations with regularization:

$$\text{LSIEP1} \quad : \quad J(\alpha) = \sum_i |\lambda_i(\alpha) - \lambda_i^d|^2 + \mu \|\boldsymbol{\alpha} - \boldsymbol{\alpha}^*\|_{l_p},$$

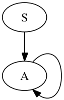
$$\text{LSIEP2} \quad : \quad J(\alpha) = \|A(\alpha) - Q\tilde{T}(\{\lambda_i^d\})Q^H\|_{\mathcal{F}}^2 + \mu \|\boldsymbol{\alpha} - \boldsymbol{\alpha}^*\|_{l_p}.$$

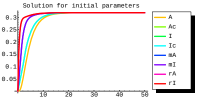


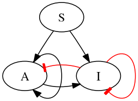


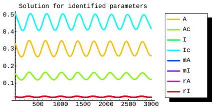
Emergence of an oscillator from bistable switch

- Time-series: the initial and identified systems





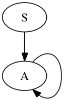


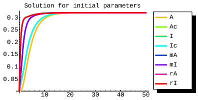


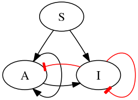
- Identified reactions:

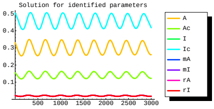
Emergence of an oscillator from bistable switch

- Time-series: the initial and identified systems







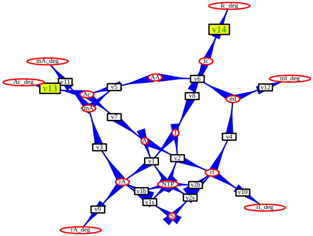


- Identified reactions:

System tG : identified reactions

tG‘v13[t] == tG‘Ac[t] tG‘D_Ac (1→0.15)

tG‘v14[t] == tG‘Ic[t] tG‘D_Ic (1→0.046)



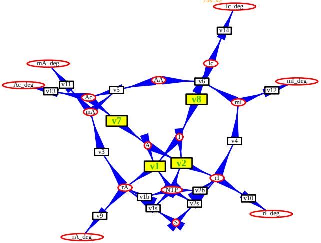
System tG : identified reactions

tG‘v7[t] == tG‘kin_A tG‘Ac[t] − tG‘A[t] tG‘kout_A (1→0.9)

$$tG‘v1[t] == \frac{tG‘Va\ tG‘A[t]^2}{\left(tG‘Ka\_A + tG‘A[t] + \frac{tG‘Ka\_A\ tG‘I[t]}{tG‘Ki\_A\ (1\to 0.14)}\right)^2}$$

$$tG‘v2[t] == \frac{tG‘Vi\ tG‘A[t]^2}{\left(tG‘Ka\_I + tG‘A[t] + \frac{tG‘Ka\_I\ tG‘I[t]}{tG‘Ki\_I\ (1\to 0.25)}\right)^2}$$

tG‘v8[t] == tG‘kin_I tG‘Ic[t] − tG‘I[t] tG‘kout_I (1→0.42)



Conclusions and Outlook

- Inverse dynamical analysis
  - Sparsity-cosntraint
    $\longrightarrow$ identify governing modules and parameters
  - Highly useful in model-experiment loop
    1. find crucial parameters for observed behaviour
    2. propose new experiments
- Evolution of the *bifurcation phenotype*
  1. duplication of auto-activator causes hypersensitivity stress
  2. analyze all possible relieving mutations by Inverse Dynamical Analysis
  3. reconstruct evolutionary pathways towards complex transcription factor networks

Conclusions and Outlook

- Inverse dynamical analysis
  - Sparsity-cosntraint
    $\longrightarrow$ identify governing modules and parameters
  - Highly useful in model-experiment loop
    1. find crucial parameters for observed behaviour
    2. propose new experiments
- Evolution of the *bifurcation phenotype*
  1. duplication of auto-activator causes hypersensitivity stress
  2. analyze all possible relieving mutations by Inverse Dynamical Analysis
  3. reconstruct evolutionary pathways towards complex transcription factor networks

Thanks: **BIRD07**

Lukas Endler, Stefan Müller
Christoph Flamm, Stefanie Müller
Peter Schuster, Heinz Engl
Douglas Murray

Funded by:

WWTF

Wiener Wissenschafts-, Forschungs- und Technologiefonds

Project Nr. MA05