1st FEBS Advanced Lecture Course
Systems Biology: From Molecules & Modeling to Cells

Mathematical Modeling of Stress Response in Yeast

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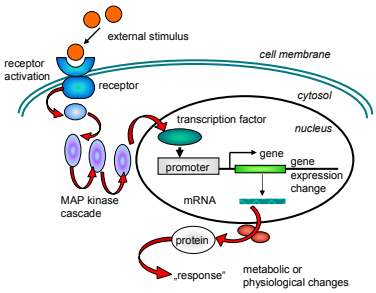
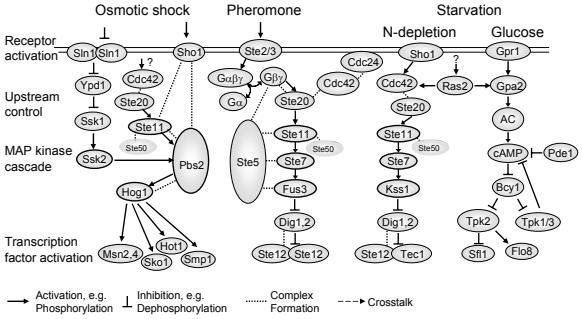
Signaling paradigm



Typical Signals:
- hormones, pheromones
- heat, cold
- osmotic pressure
- concentration changes (K, Ca, cAMP,..)

Signaling Pathways in Baker's Yeast



Aim

All models are wrong, some are useful.
George B.P. Box

Signaling pathways are composed of different modules:
- Receptors
- G proteins
- Ras protein
- MAP kinase cascades
- ....

These modules are ubiquitous.

They contribute in different ways to signal transmission, signal processing, and regulation.

Overview

Comparison of signaling and metabolic pathways
- On the level of individual reaction kinetics
- On the network level

Modeling of signaling pathway modules
- Receptor kinetics
- G protein cycle
- Phospho-relay system
- MAP kinase cascade

Model of the pheromone pathway
- Integration of moduls
- Effect of regulatory interactions

Characteristics of Signaling Pathways

- Complex units
- Interaction of different types of components (proteins, ions, small metabolites,...)
- Involvement of various compartments and places in the cell
- Regulatory interactions: feedback and feedforward loops

- Uncomplete knowledge about components and their interactions
- Interpretation of data is dependent on background knowledge and context

- The action of the signal changes the state of the cell.
  ➡ Difficulties in determination of system limits

Common properties of metabolic and signaling pathways

Cellular network has a high degree of connectivity.

The processes are reactions, molecular interactions.
- binding
- intramolecular transformations
- release

Differences in modeling of different parts are due to appropriate approximations.

Network characteristics

Metabolism

All reactions are catalyzed by enzymes.

→The network is determined by the existing enzymes (which not necessarily interact).

Metabolites need not to be there initially.

Signaling

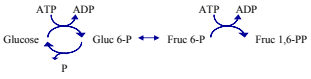
Reactions can be
- catalyzed by enzymes
- autocatalytic.

→The network is given by the existing protein and their interactions.

The network forms dynamically.

Network characteristics

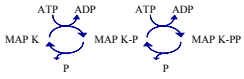
Metabolism



Important feature:
Flux through the pathway,
(final) transformation of metabolites

Phosphorylation ⟷ energy transfer

Signaling



State changes:
change in phosporylation states
Coding of information

But: Conservation
(MAPK + MAPK-P + MAPK-PP)
in the considered time window

Concentrations

Signaling

Proteins low

~ 100-300 nmol/L
(~ 10^3-10^4 molecules per cell)

(catalysts and substrates)

ATP ~ 2 mmol/L

Metabolism

Enzymes low

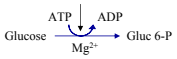
Metabolites higher

Metabolite	Concentration (mmol/L)
Glucose-6-phosphate	0.93 ± 0.01
Fructose-6-phosphate	0.17 ± 0.005
Fructose-1,6-bisphosphate	0.11 ± 0.005
Glyceraldehyde-3-phosphate	0.063 ± 0.001
3-Phosphoglycerate	1.33 ± 0.02
Phosphoenol pyruvate	0.053 ± 0.004
Pyruvate	0.79 ± 0.02
Adenine nucleotides (cytoplasm)	
ATP	2.1 ± 0.1 mmol/L
ADP	0.47 ± 0.05 mmol/L
AMP	0.11 ± 0.03 mmol/L

Theobald U et al., 1996, Biotechnol Bioeng 55, 305

Rate equations: Modelers choice

Metabolism



Signaling

Mass action kinetics

Typical choice:
Michaelis-Menten-Kinetics

$E+S \underset{slow}{\overset{fast}{\rightleftharpoons}} ES \xrightarrow{slow} E+P$ $\quad v = \frac{V_{max} S}{K_M + S}, \quad V_{max} = k_2 \cdot E_{tot}$

Requirement: E << S

Hexokinase (Fromm and Zewe, 1962):

$$r_{Glc} = \frac{r_{Glc}^{max}}{1 + \frac{K_{A,ATP,2}}{C_{A,ATP}}\left[1 + \frac{C_{A,ADP}}{K_{A,ADP,2}}\right] + \frac{K_{Glc,2}}{C_{Glc}} + \frac{K_{A,ATP,2} K_{Glc,2}}{C_{A,ATP} C_{Glc}}\left[1 + \frac{C_{A,ADP}}{K_{A,ADP,2}}\right]}$$

Catalyst and Substrate have about the same concentration (E≅S)

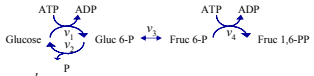
Binding slow compared to intramolecular rearrangements.

First order kinetics

$v = k \cdot E \cdot S, \quad k = k(ATP)$

Balance equations, Stoichiometry

Metabolism



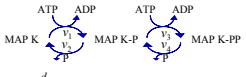
$$\frac{d}{dt} Gluc6-P = v_1 - v_2 - v_3$$

$$N = \begin{pmatrix} -1 & 1 & 0 & 0 \\ 1 & -1 & -1 & 0 \\ 0 & 0 & 1 & -1 \\ 0 & 0 & 0 & 1 \\ -1 & 0 & 0 & -1 \\ 1 & 0 & 0 & 1 \end{pmatrix} \begin{matrix} Glucose \\ Gluc6-P \\ Fruc6-P \\ Fruc1,6-PP \\ ATP \\ ADP \end{matrix}$$

Fewer conservation relations
Fewer independent fluxes

Flow of matter

Signaling



$$\frac{d}{dt} MAPK-P = v_1 - v_2 - v_3 + v_4$$

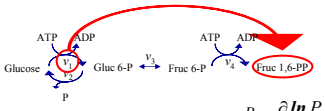
$$N = \begin{pmatrix} -1 & 1 & 0 & 0 \\ 1 & -1 & -1 & 1 \\ 0 & 0 & 1 & -1 \\ -1 & 0 & -1 & 0 \\ 1 & 0 & 1 & 0 \end{pmatrix} \begin{matrix} MAPK \\ MAPK-P \\ MAPK-PP \\ ATP \\ ADP \end{matrix}$$

Many conservation relations
Many independent fluxes
→Modules

Flow of information

Application of Control Analysis

Metabolism



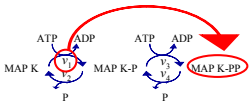
$$C^{P}_{v_1} = \frac{\partial \ln P}{\partial \ln v_1}$$

Steady state
Small perturbations

Steady state as useful approximation

As used in MCA or FBA

Signaling



Steady state as useful approximation
Interesting:
temporal changes during signalling

Spatial effects

Signaling

„well stirred" ???

Low number of molecules,
Highly organised complexes,
Often membrane-bound.

Spatial effects should be considered.
(problem with ODEs)
At least as „compartmentalisation"

Metabolism

„well stirred"

Molecules are considered to meet with probability
according to their concentration
(mass action).

Spatial effects usually neglected.

Overview

Comparison of signaling and metabolic pathways
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- G protein cycle
- Phospho relay system
- MAP kinase cascade

Model of the pheromone pathway
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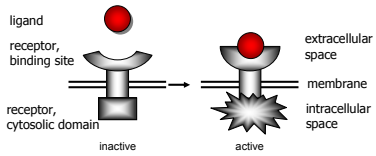
Receptor kinetics



- membrane bound
- receive and transmit signal
- conformation change
- active or inactive forms

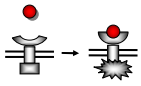
Simplest approach:

$$L + R \rightleftharpoons LR$$

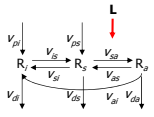
L – ligand
R – receptor
LR – ligand-receptor-complex

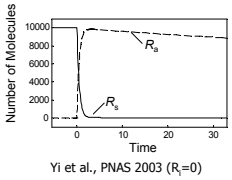
$$K_D = \frac{L \cdot R}{LR}$$

typical values:
$K_D = 10^{-12}$ M ....10^{-6} M



Receptor kinetics





Yi et al., PNAS 2003 (R_i=0)

Differential equations

$$\frac{d}{dt}R_i = v_{pi} - v_{di} - v_{is} + v_{si} + v_{ai}$$

$$\frac{d}{dt}R_s = v_{ps} - v_{ds} + v_{is} - v_{si} - v_{sa} + v_{as}$$

$$\frac{d}{dt}R_a = -v_{da} + v_{sa} - v_{as} - v_{ai}$$

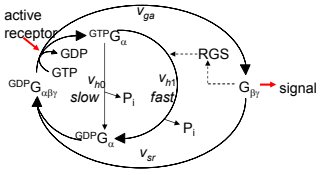
Rate expressions ??

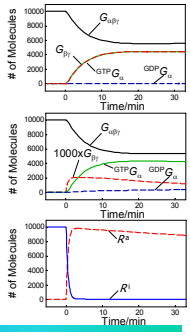
$$v_{xy} = k_{xy} \cdot R_x$$ Mass action

$$v_{sa} = k_{sa} \cdot R_s \cdot L$$

$$v_{sa} = k_{sa} \cdot R_s \cdot \frac{K_b \cdot L^n}{1 + K_b \cdot L^n}$$ Hill kinetics

G protein cycle





Differential equations

$$\frac{d}{dt}G_{\alpha\beta\gamma} = -v_{ga} + v_{sr}$$

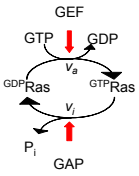
$$\frac{d}{dt}G_{\alpha}GTP = v_{ga} - v_{h0} - v_{h1}$$

Conservation relations

$$G_{total} = G_{\alpha\beta\gamma} + G_{\beta\gamma}$$

$$G_{total} = G_{\alpha\beta\gamma} + G_{\alpha}GTP + G_{\alpha}GDP$$

Small monomeric G-Protein



Small monomeric G-proteins

Belong to 5 families: Ras, Rho, Rab, Ran, and Arf

Ras protein cycles between active and inactive state

GEF – guanine nucleotide exchange factor

GAP – GTPase activating protein

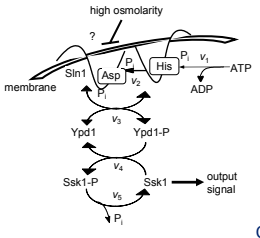
Differential equations

$$\frac{d}{dt}{}^{GDP}Ras = -v_a + v_i$$

Conservation relations

$$Ras_{total} = {}^{GDP}Ras + {}^{GTP}Ras$$

Phospho relay system



$$\frac{d}{dt}Sln1 = -k_1 \cdot Sln1 + k_3 \cdot Sln1A - P \cdot Ypd1$$

$$\frac{d}{dt}Sln1H - P = k_1 \cdot Sln1 - k_2 Sln1H - P$$

$$\frac{d}{dt}Sln1A - P = k_2 \cdot Sln1H - P - k_3 \cdot Sln1A - P \cdot Ypd1$$

$$\frac{d}{dt}Ypd1 = k_4 \cdot Ypd1 - P \cdot Ssk1 - k_3 \cdot Sln1A - P \cdot Ypd1$$

$$\frac{d}{dt}Ypd1 - P = -k_4 \cdot Ypd1 - P \cdot Ssk1 + k_3 \cdot Sln1A - P \cdot Ypd1$$

$$\frac{d}{dt}Ssk1 = k_5 \cdot Ssk1 - P - k_4 \cdot Ypd1 - P \cdot Ssk1$$

$$\frac{d}{dt}Ssk1 - P = -k_5 \cdot Ssk1 - P + k_4 \cdot Ypd1 - P \cdot Ssk1$$

Conservation relations

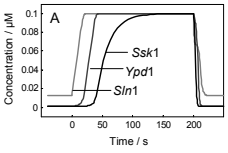
$$Sln1_{total} = Sln1 + Sln1H - P + Sln1A - P$$

$$Ypd1_{total} = Ypd1 + Ypd1 - P$$

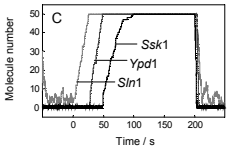
$$Ssk1_{total} = Ssk1 + Ssk1 - P$$

Phospho relay system

Time courses

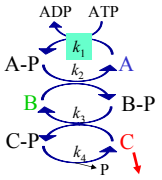


Deterministic simulation



Stochastic simulation

Phospho relay system



$$\frac{d}{dt}A = -k_1 \cdot A \cdot ATP + k_2 \cdot AP \cdot B$$

$$\frac{d}{dt}B = -k_2 \cdot B \cdot AP + k_3 \cdot BP \cdot C$$

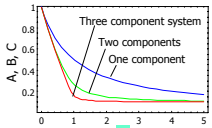
$$\frac{d}{dt}C = -k_3 \cdot C \cdot BP + k_4 \cdot CP$$

$$A + AP = A_{total}$$

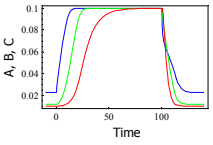
$$B + BP = B_{total}$$

$$C + CP = C_{total}$$

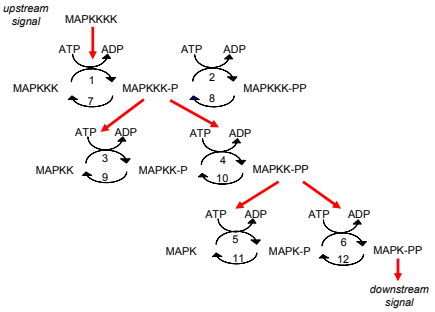
Dependence of steady state values On stimulus strength



Temporal behavior upon stress and relieve of stress

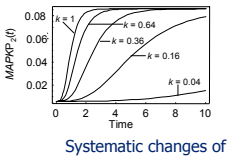


MAP kinase cascades



MAP kinase cascades

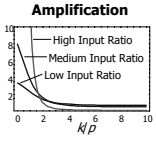
Time courses



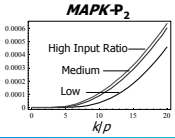
- Sigmoidal input/output dependence
- Signal amplification
- ⋮
- *but*: parameter dependence

Systematic changes of kinase or phosphatase activities

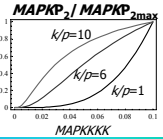
Amplification



***MAPK-P*$_2$**



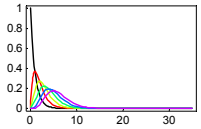
***MAPKP*$_2$ / *MAPKP*$_{2max}$**



Why so complicated?

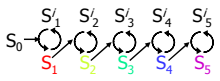
Metabolic pathway

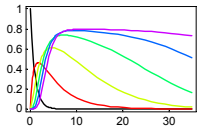
$S_0 \rightarrow S_1 \rightarrow S_2 \rightarrow S_3 \rightarrow S_4 \rightarrow S_5$



Mass action kinetics,
all rate constants equal to 1.

Signaling cascade



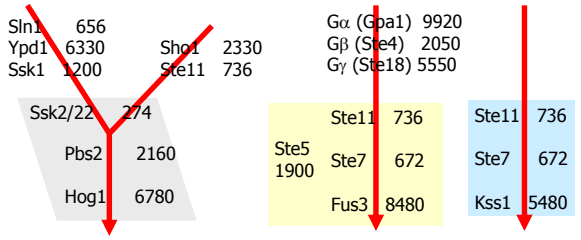


Mass action kinetics,
$k = 1, l\ p = 0.2$.

Robust against parameter changes

Signal Amplification in Numbers

http://yeastgfp.ucsf.edu/



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Model of the pheromone pathway
- Integration of moduls
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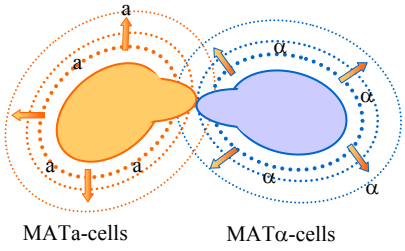
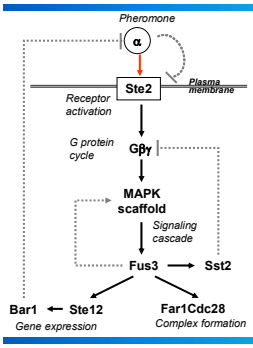
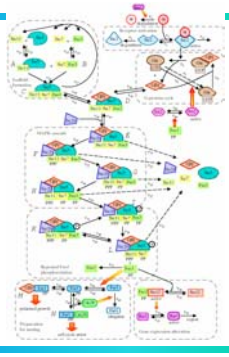
Putting all together: the Pheromone pathway

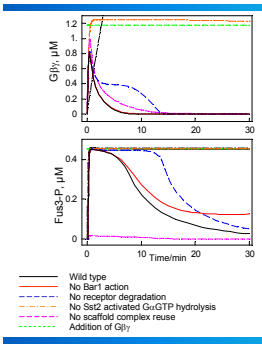


Pheromone pathway: structural parts



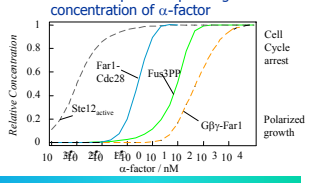


Pheromone pathway: time courses

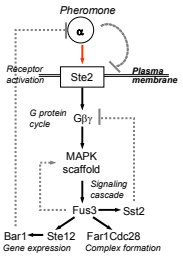


In comprehensive model:
regulatory feedback loops are considered
mutant phenotypes can be investigated

Graded response depending on concentration of α-factor



Comparison of modeling approaches

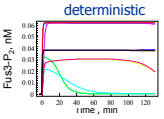


Petri net

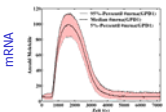
→Signal flow

Differential equations

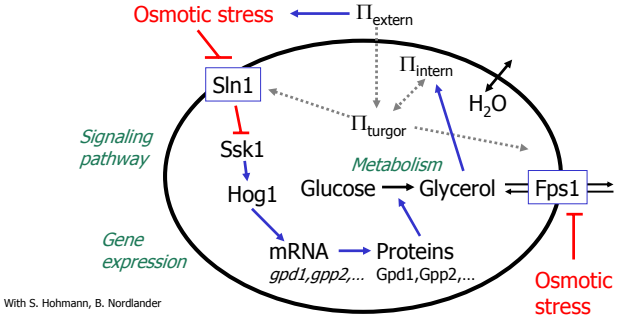
deterministic



stochastic

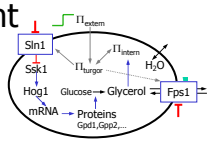


A Model for Osmostress Response

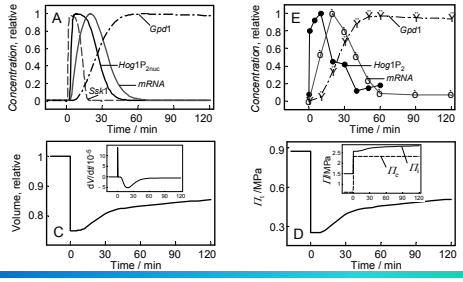


With S. Hohmann, B. Nordlander

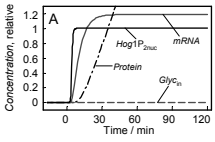
The Standard Experiment

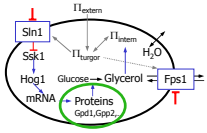


Wild Type Cells, shock with 0.5 M NaCl

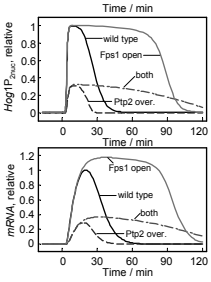


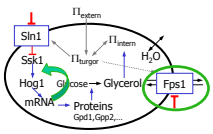
Test case: gpd1Δ gpd2Δ mutant



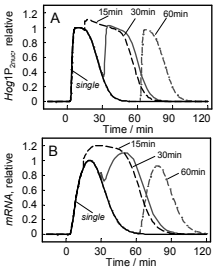


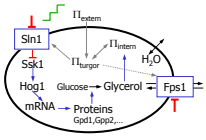
Test cases: Fps1 open / Ptp2 up





Test case: Double shock





Cells are competent to respond to a second shock.

Conclusions

Models for Metabolism and Signaling can use the same design principles.

Metabolism and Signaling may take place in
- various areas of the cells
- various regions of the concentration space
- various time scales

Signaling models have to account for the hierarchy in the system

Regulatory couplings (feedback) distribute the control in both cases.

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