Rule-Based Kinetic Modeling of Signal Transduction Networks

Part I. Motivation

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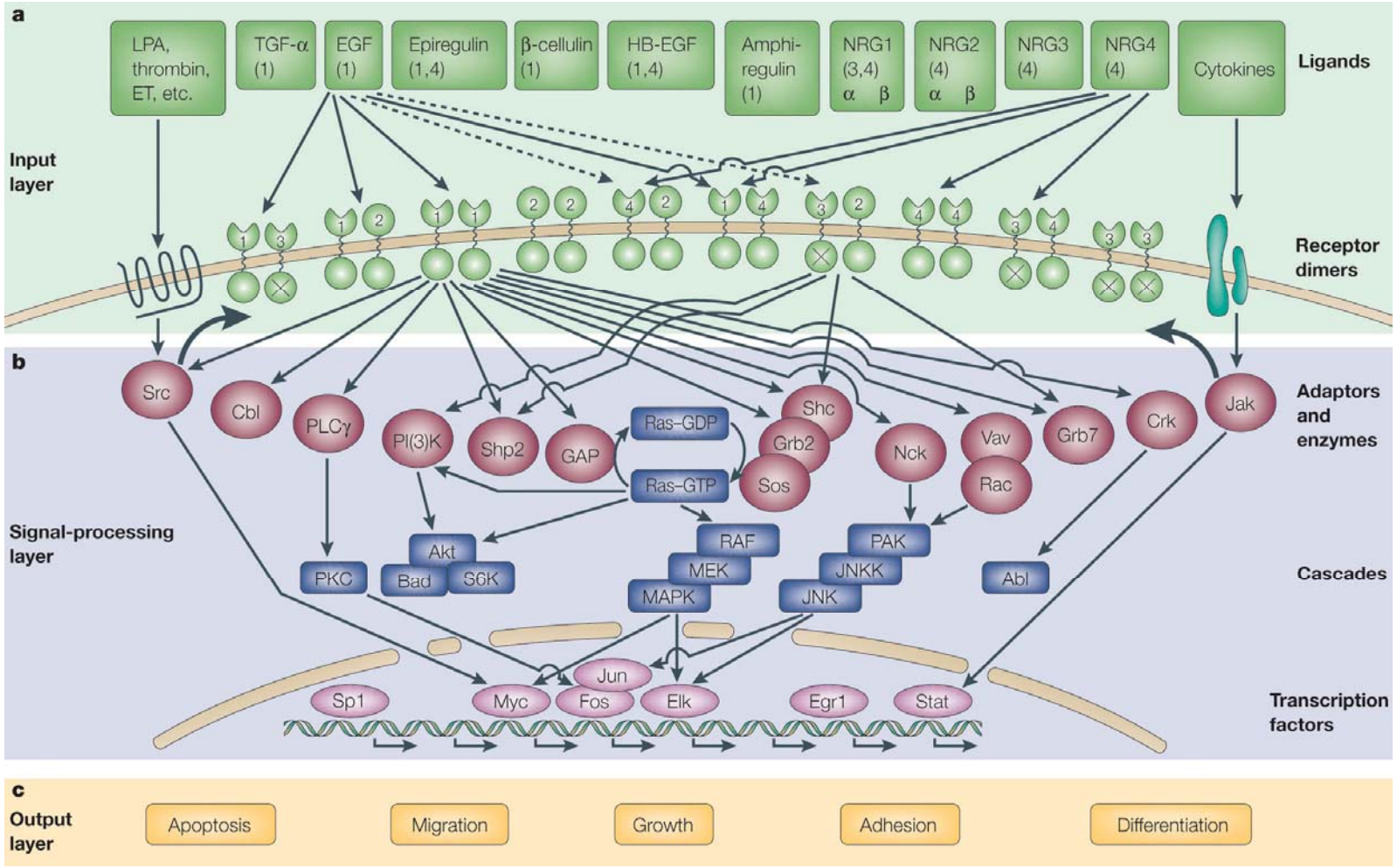
NIH
DOE
LANL-LDRD

Example 1: Early events in signaling by Epidermal Growth factor Receptor

Large networks of proteins and other molecules are involved in signaling



Yarden & Sliwkowski, *Nature Rev. Mol. Cell Biol.* **02**: 127-137 (2001).

Phenomenological vs. Mechanistic Modeling

- Type of model depends on the questions one wants to ask (and answer).
- *Phenomenological models* are good for establishing correlations among the measured variables.
- *Mechanistic models* attempt to put known information into a model that can describe data and make predictions about how manipulating the components affects the outcome.

Multiplicity of sites and binding partners gives rise to combinatorial complexity

Epidermal growth factor receptor (EGFR)

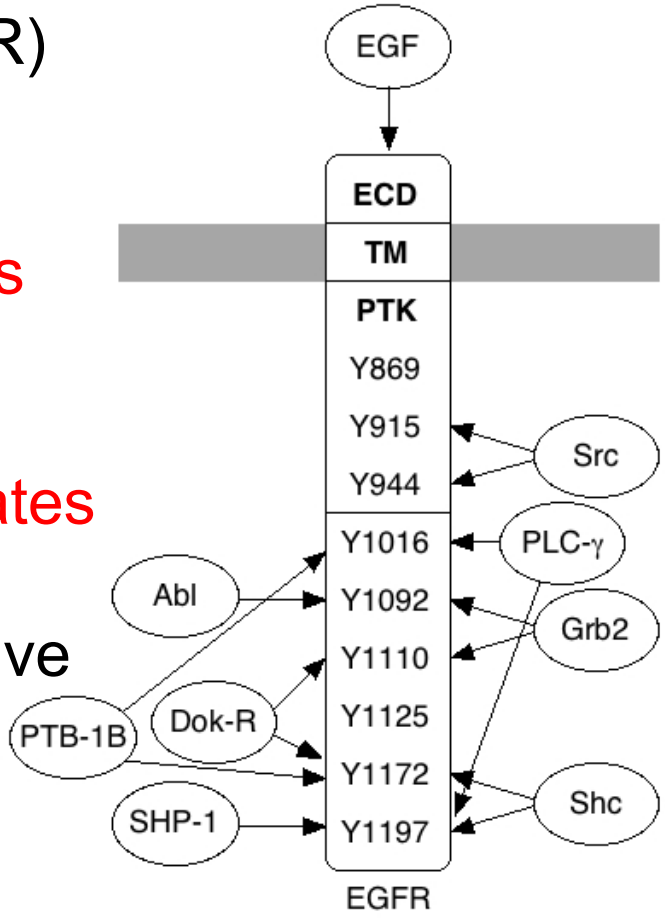
9 sites $\Rightarrow$ 2^9=512 phosphorylation states

Each site has ≥ 1 binding partner

$\Rightarrow$ more than 3^9=19,683 total states

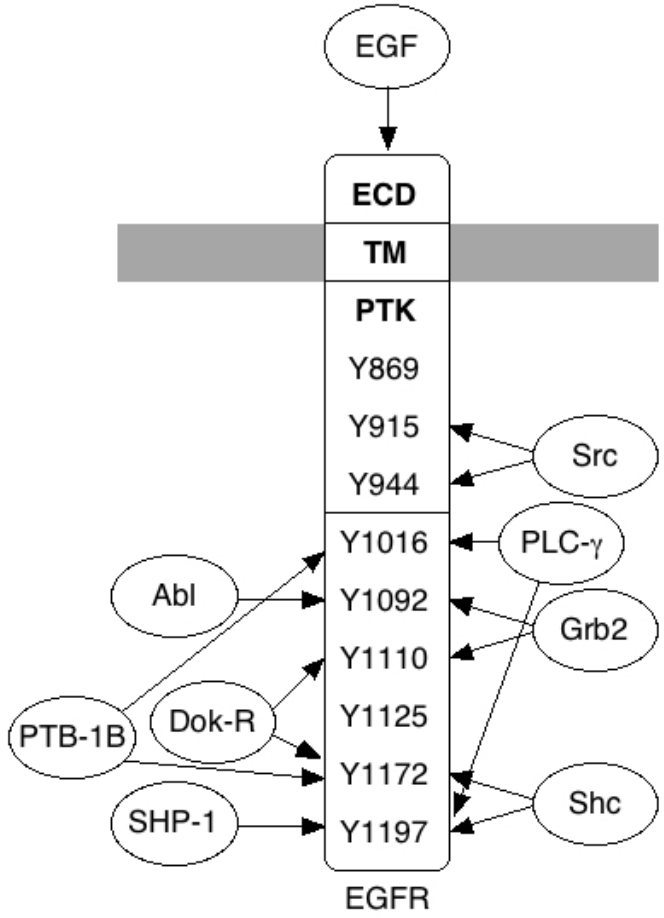
EGFR must form *dimers* to become active

$\Rightarrow$ more than 1.9×10^8 states



Multiplicity of sites and binding partners gives rise to combinatorial complexity

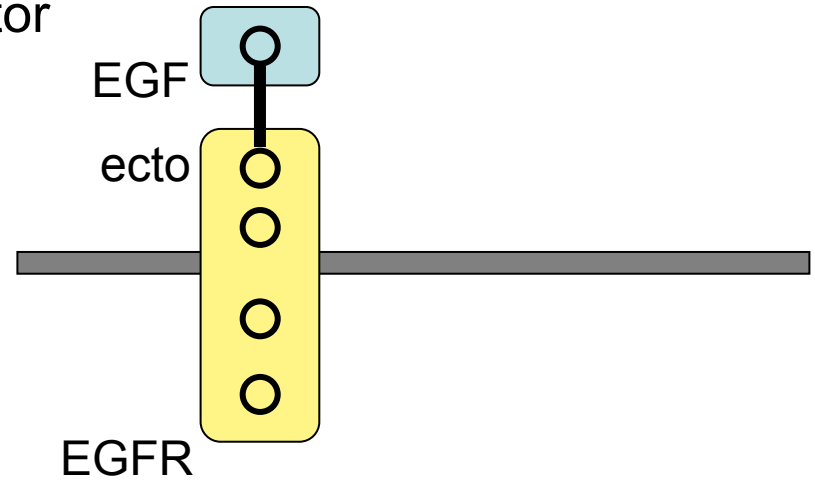
…but the number of interactions is relatively small.



Early events in EGFR signaling

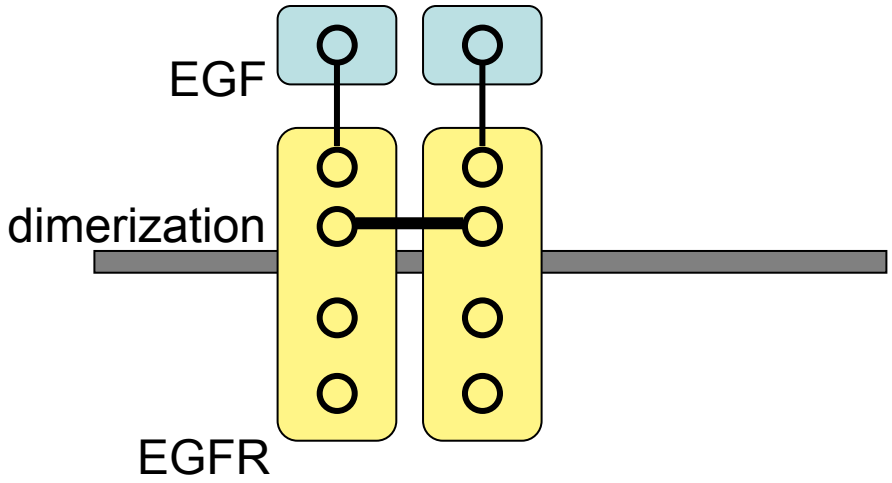
EGF = epidermal growth factor
EGFR = epidermal growth factor receptor

1. EGF binds EGFR



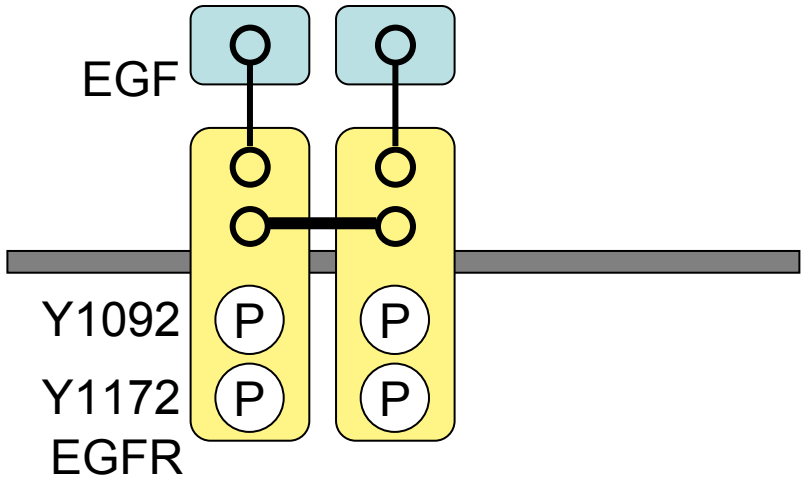
Early events in EGFR signaling

1. EGF binds EGFR
2. **EGFR dimerizes**



Early events in EGFR signaling

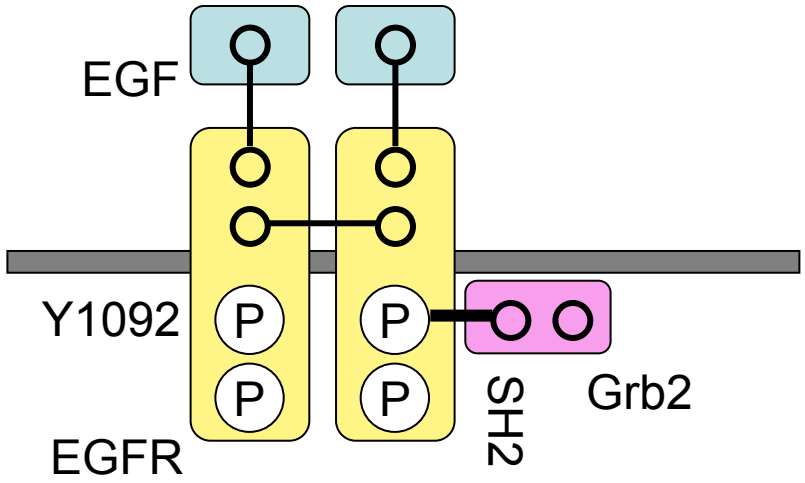
1. EGF binds EGFR
2. EGFR dimerizes
3. **EGFR transphosphorylates itself**



Early events in EGFR signaling

Grb2 pathway

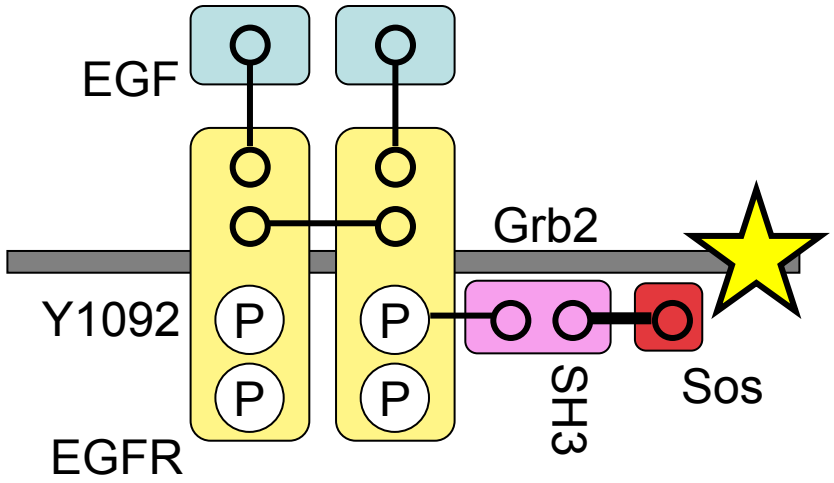
1. EGF binds EGFR
2. EGFR dimerizes
3. EGFR transphosphorylates itself
4. **Grb2 binds phospho-EGFR**



Early events in EGFR signaling

Grb2 pathway

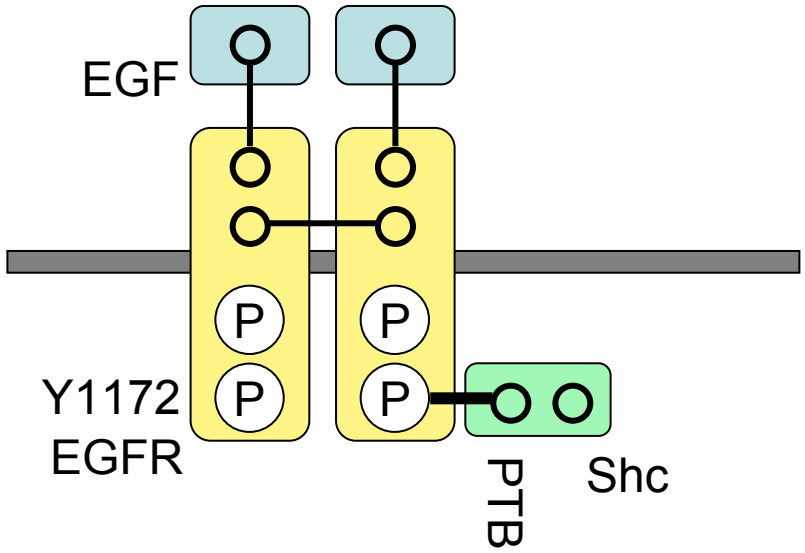
1. EGF binds EGFR
2. EGFR dimerizes
3. EGFR transphosphorylates itself
4. Grb2 binds phospho-EGFR
5. **Sos binds Grb2 (Activation Path 1)**



Early events in EGFR signaling

Shc pathway

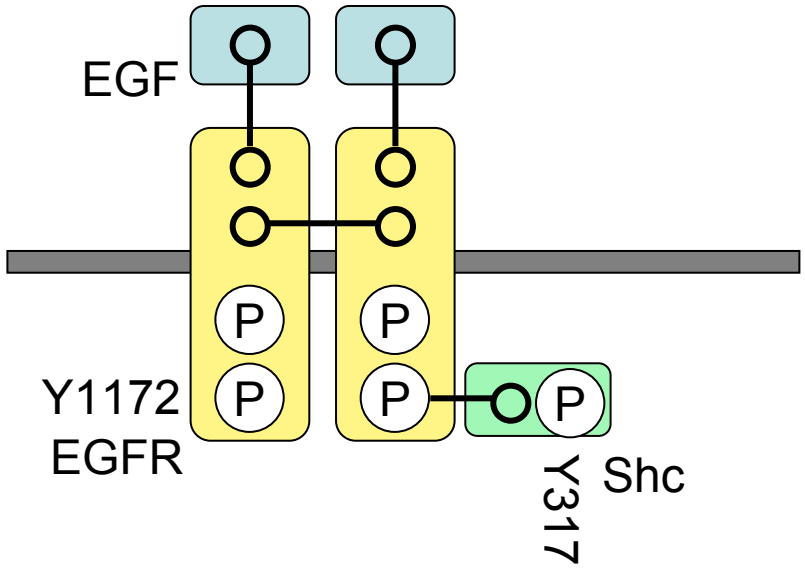
1. EGF binds EGFR
2. EGFR dimerizes
3. EGFR transphosphorylates itself
4. **Shc binds phospho-EGFR**



Early events in EGFR signaling

Shc pathway

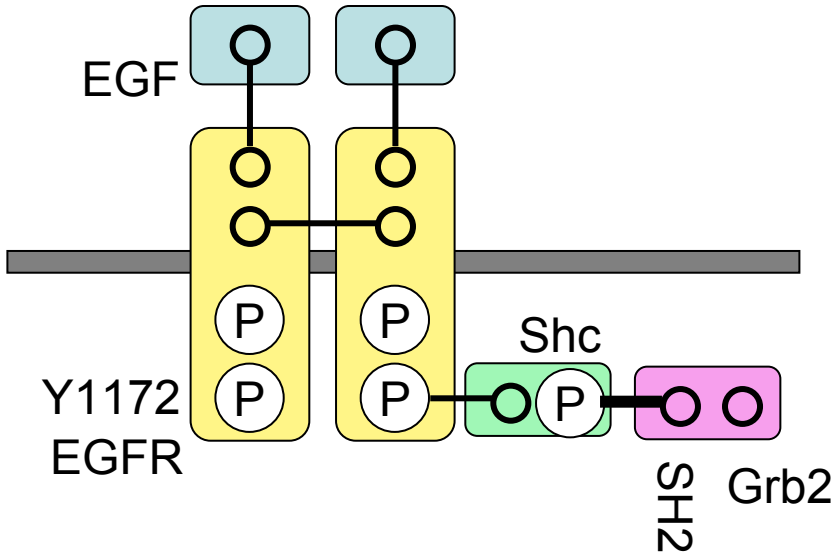
1. EGF binds EGFR
2. EGFR dimerizes
3. EGFR transphosphorylates itself
4. Shc binds phospho-EGFR
5. **EGFR transphosphorylates Shc**



Early events in EGFR signaling

Shc pathway

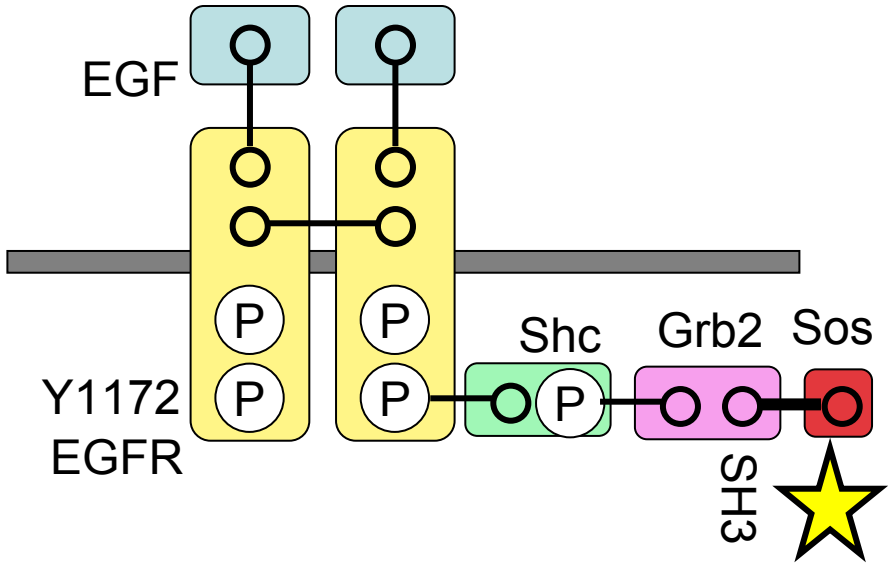
1. EGF binds EGFR
2. EGFR dimerizes
3. EGFR transphosphorylates itself
4. Shc binds phospho-EGFR
5. EGFR transphosphorylates Shc
6. **Grb2 binds phospho-Shc**



Early events in EGFR signaling

Shc pathway

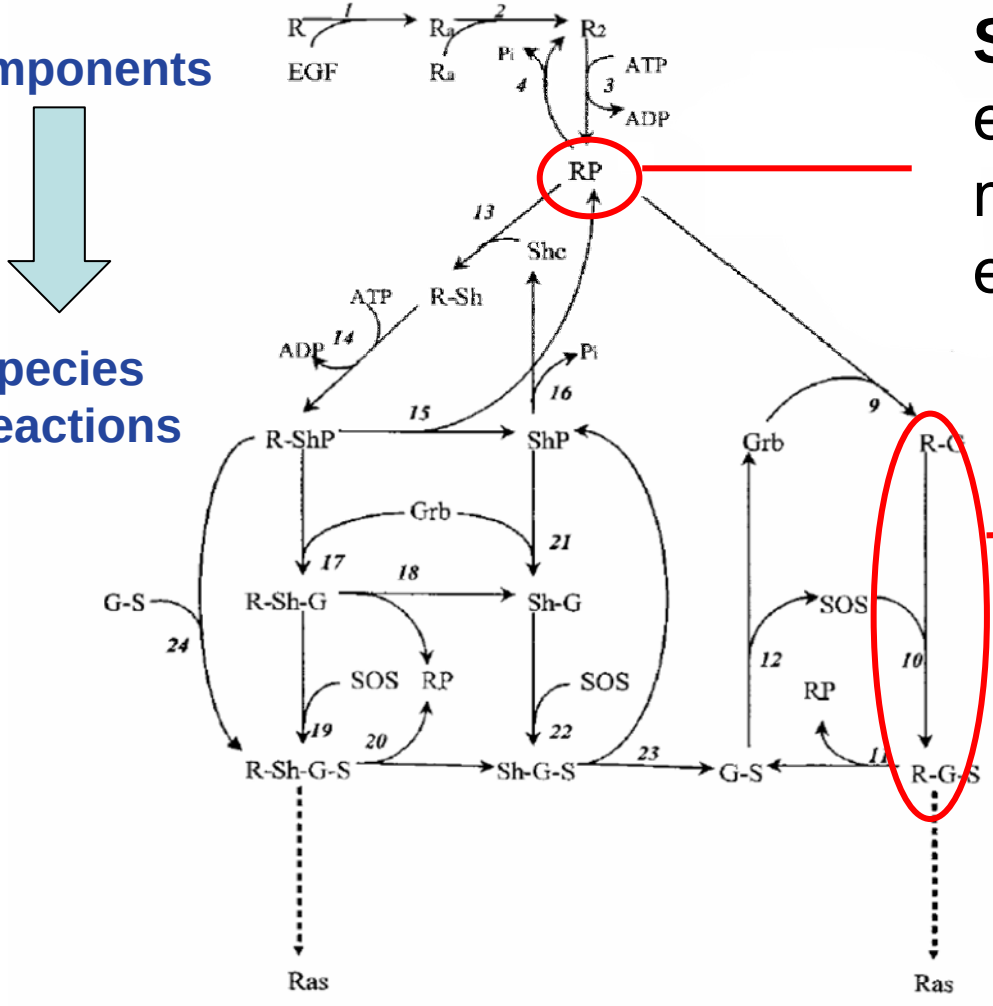
1. EGF binds EGFR
2. EGFR dimerizes
3. EGFR transphosphorylates itself
4. Shc binds phospho-EGFR
5. EGFR transphosphorylates Shc
6. Grb2 binds phospho-Shc
7. **Sos binds Grb2 (Activation Path 2)**



A conventional model for EGFR signaling

5 components

18 species
34 reactions



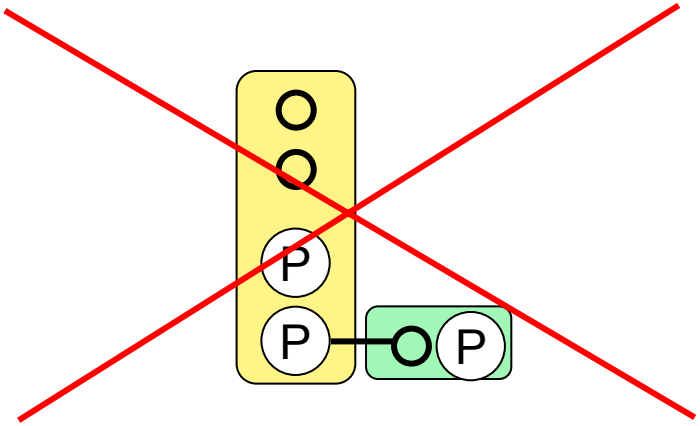
Species: One for every possible modification state of every complex

Reactions: One for every transition among species

Kholodenko et al., *J. Biol. Chem.* **274**, 30169 (1999)

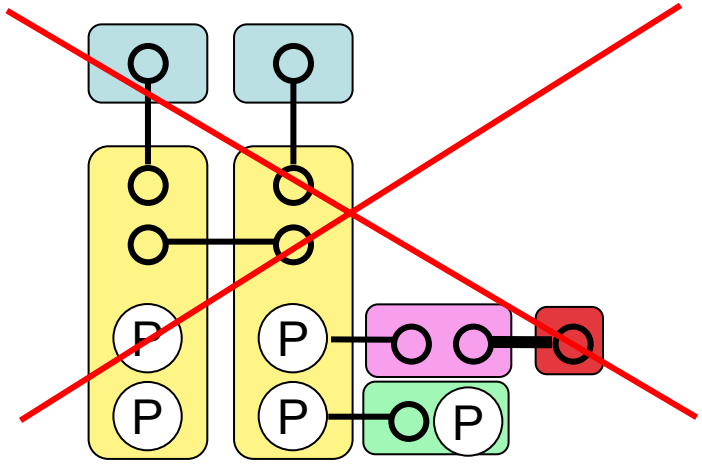
Excluded from the scheme

No modified monomers



1. Phosphorylation inhibits dimer breakup

No complexes



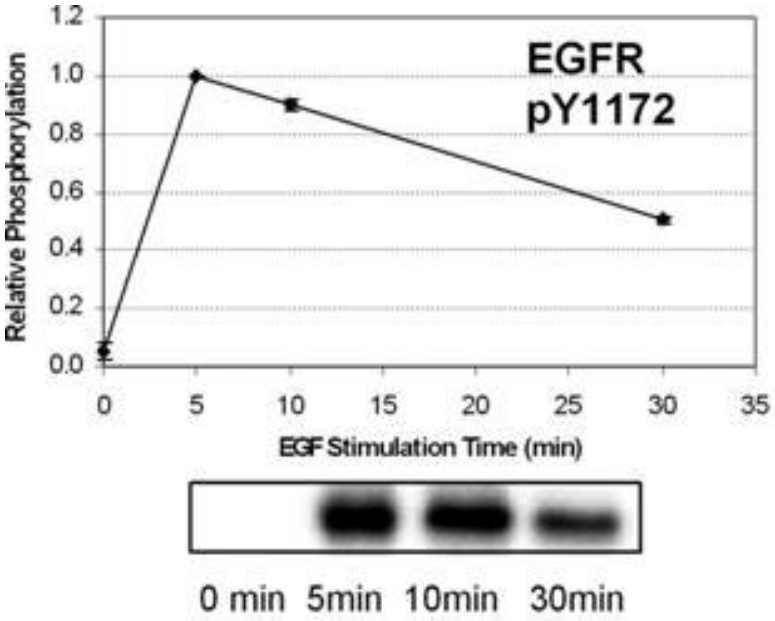
2. Adaptor binding is competitive

Summary of the conventional approach

- Combinatorial complexity gives rise to a multitude of species and reactions.
- Modelers assume (often implicitly) only some of these combinations are important.
- Assumptions are based on convenience rather than physical knowledge.
- Assumptions may be valid under some conditions, but not others.
- These assumptions cannot be tested without addressing combinatorial complexity.

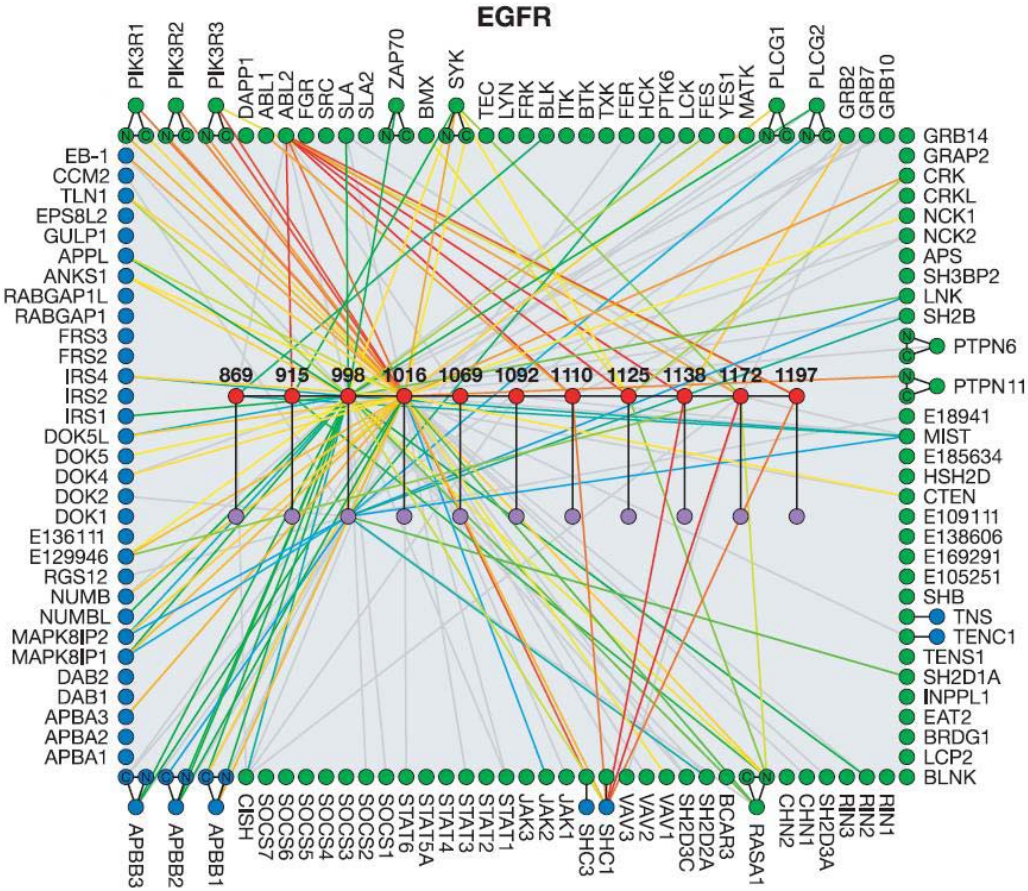
Experiments probe

the kinetics of multiple phosphorylation sites



Zhang et al.,
Mol. Cell. Proteomics **4**, 1240 (2005).

affinities for multiple binding partners



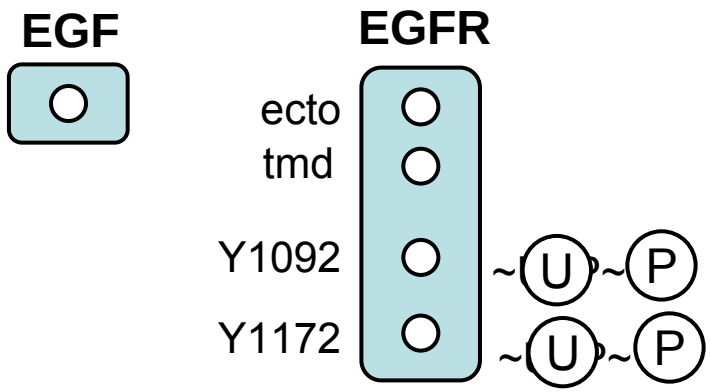
Richard B. Jones et al.,
Nature 439, 168-174 (2006).

Rule-based modeling is a way to handle combinatorial complexity

- **Assumption of proteins modularity**:
  - Signalling molecules consist of functional domains
  - Interactions depend on a limited set of features of signalling molecules, and are “local” with respect to these functional domains.
- The evolution of biological system is defined by **rules** describing activities, potential modifications and interactions of the domains of signaling molecules.
- Computer algorithm **automatically generates** all molecular species and reactions implied by rules.

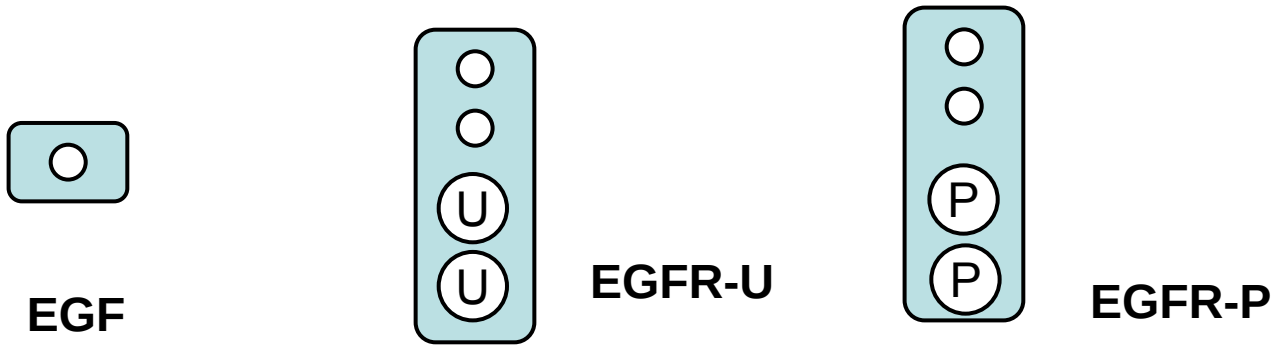
Instead of the list of species a user specifies

a) Biomolecules and their components



Components of proteins may have attributes, e.g. conformation or phosphorylation state.

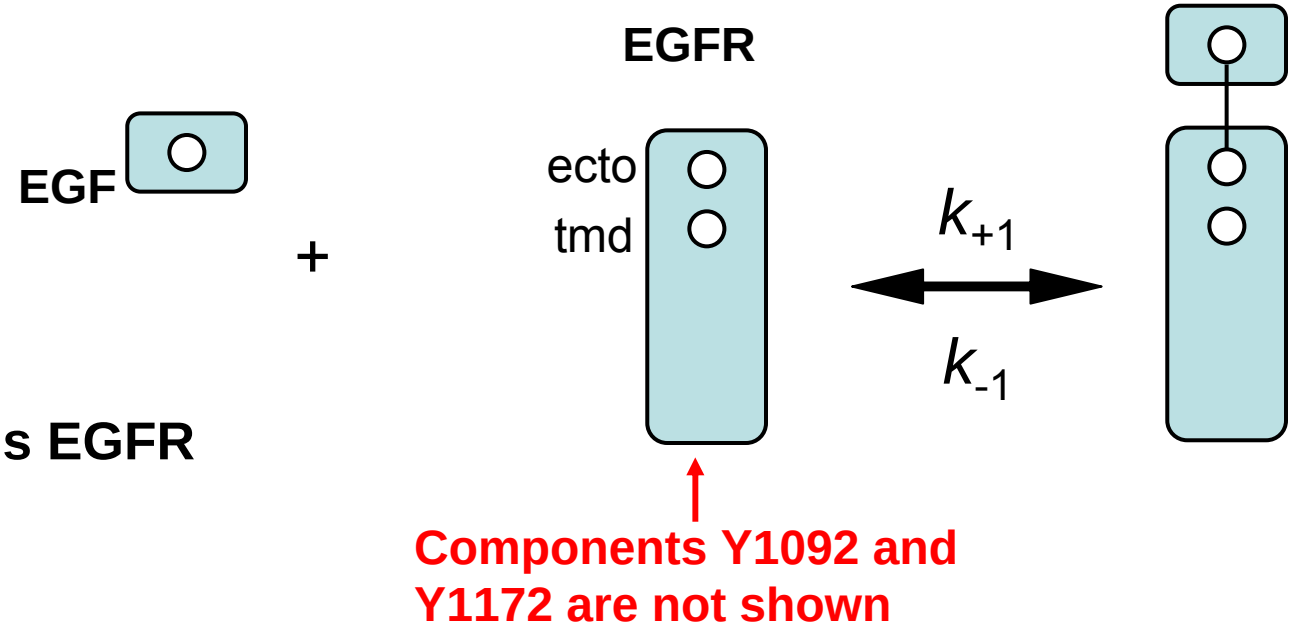
b) Species existing before simulation



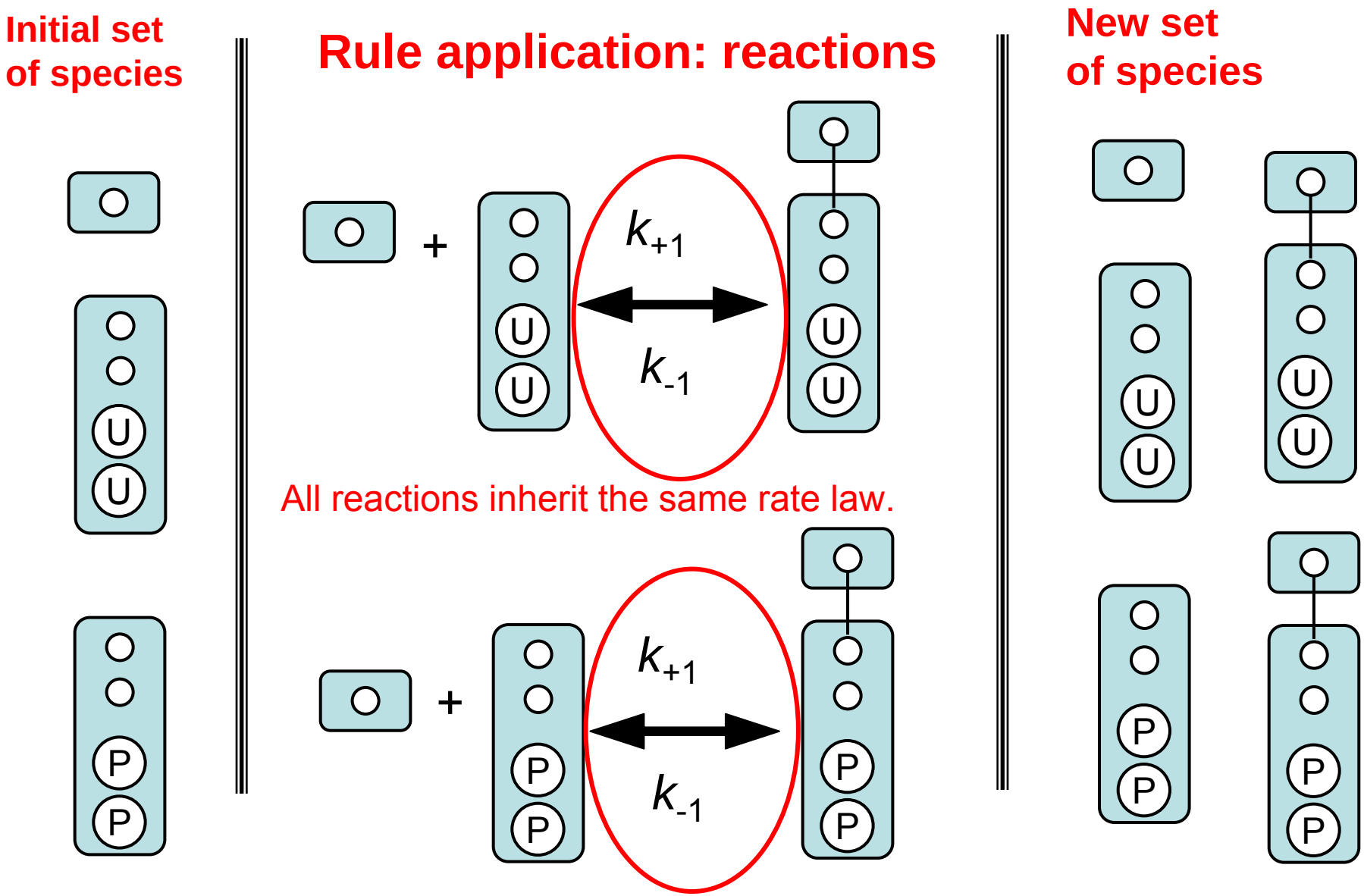
Instead of the list of reactions a user specifies

c) Rules that generate reactions and species

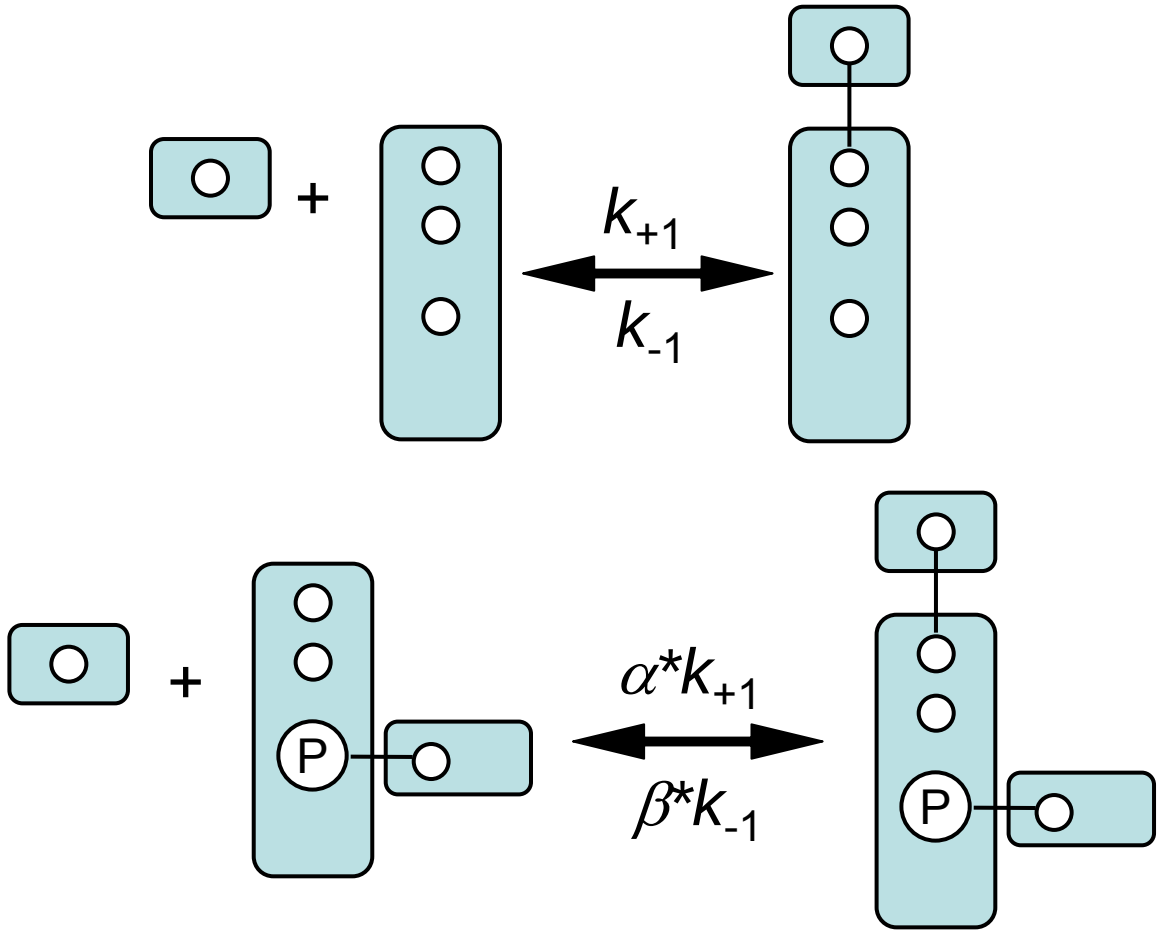
- User specifies a rule for each **experimentally-testable** feature of the system (*Example: kinetics of ligand-receptor binding is independent of receptor cytosolic tail modifications*).



Rules generate reactions and new chemical species

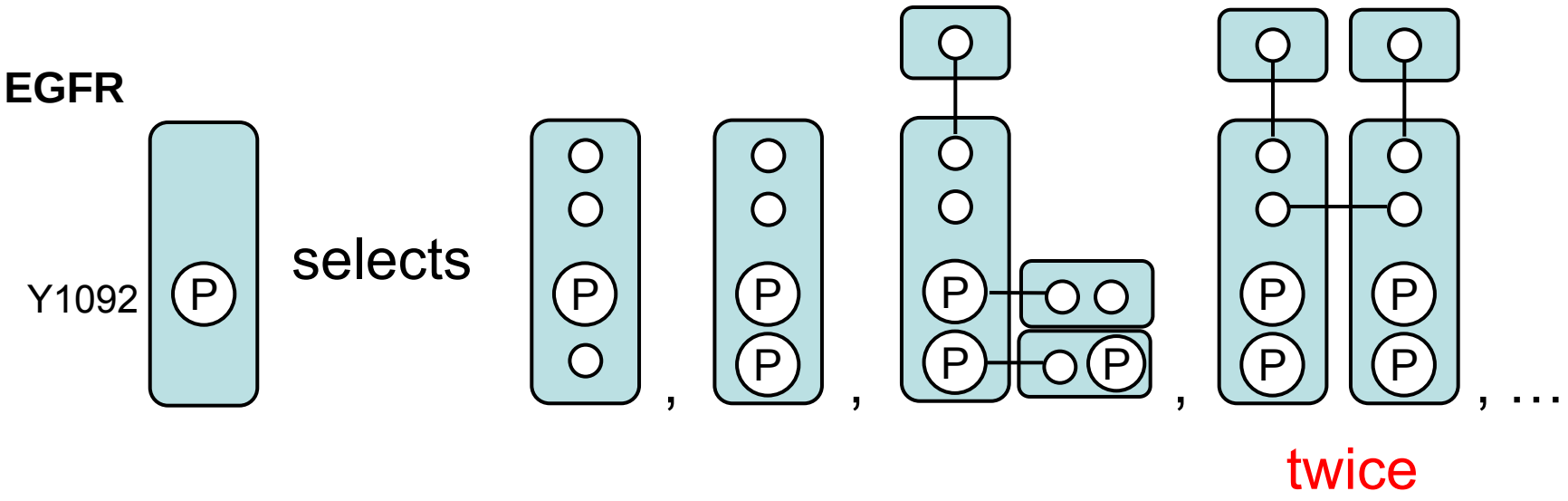


Extendibility and refinement of rules



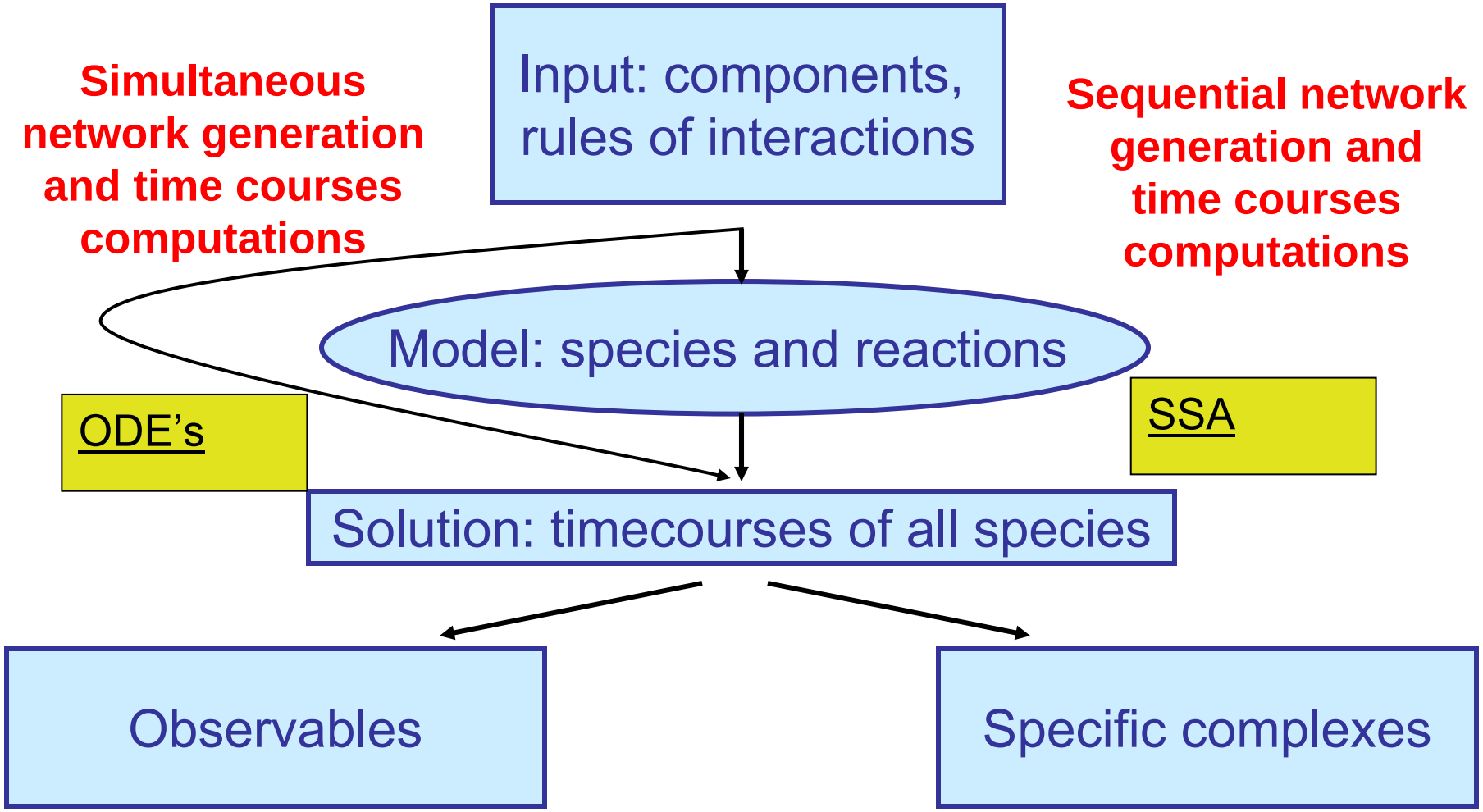
Revise rules to account for context (steric clashes, cooperativity).

Predictions are reported as “observables”, corresponding to groups of species with the same properties



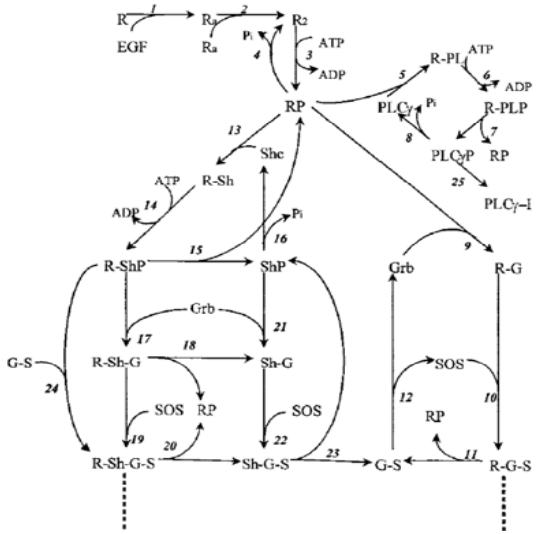
Pattern that selects EGFR phosphorylated at Y1092.

BioNetGen modeling



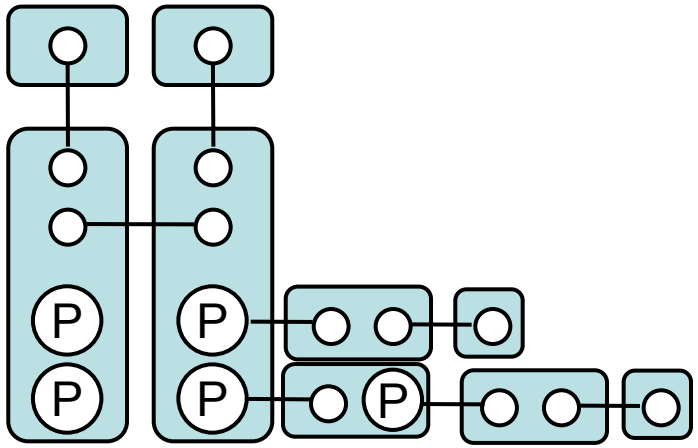
Rule-based version of a reaction scheme

18 species
34 reactions
37 parameters



Kholodenko et al. *JBC* (1999).

356 species
3749 reactions



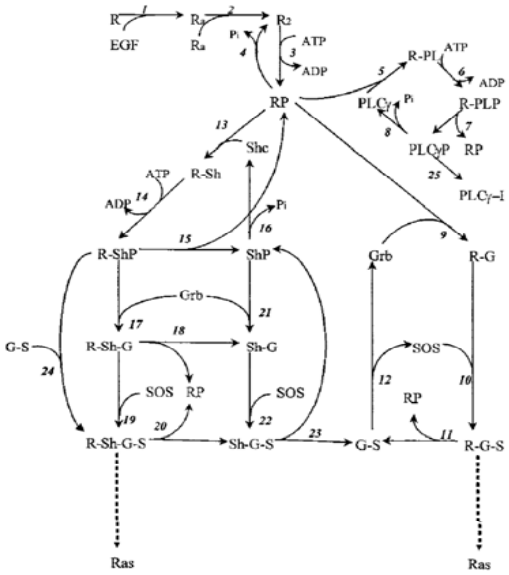
Blinov et al. *Biosystems* (2006).

- Same number of parameters as in reaction scheme
- Physical basis for rate parameters (e.g. binding constants)

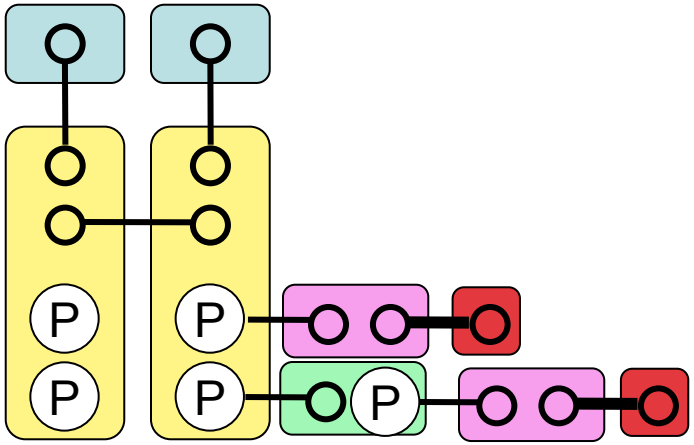
Rule-based version of the Kholodenko model

- 5 molecule types
- 23 reaction rules
- No new rate parameters (!)

18 species
34 reactions

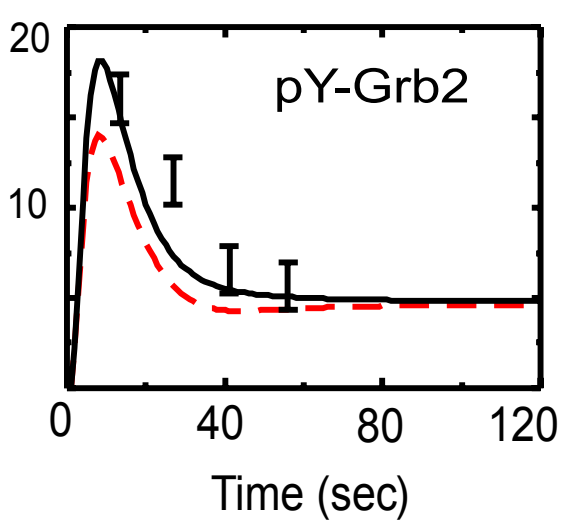


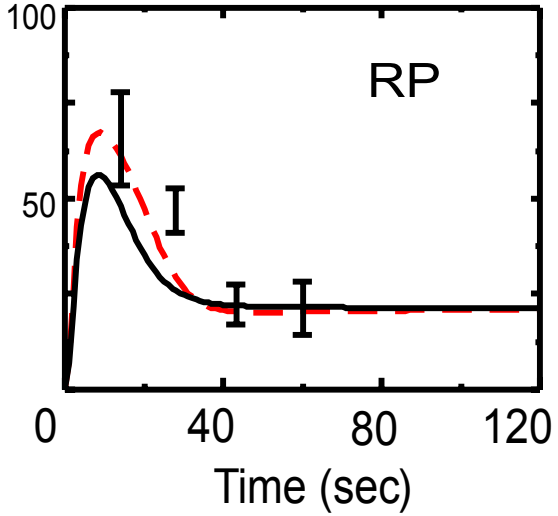
356 species
3749 reactions



Blinov et al. *Biosystems* **83**, 136 (2006).

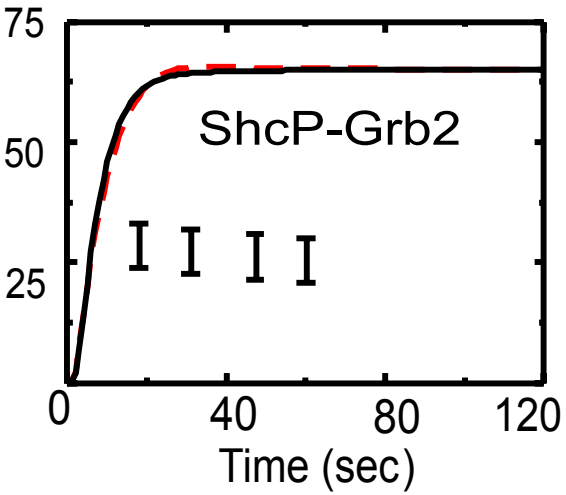
Fit to experimental measurements

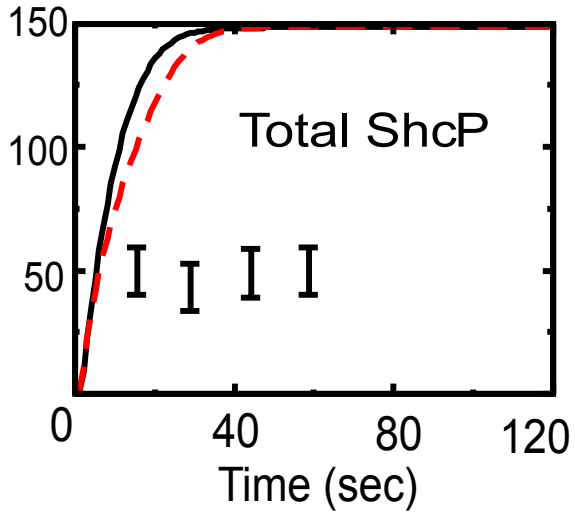




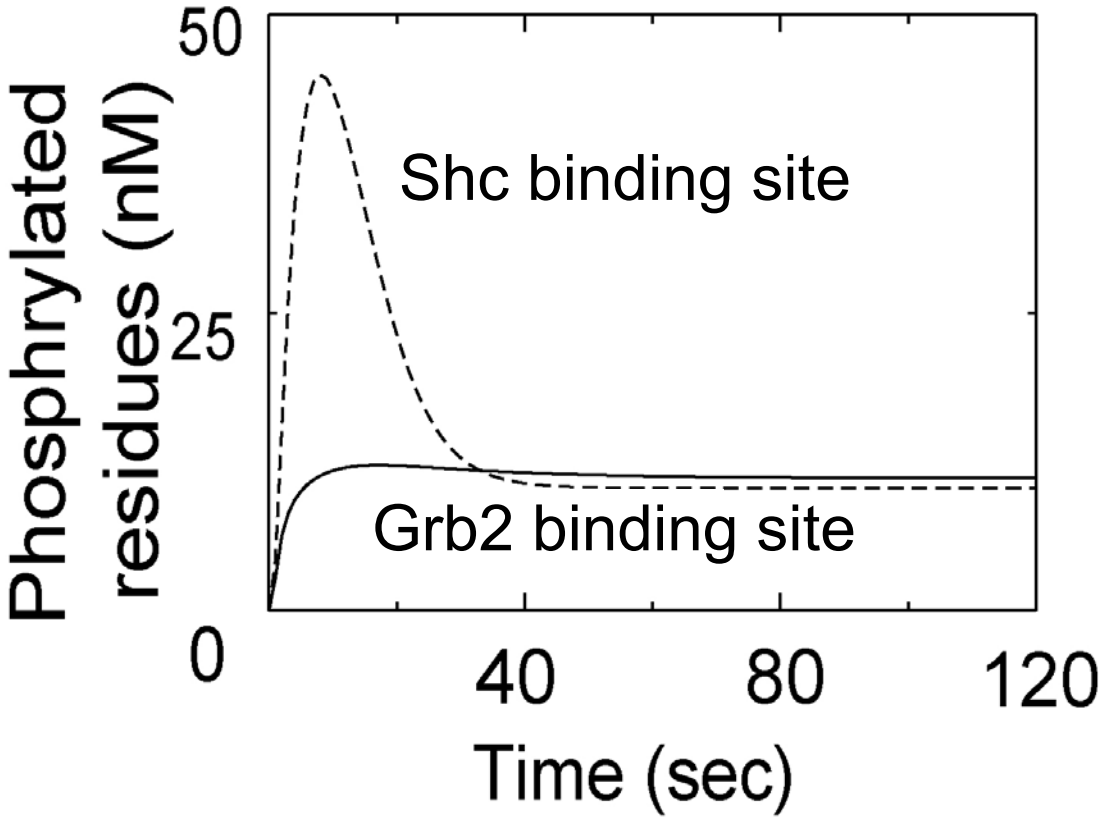
Pathway-like model

Network model





Rule-based model predicts distinct kinetics for two phosphorylation sites

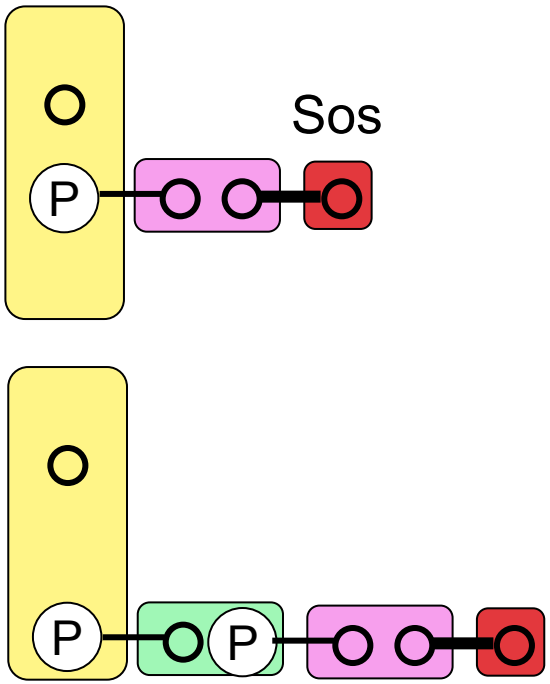


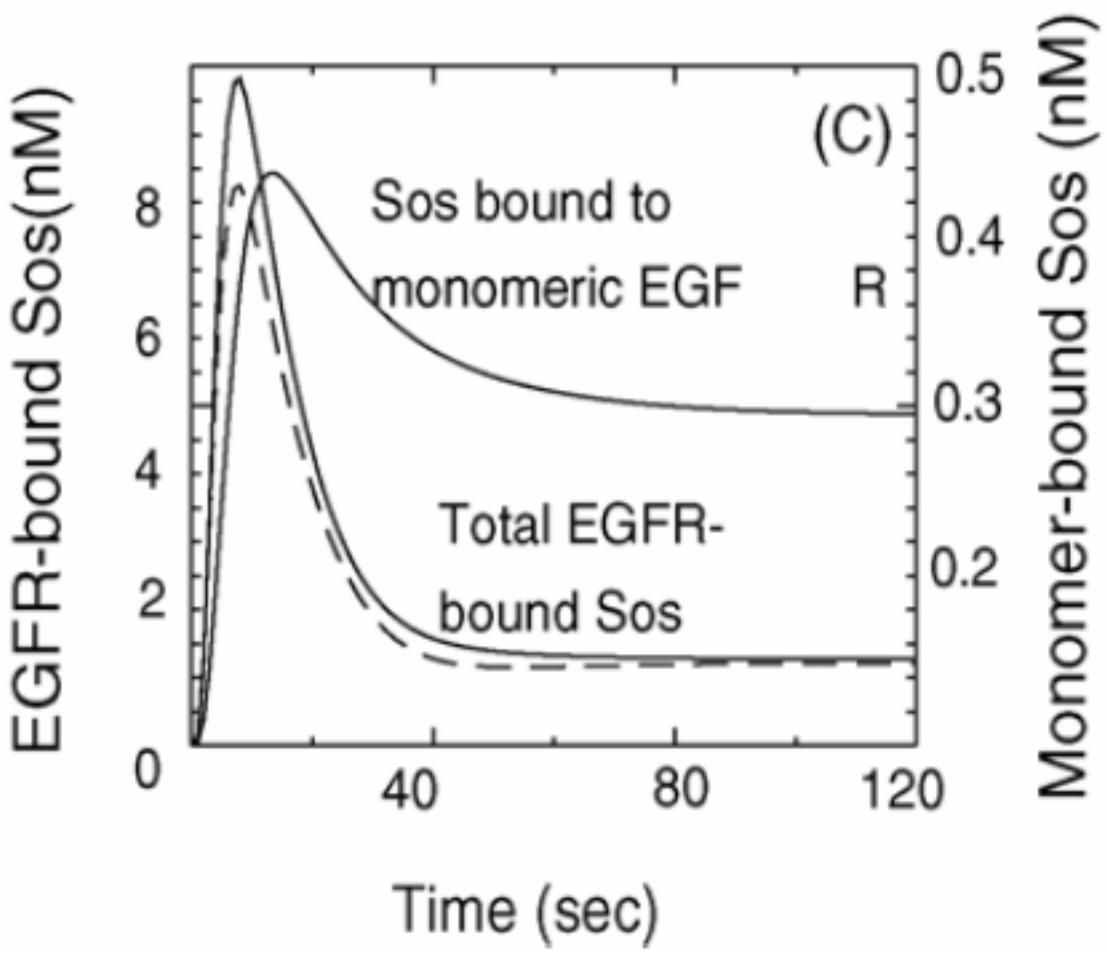
Recent experiments suggest that binding partner can affect phoshporylation kinetics.

Schulze et al., *Mol. Syst. Biol.* (2005)

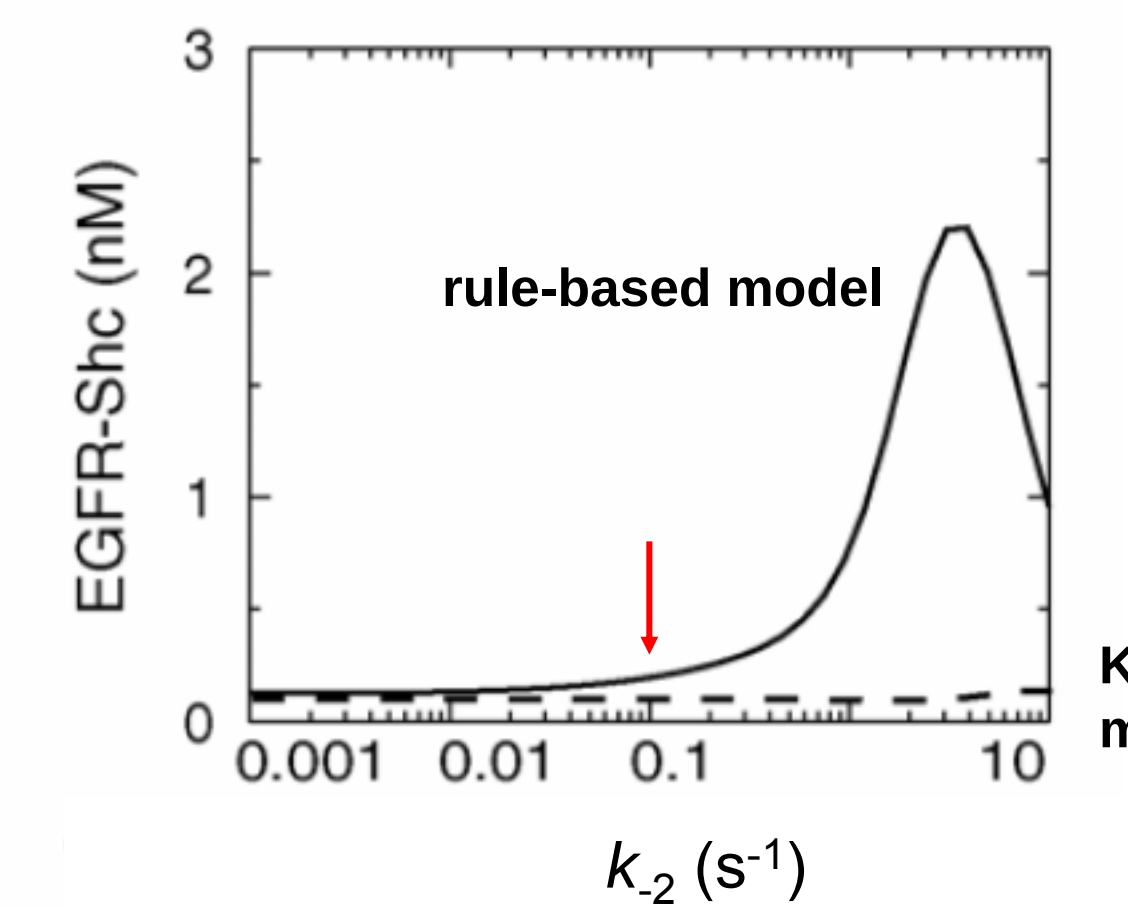
Also predicts monomers make substantial contribution to steady state Sos activation

36% of active Sos associates with EGFR monomers



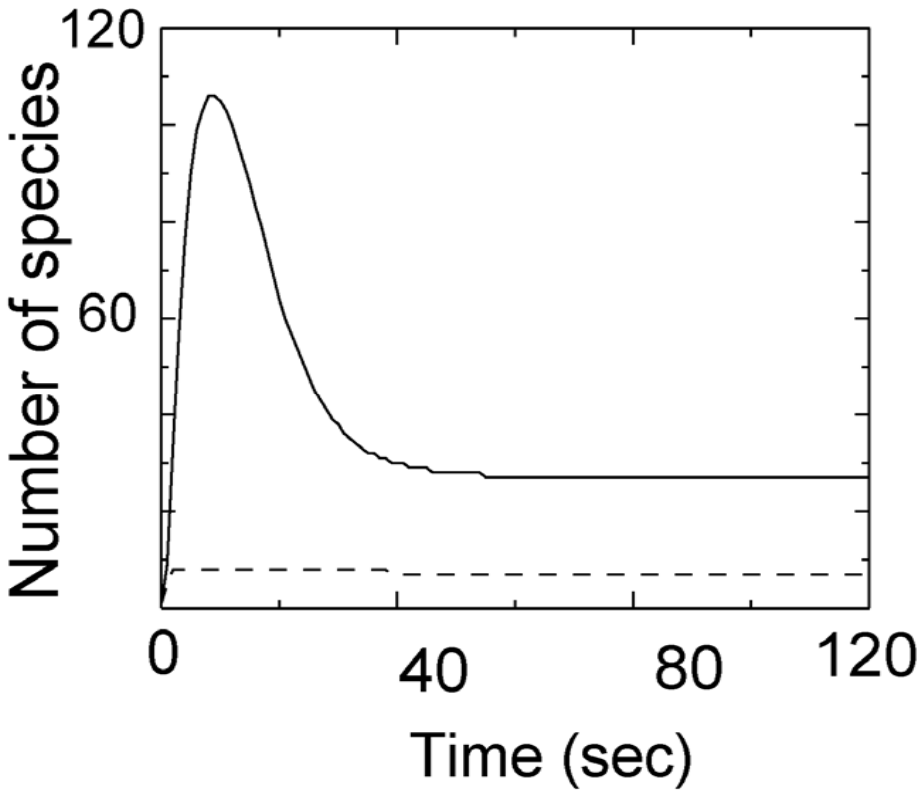


Strong differences when dimer dissociation rate is varied



Molecular diversity

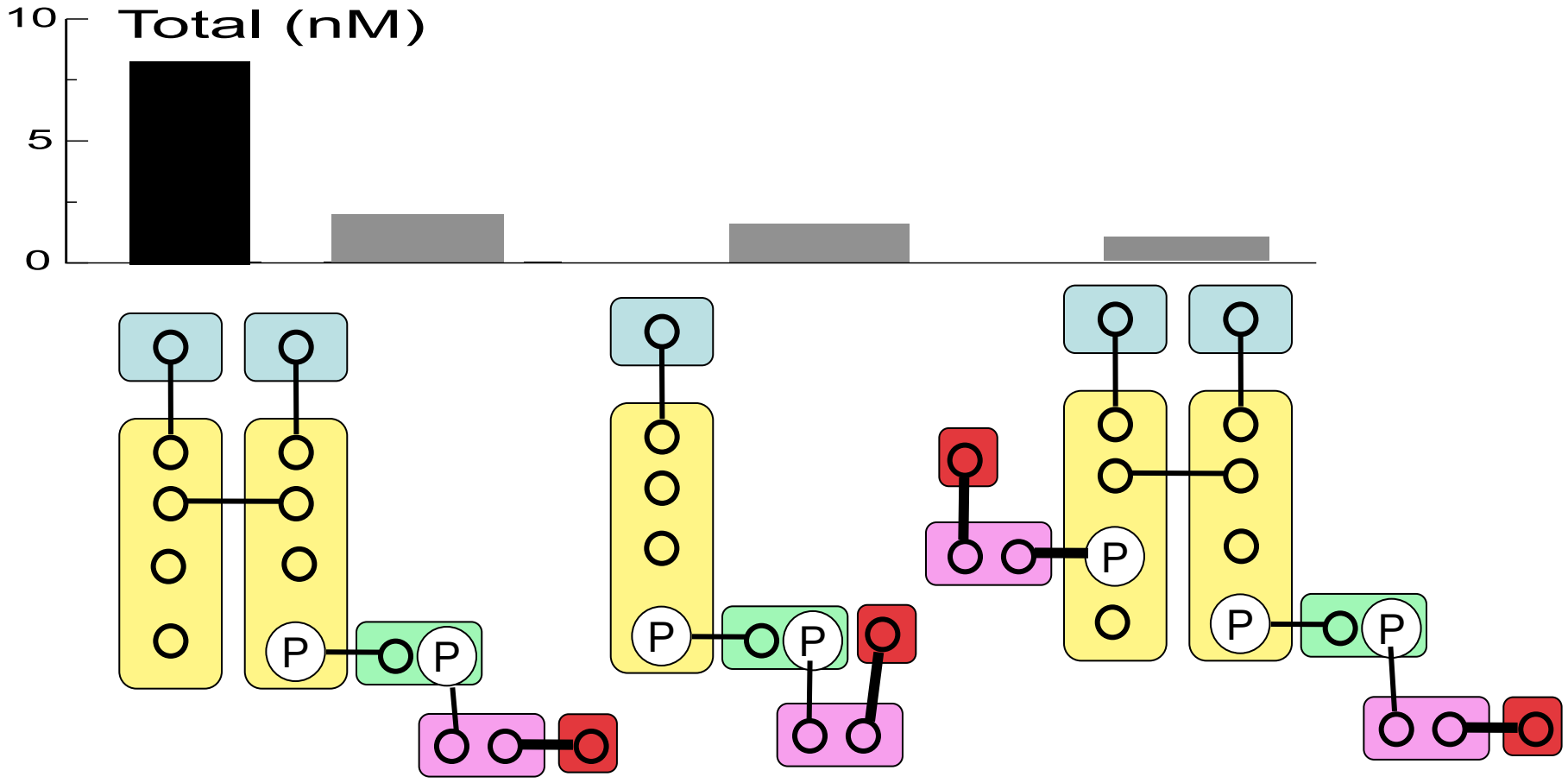
Much larger number of distinct chemical species is predicted to participate in signaling at short times than at steady state



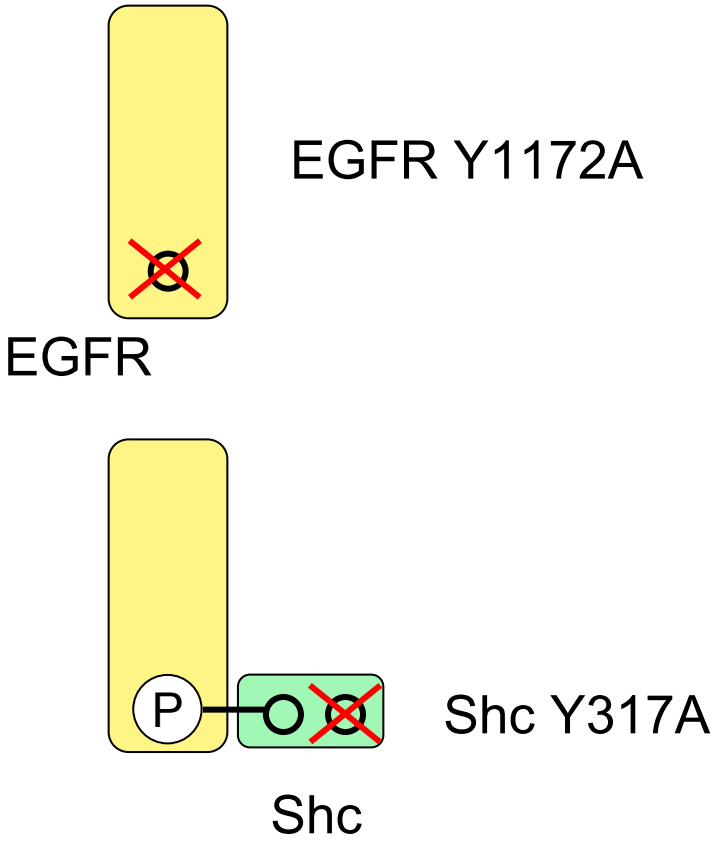
Solid line – network,
dashed line – pathway-like

Dominant molecular complexes

Few chemical species are predicted to account for almost all recruited Sos at steady state.



Results for two different knockouts of the Shc pathway



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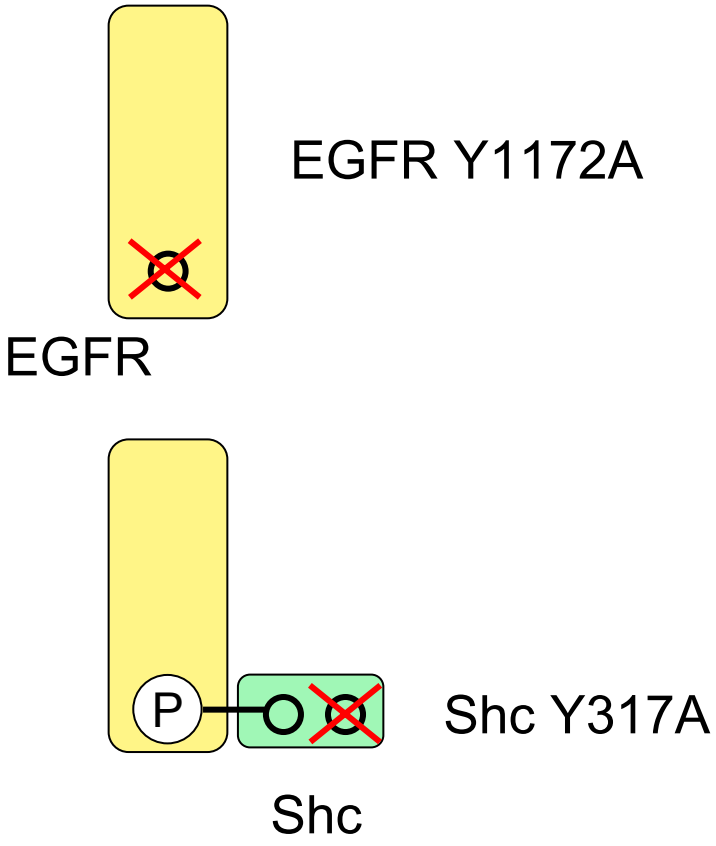
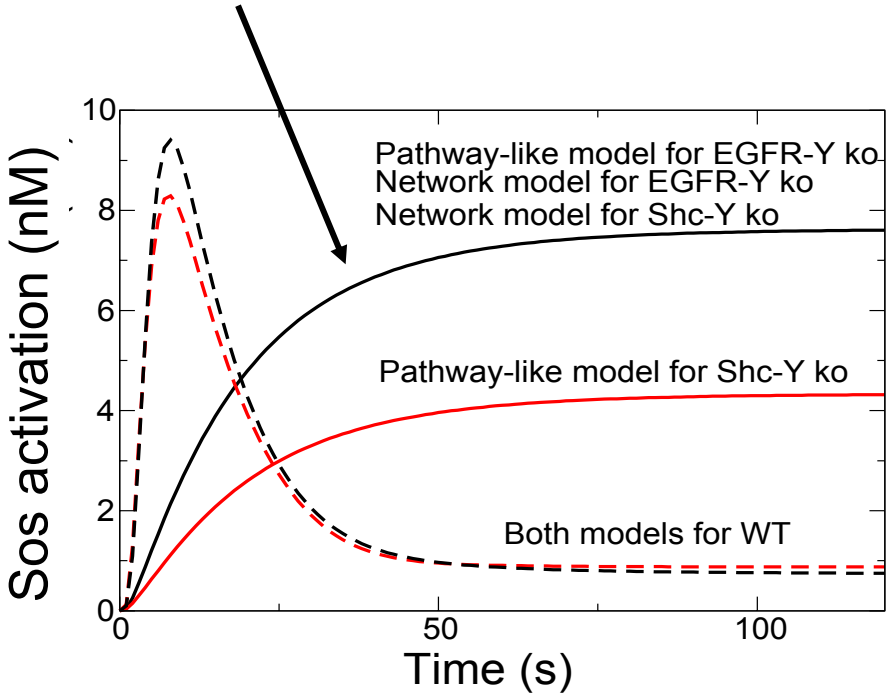
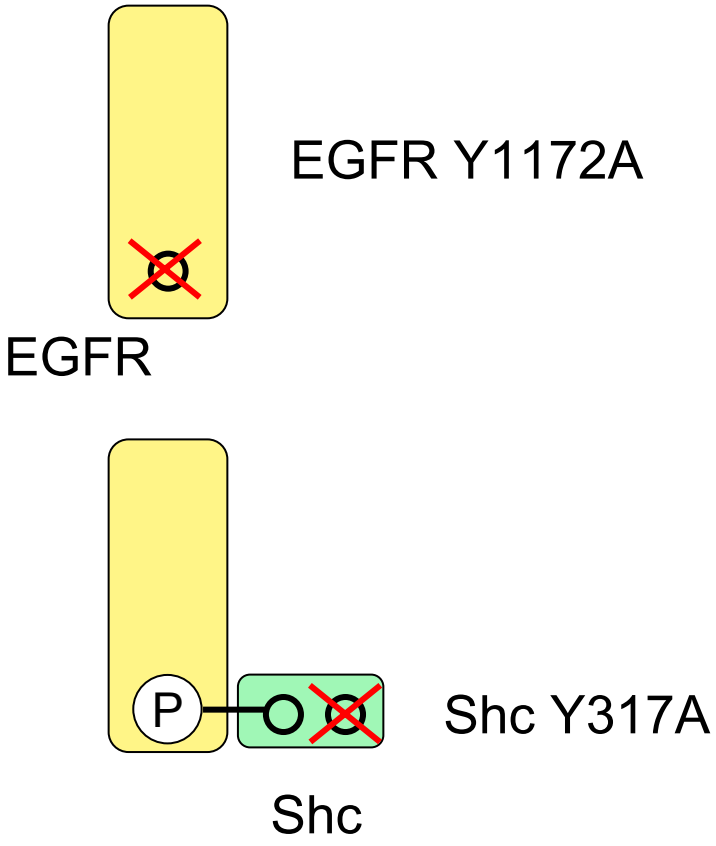


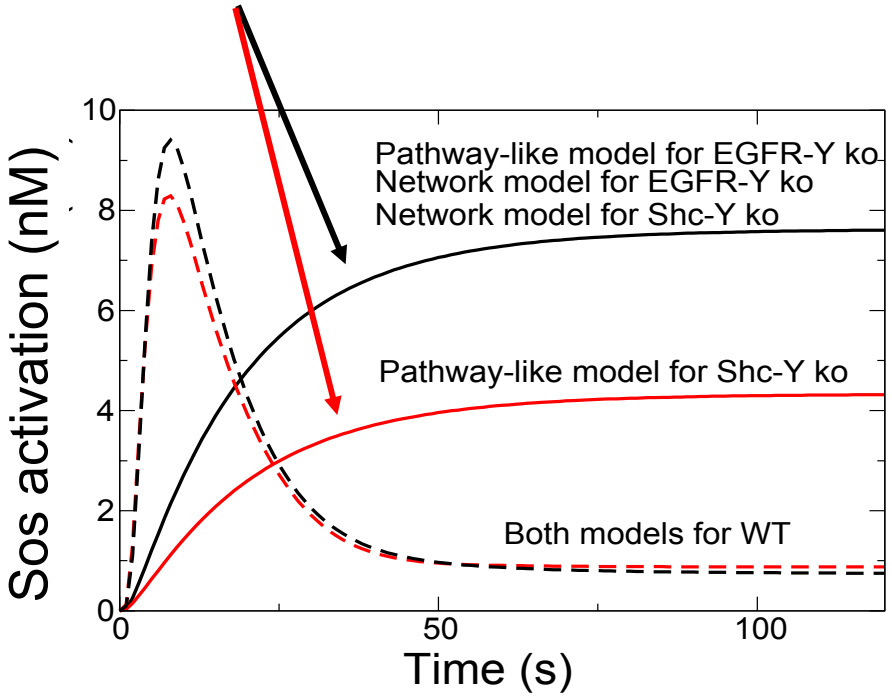
Rule-based model predicts same behavior for both knockouts



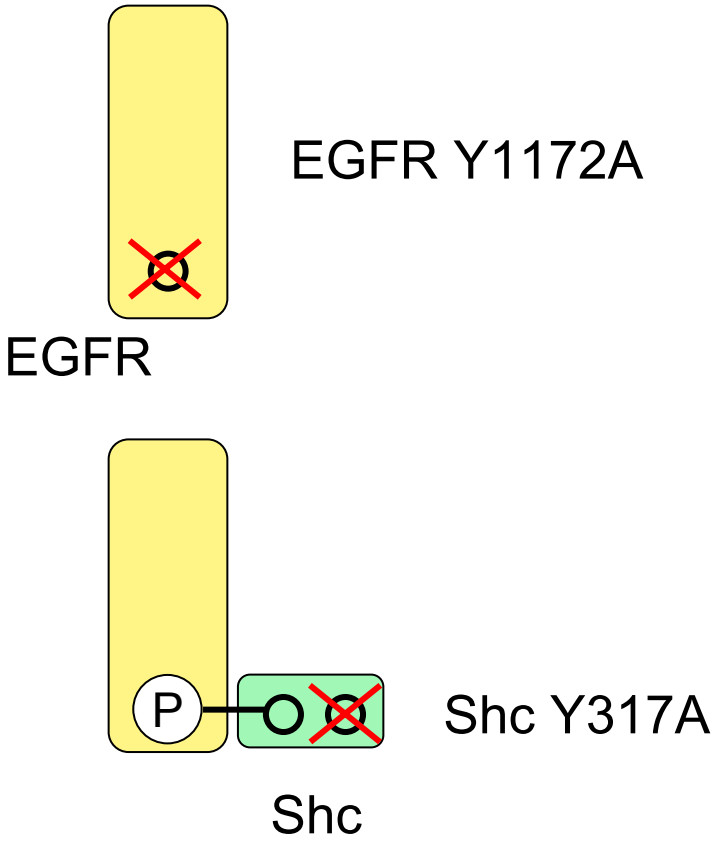
Results for two different knockouts of the Shc pathway



Kholodenko model predicts lower activation for Shc Y317A



Results for two different knockouts of the Shc pathway



Kholodenko model predicts lower activation for Shc Y317A

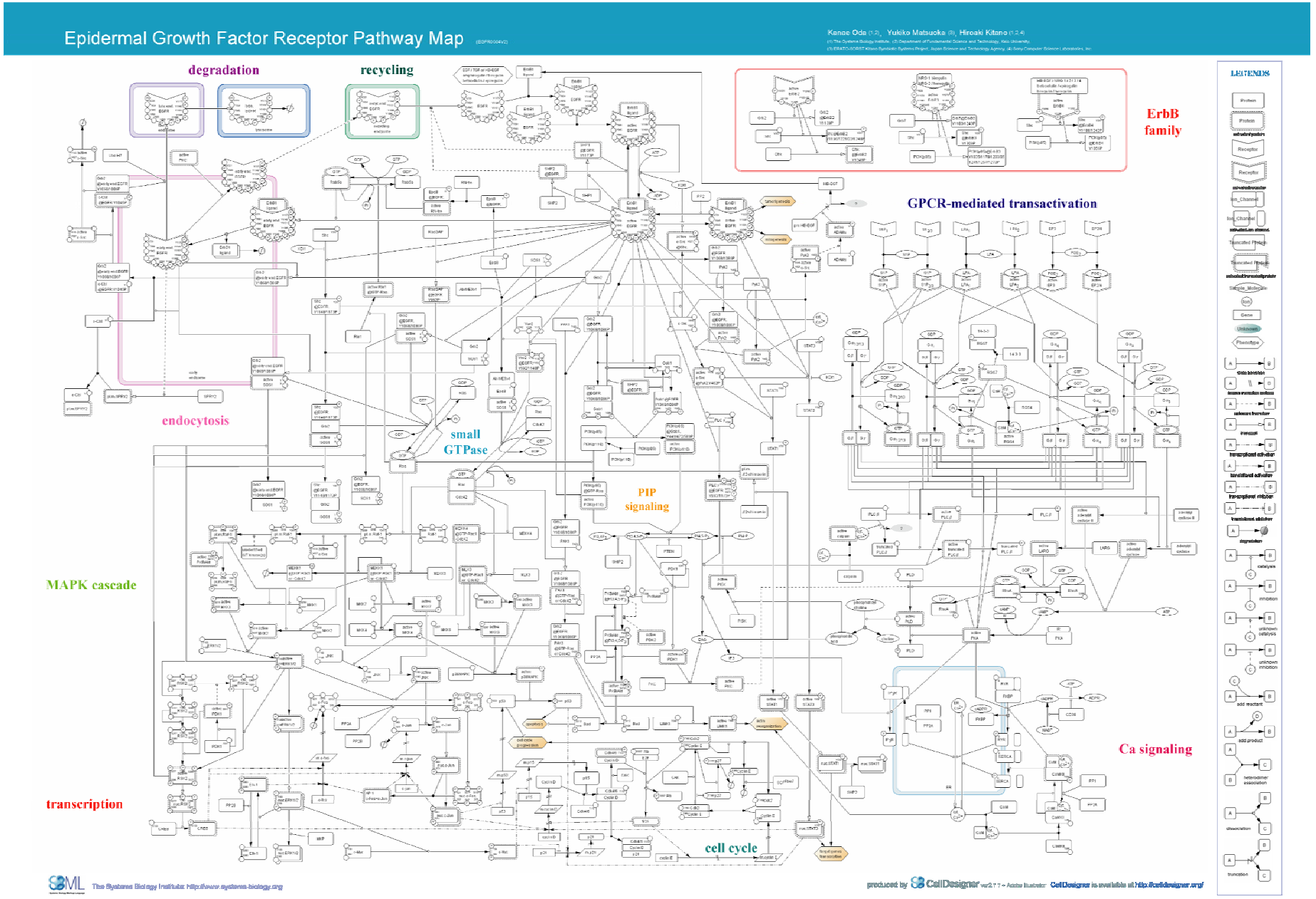
… because mutant Shc blocks binding of Grb2 (competitive binding)

What do we gain

- New quantitative predictions about specific domains, complexes, and interactions, in contact with kind of experiments biologists do (monitoring levels, knocking out and over-expression of specific domains).
- New qualitative predictions (tracing reaction sequences, dominant molecular species).
- Testing hypotheses about signalling mechanisms, e.g. competitive versus non-competitive protein binding.
- Testing effects of specific genetic manipulations, e.g. effects of knock-outs.

BNGL as a collaborative framework

Grand challenge: create a comprehensive rule-based model



Spatial modeling meets rule-based approach: BioNetGen at the Virtual Cell

- Rules can be used to generate spatial models accounting for combinatorial complexity:
  - Populate compartments with initial species.
  - Define “compartment-based” rules, with some rules generating species inside compartments and some rules defining trafficking between compartments.
  - Finally, define “spatial-based” rules, with each reactant and product species having a spatial location.