Alliance for Cellular Signaling

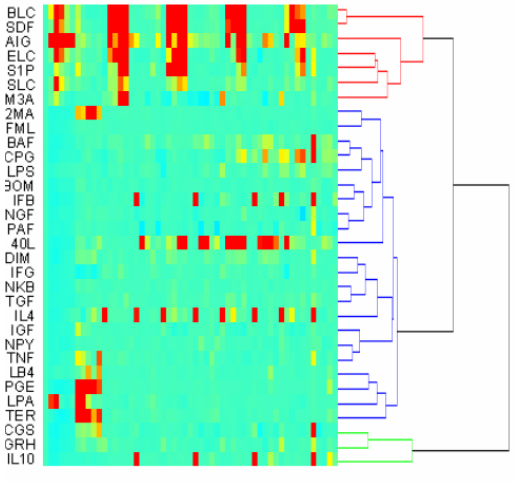
- 40 laboratories investigating basic questions in cell signaling
  - How complex is signal processing in cells?
  - What is the structure and dynamics of the network?
  - Can functional modules be defined?
- Key Advantage of AfCS: High quality data from single cell type
- All findings will be available to signaling community (www.signaling-gateway.org)

AfCS data

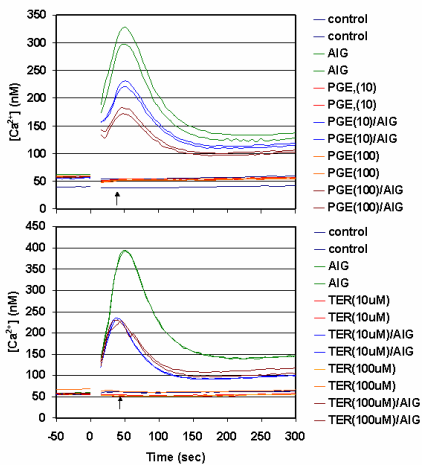
Inputs:

Cells are treated with uniform stimulus. There are 25 ligands applied individually and in pairs

Ouputs:



Gene Expression



Ca^{2+}, CAMP Time traces

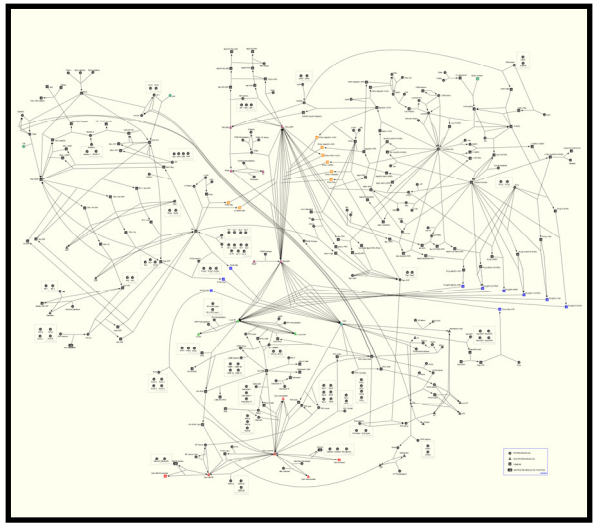
Other data:

- Protein-Protein interactions
- Phosphorylation states
- All can be repeated with knockdowns

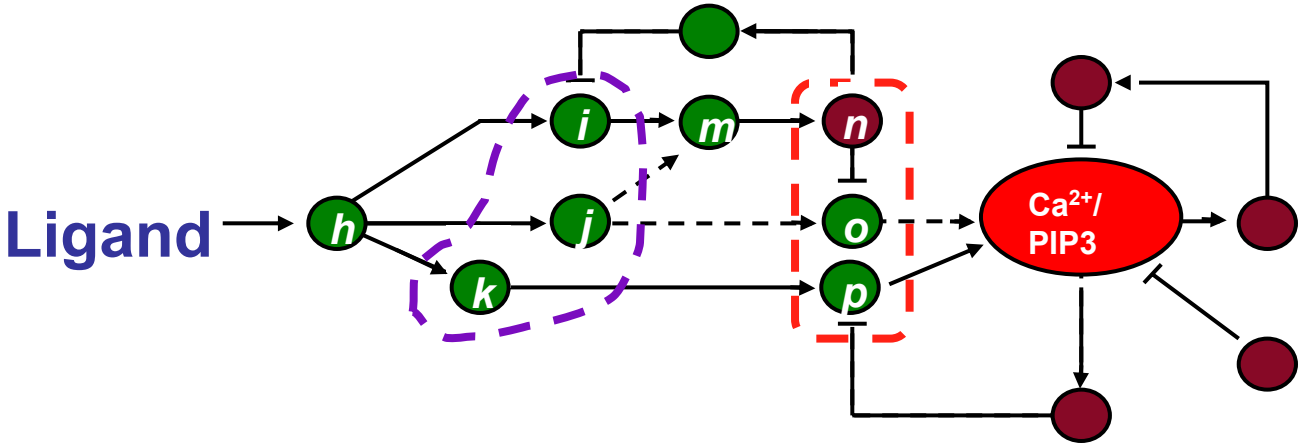
Mechanistic Modeling

Two efforts: large connection maps and evaluation of minimal models

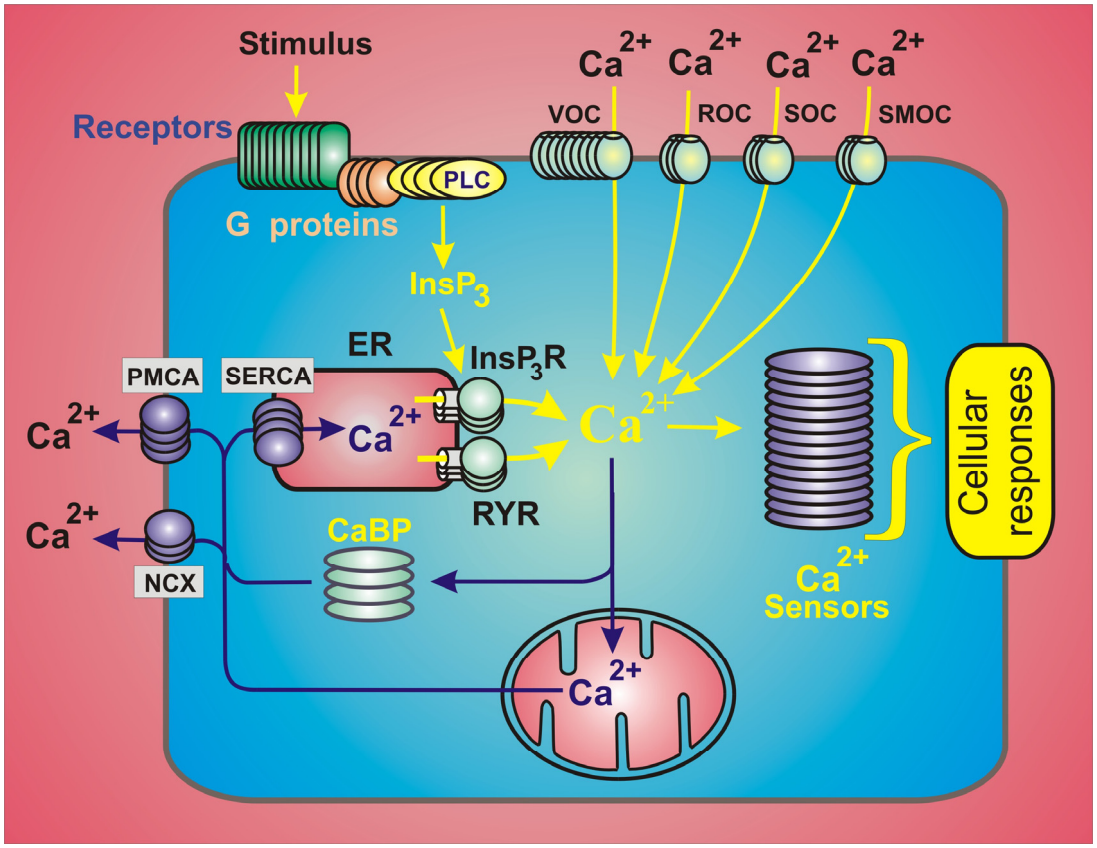
Detailed structure, no kinetics



FXM project: Smaller chunk with Kinetics.



More about Ca^{2+} signaling

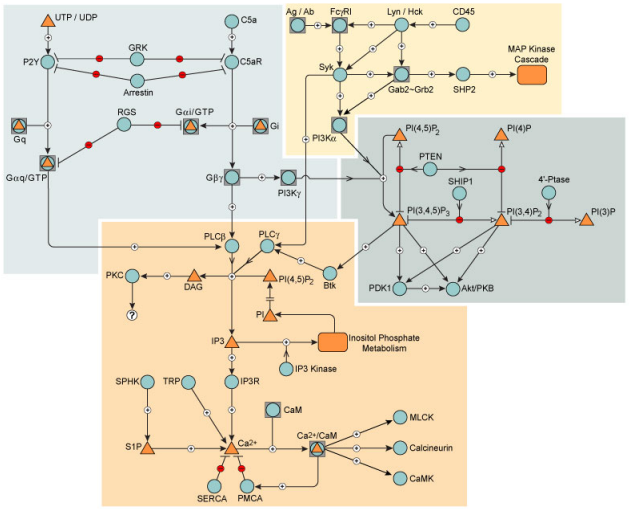


Calcium regulates many responses:

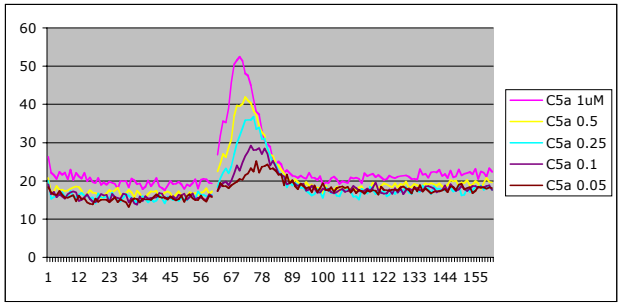
- Synaptic signaling (microseconds)
- Muscle Contraction (~milliseconds)
- Gene transcription (hours)
- Embryonic development (days)

AfCS FXM Ca^{2+} data and RNAi Knockdowns

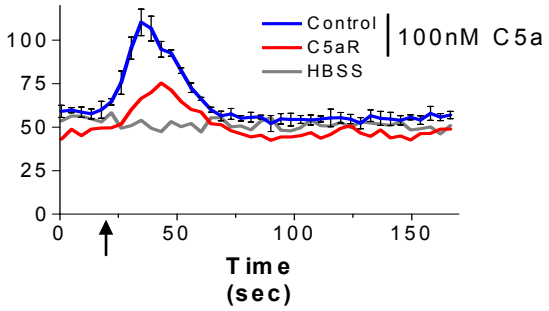
Chunk of pathway with knockdown points (red dots)



Dose Response

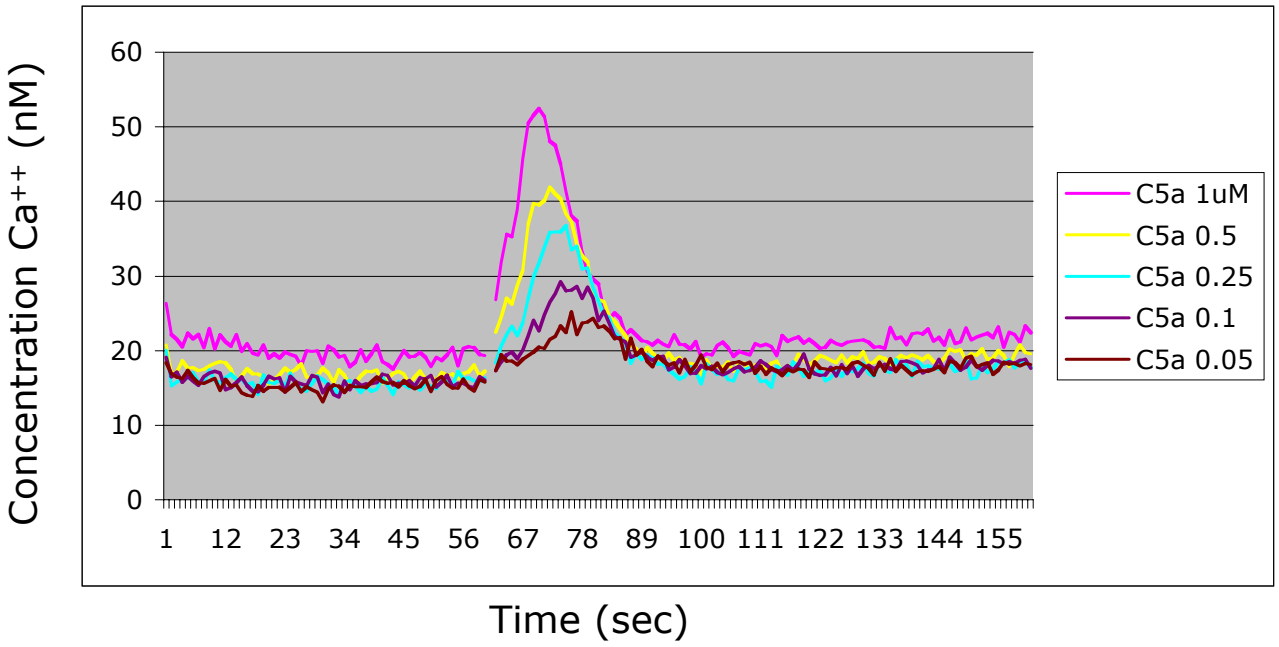


Receptor knockdown



Models, Data, and Uncertainty

- We've examined two Models:
  - A. Goldbeter, Proc. Natl. Acad Sci. 1990
  - T. Wiesner, Modeling in Physiology. 1996
- Today we will focus on the Wiesner model
- Ligand(c5a) dosage data from AFCS



Scalar-valued Data Features

- Considered Ca^{++} response to 4 different ligand dosages:
  - 1000nM
  - 500nM
  - 250nM
  - 100nM
- From each time-series we extracted 4 scalar-valued features:
  - 95% rise time (sec)
  - Time to peak (sec)
  - Decay time (relative to peak time) to settle to within 5% of peak (sec)
  - Peak concentration relative to steady-state level (μM)

Extracting data from time series

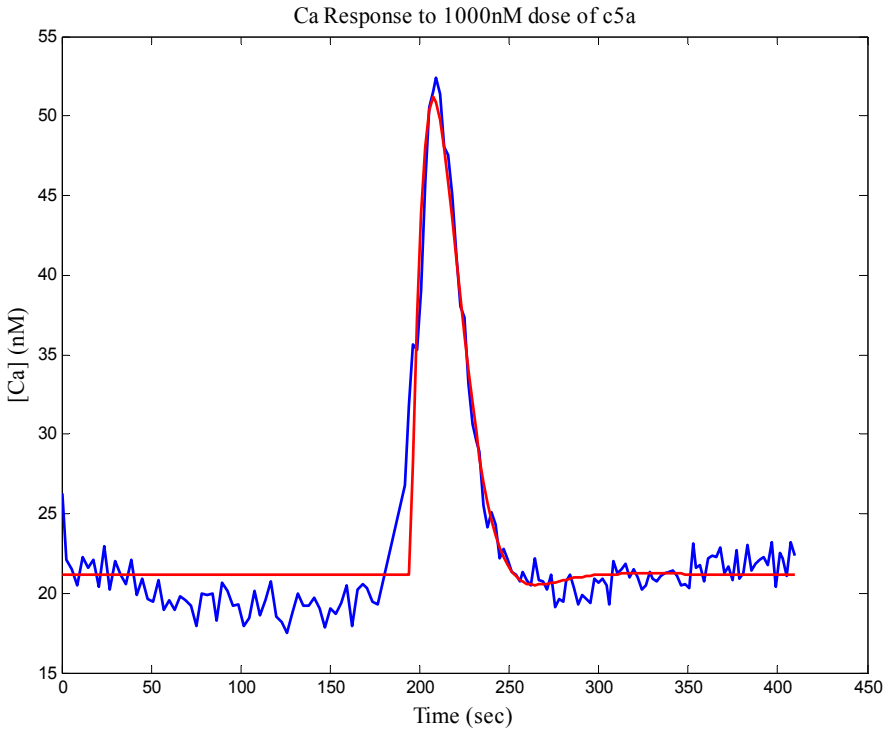
Fit a equation of the following form to the time series data to effectively filter the data in a manner that preserved the character of the spike

$$f(K,\omega,\zeta,\tau,\alpha) = K\frac{\omega}{\sqrt{1-\zeta^2}}e^{-\zeta\omega*(t-\tau)}\sin\left(\omega\sqrt{1-\zeta^2}*(t-\tau)\right)+\alpha$$

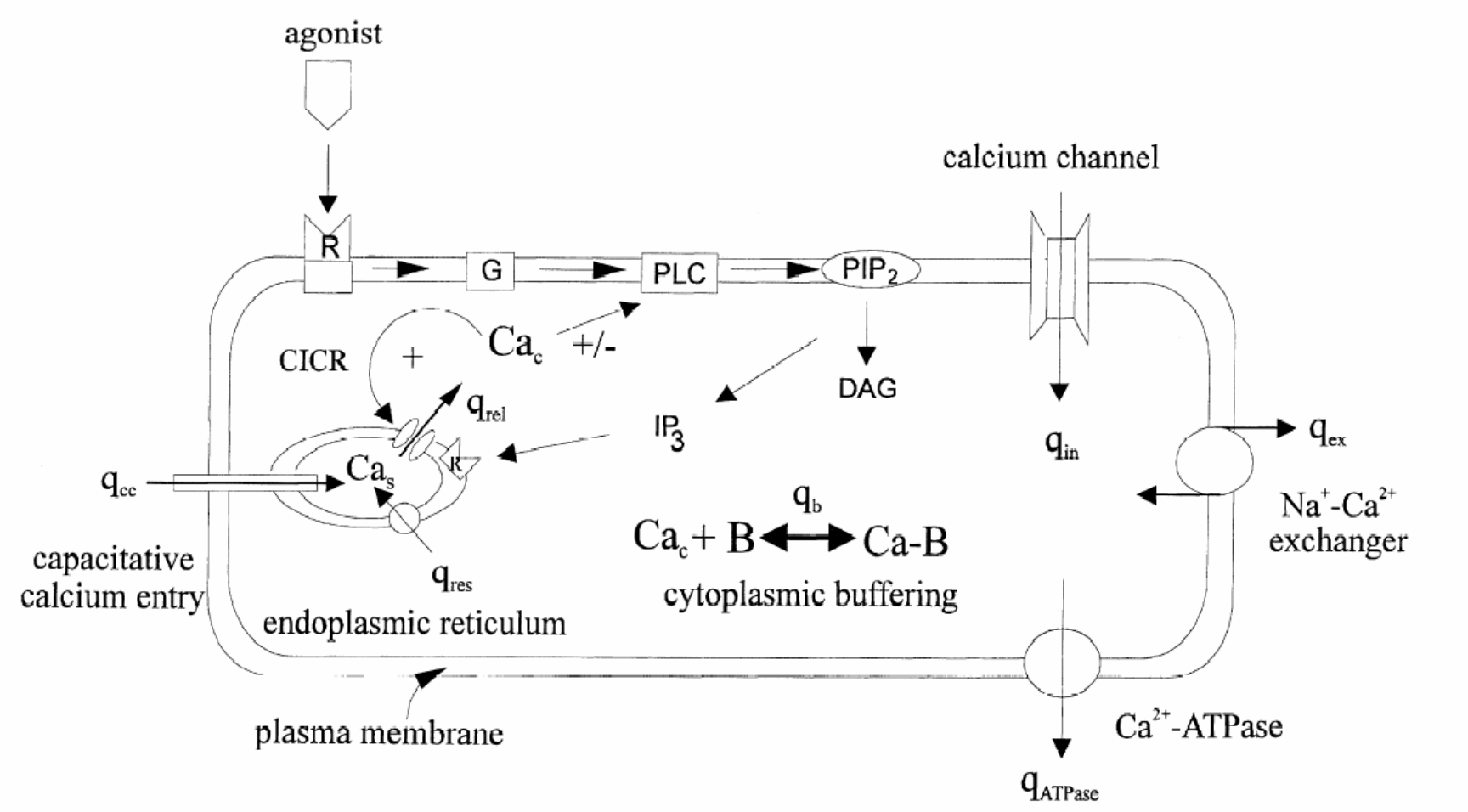
Feature values obtained from f were used as the "data" d_i in our analysis.

For lack of documentation, we specified an uncertainty level $u_i = 0.2d_i$

It would be interesting to use K, ω, ζ, etc. as the features



Schematic of Wiesner Model

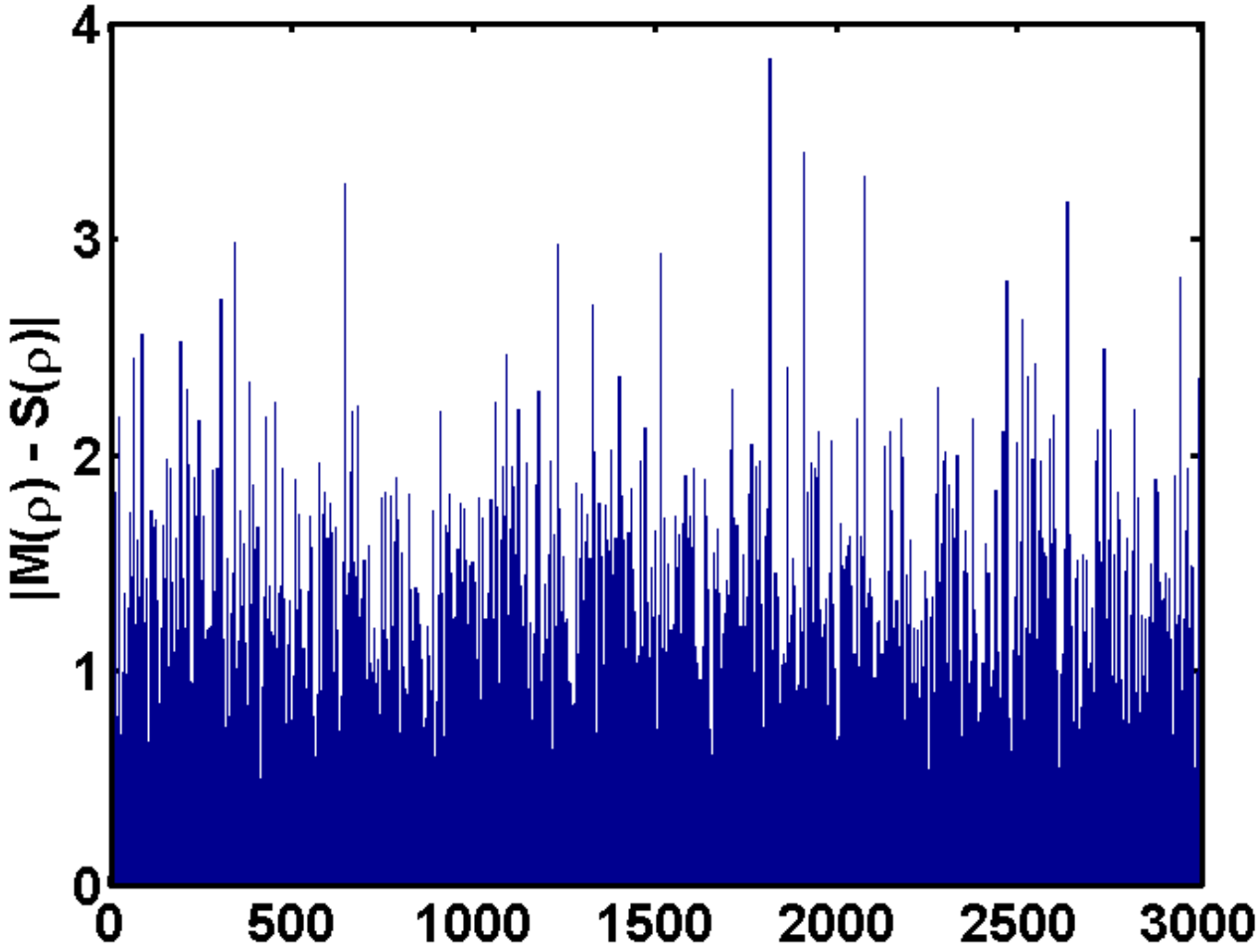


Parameters

- 27 Parameters drawn from
  - Rate constants
  - Cell features (e.g., # of receptors)
  - Initial conditions of species concentration
  - Binding coefficients
- Papers/literature provided nominal parameter values.
- We arbitrarily specified the parameter uncertainty level.
  - Typically an order of magnitude for rate constants.
  - Same or less for other parameter types.
- Parameters were normalized to lie in the hypercube $[-1,1]^n$
- 16 variables are relevant; the remaining 11 parameters have negligible influence (over the examined ranges) on the model predictions for the chosen features.

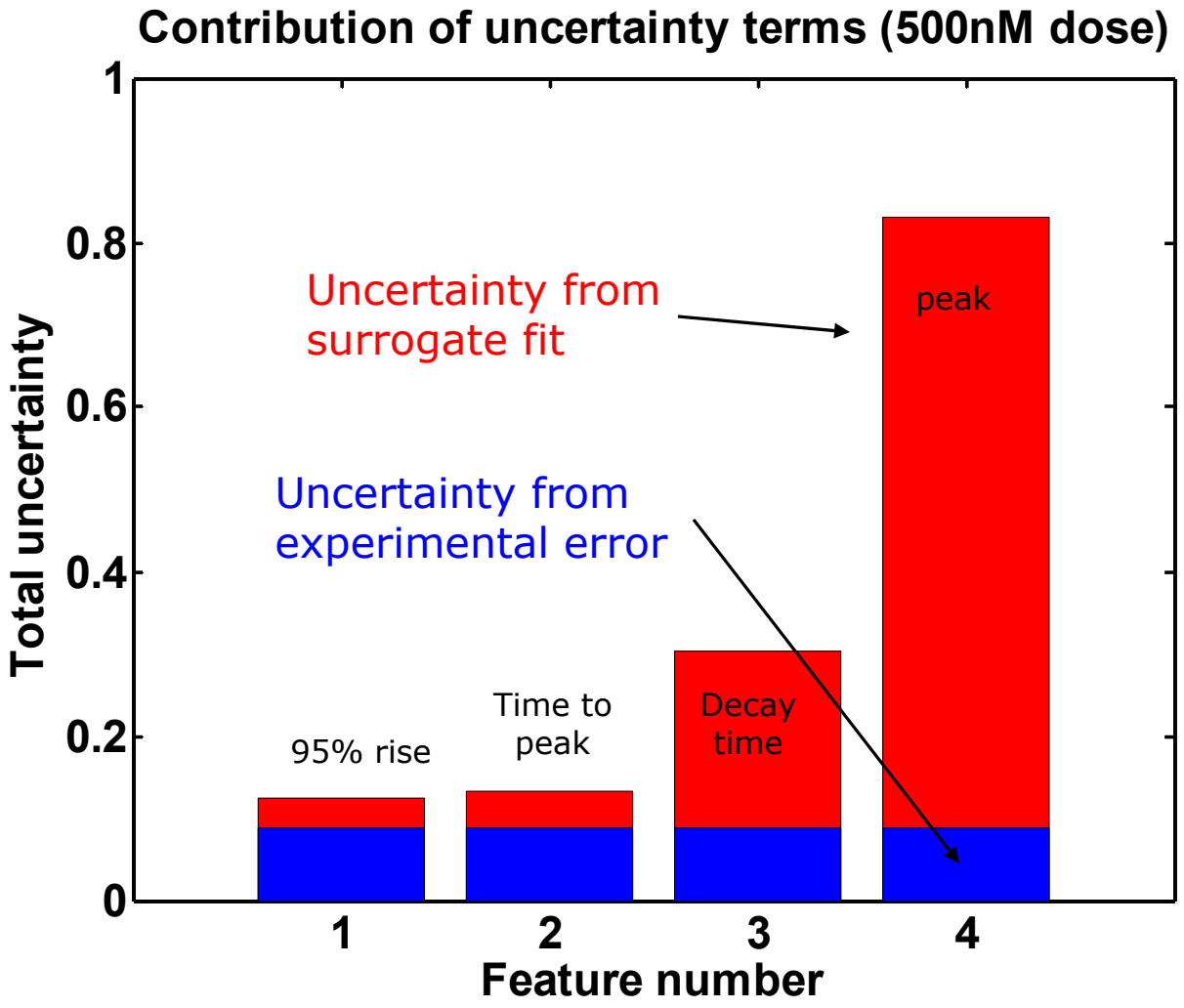
Surrogate Fitting Error Example

Fitting error for peak time at 500nM ligand dose



Surrogate Fits – final analysis

- Large fitting errors in peak Ca^{++} concentration



Consistency analysis

- Question: Does there exist a vector ρ 2 H so all feature observations are predicted within their respective uncertainty ranges?
- Consider

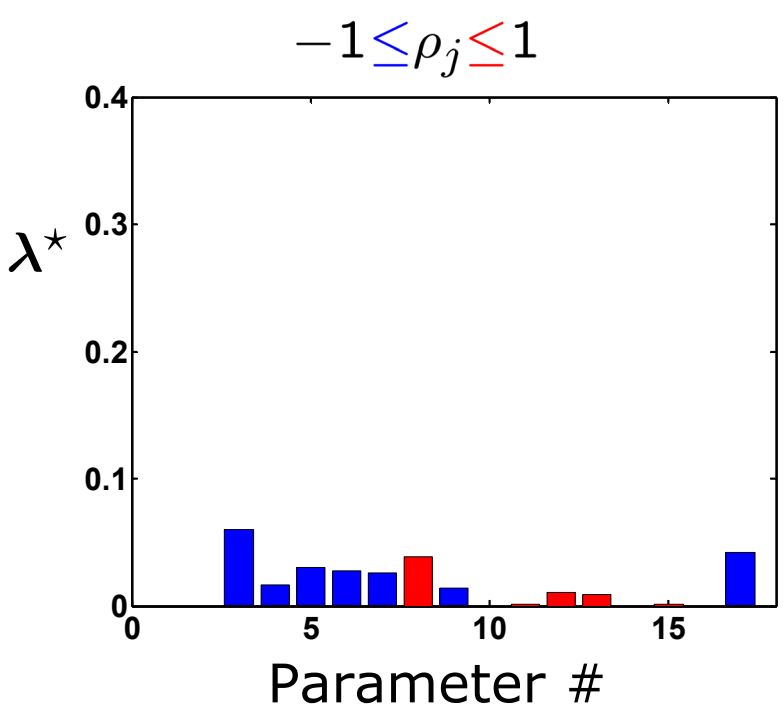
$$\mathcal{C} = \text{ maximum value of } \gamma$$

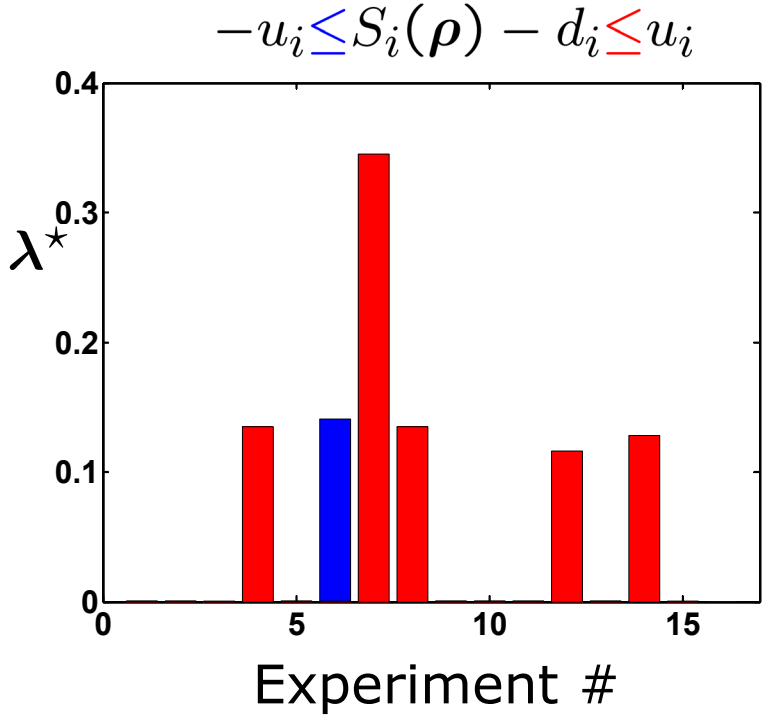
$$\text{subject to the constraints: } \begin{cases} |\rho_j| \leq 1 & j = 1, \ldots, n \\ |S_i(\boldsymbol{\rho}) - d_i| \leq u_i - \gamma & i = 1, \ldots, m. \end{cases}$$

- Negative C indicates there is no such vector so our assertions regarding the system are inconsistent.

Consistency Analysis Con't

- Including the fitting error in the analysis, our consistency test returns inconclusive results: C 2 [-0.07 0.10]

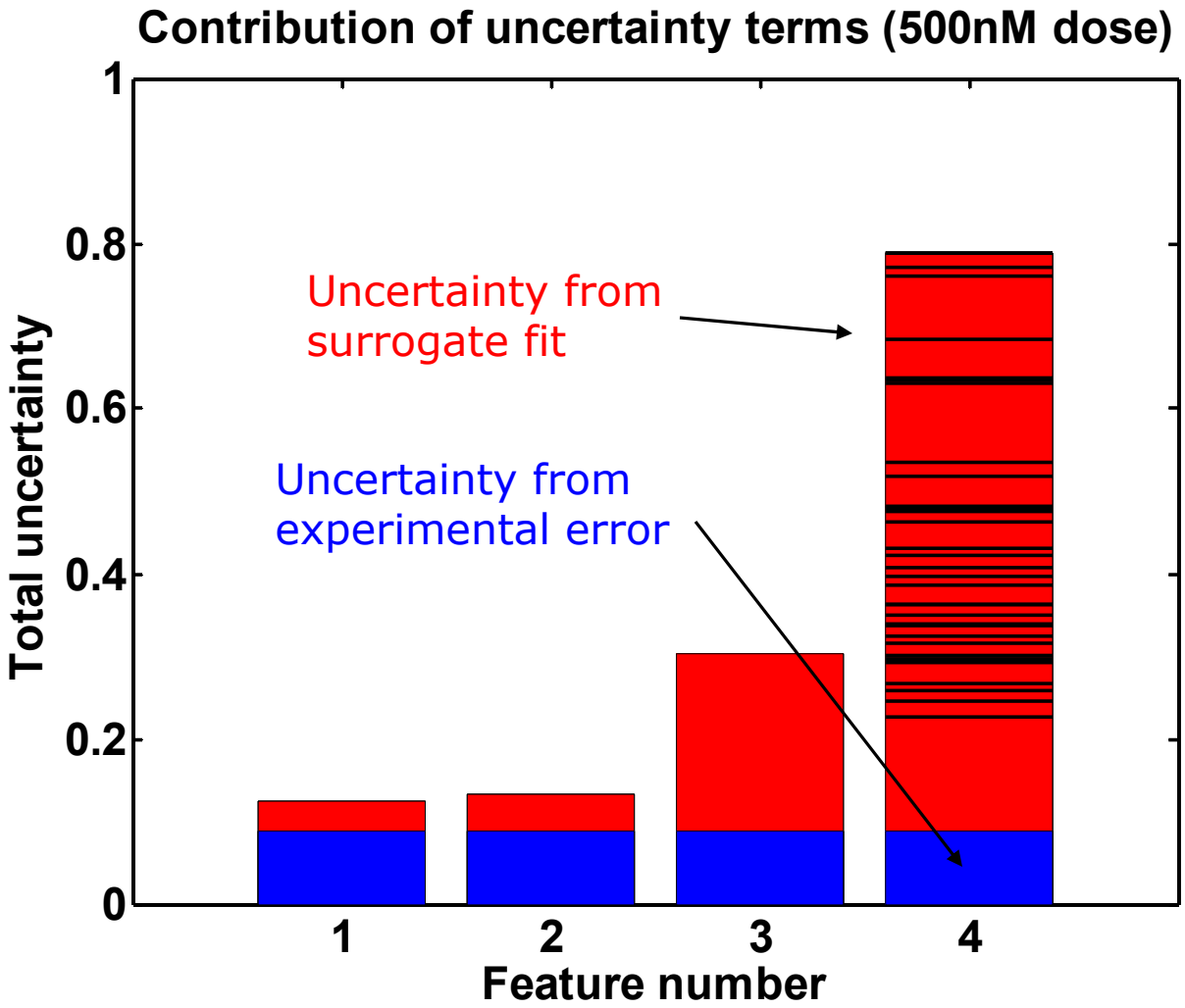




- Subdivide the cube, re-compute surrogate models and associated fitting errors for the peak concentration models

Partition and Recompute

Still large fitting errors on a few partitions



But it worked…

- In the worst of the 32 cases, C 2 [-0.15 -0.03]
- In this case, the consistency measure was largest (but still negative) on the 26^{th} cube.
- This was not the cube with the worst surrogate fitting errors