An Approach To Modeling Tracer Experiments In Metabolic Non-Steady States

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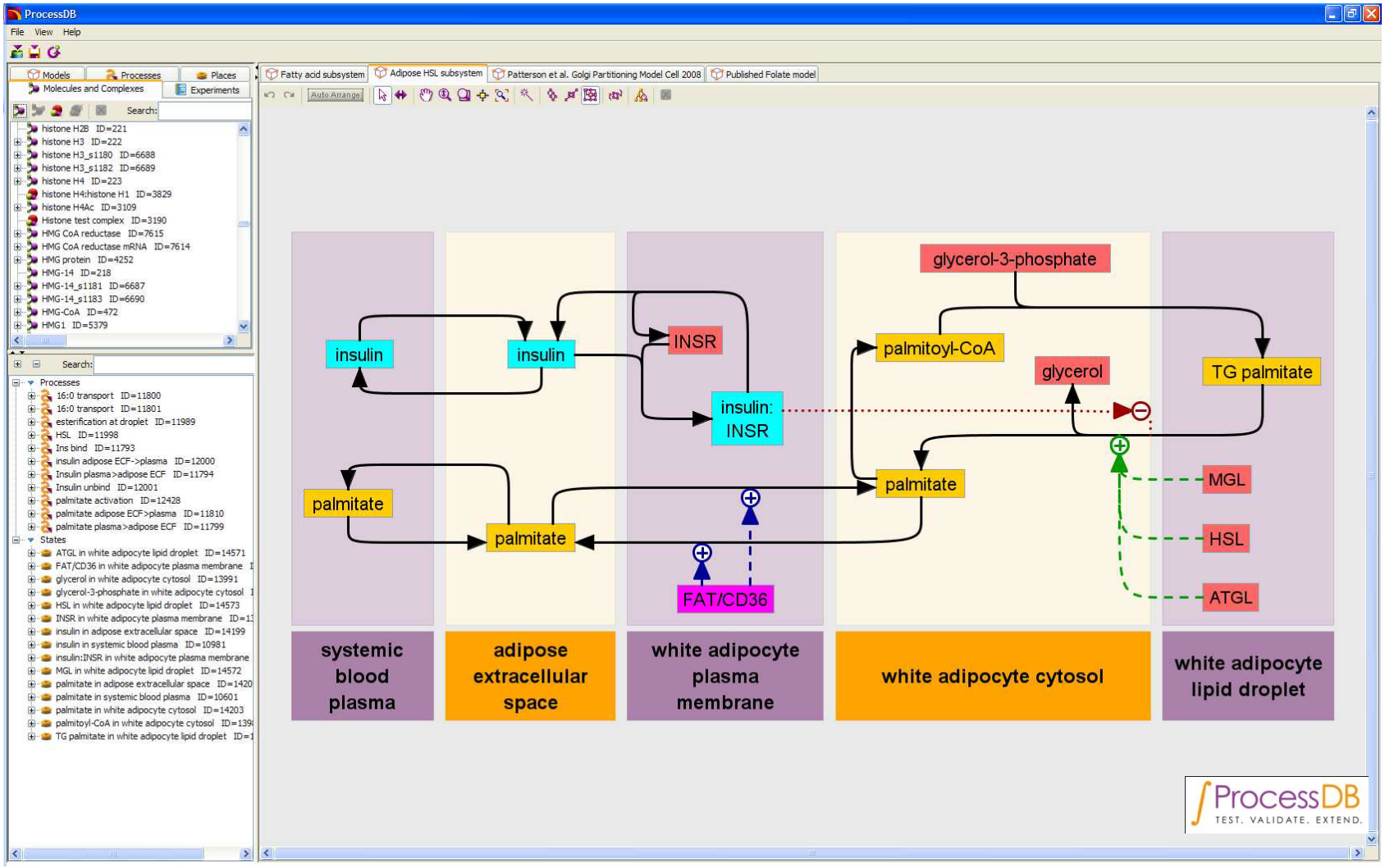
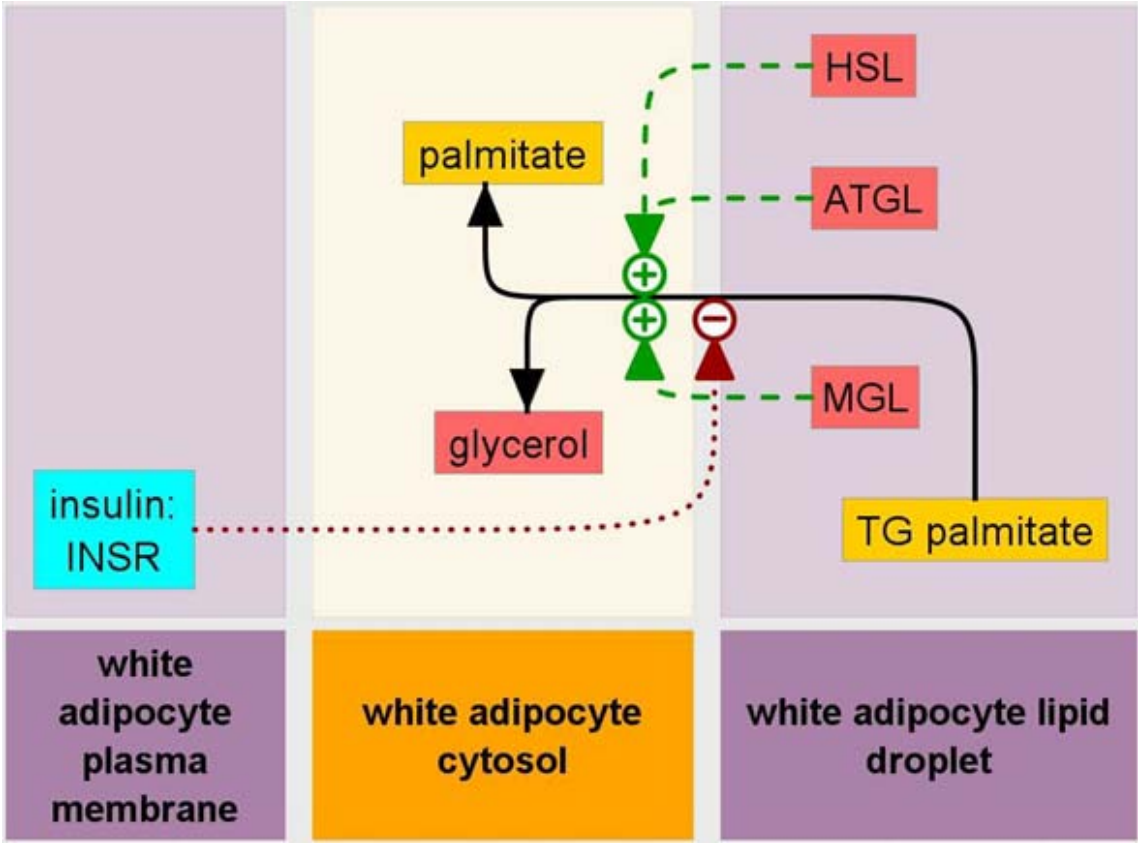
Starting point: A simple mechanistic model of insulin regulated adipose lipolysis



Adipose lipolysis is mediated by 3 enzymes

HSL is well known to be inhibited by insulin.

ATGL is probably also insulin sensitive.

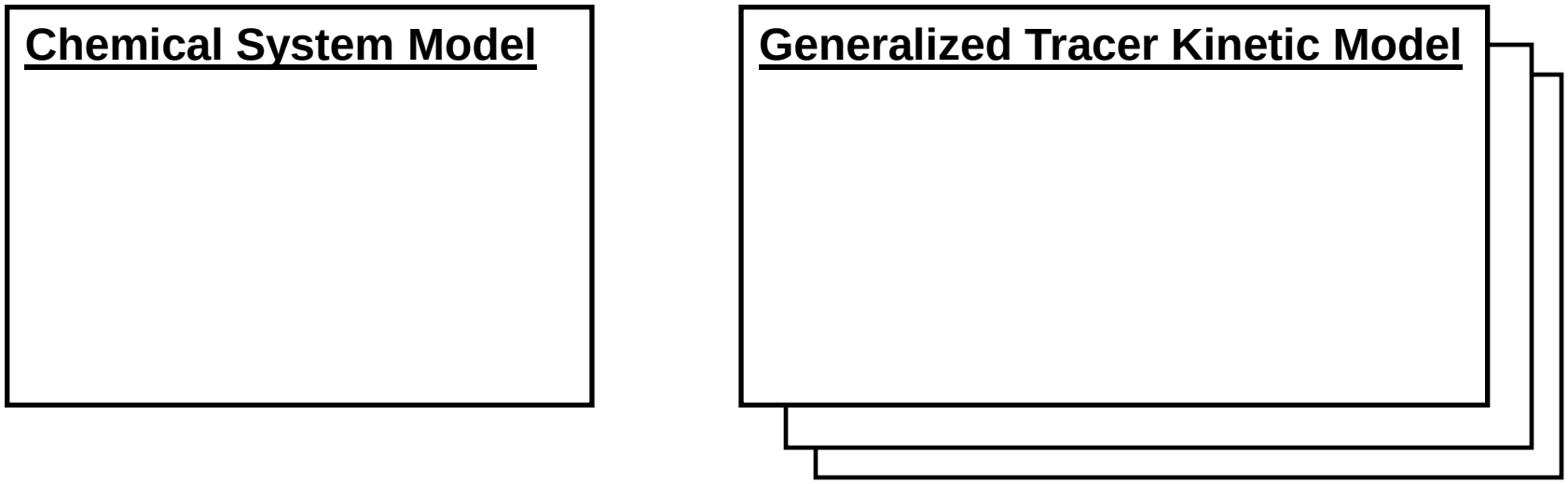


A simple rate law for lipolysis

$$P_{lipolysis} = \frac{\frac{V_{\max}^{lipolysis}}{K_{TG}}}{1 + \frac{[TGpalmitate]_{droplet}}{K_{TG}} + \frac{[Insulin:INSR]}{K_i^{insulin}}} [TGpalmitate]_{droplet}$$

Rate constant

NSSTK[1] models require two (or more) linked ODE systems



[1]NSSTK = Non-Steady State Tracer Kinetic

Chemical System Model

- is nonlinear because it includes molecular cell biology & physiology
- is mechanistic
- is dynamic
- its state variables are the sums of the tracer and traced molecules in a given state

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Generalized Tracer Kinetic Models

- have same structure as Chemical System Model, but…
- user can (and must) specify which states and processes are traced by each tracer

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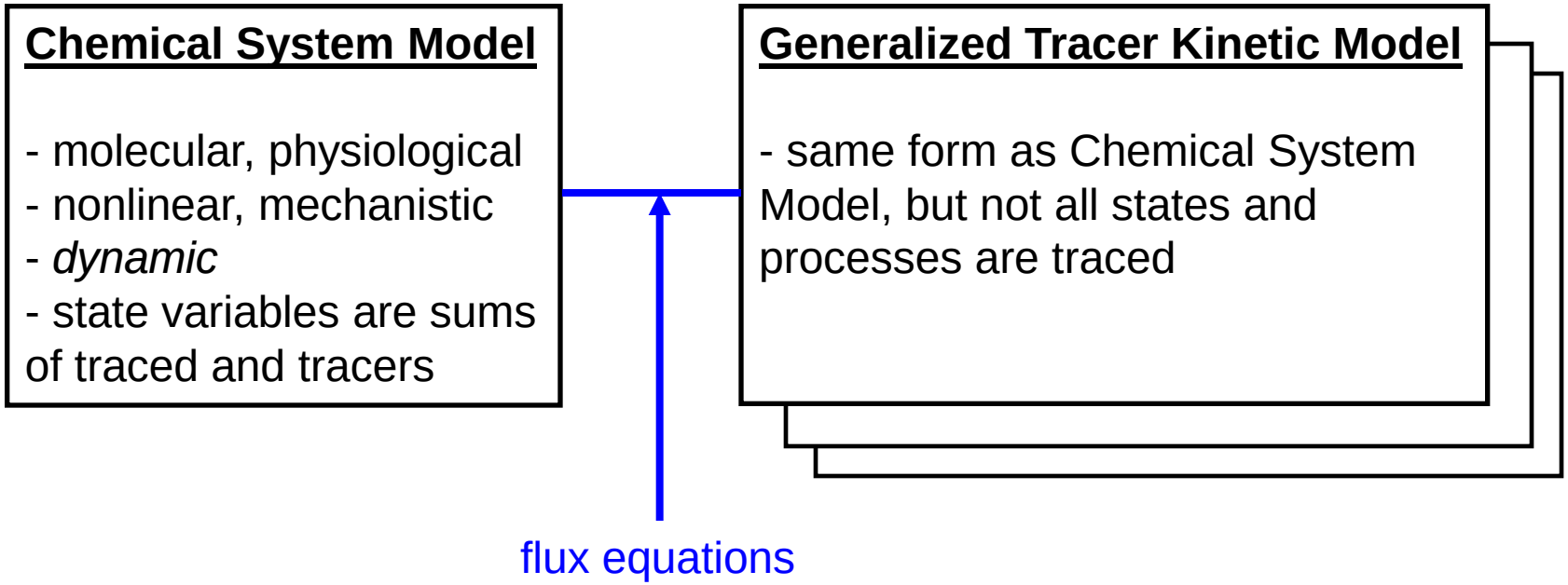
Tracer models are closely related to the chemical system model



How to calculate a tracer flux:

Start with the flux of the native molecules and apply the indistinguishability principle.

If the biology does not distinguish between tracer and traced molecule then the tracer fraction of the total flux leaving a given compartment is the same as the probability that a molecule in the compartment is a tracer.

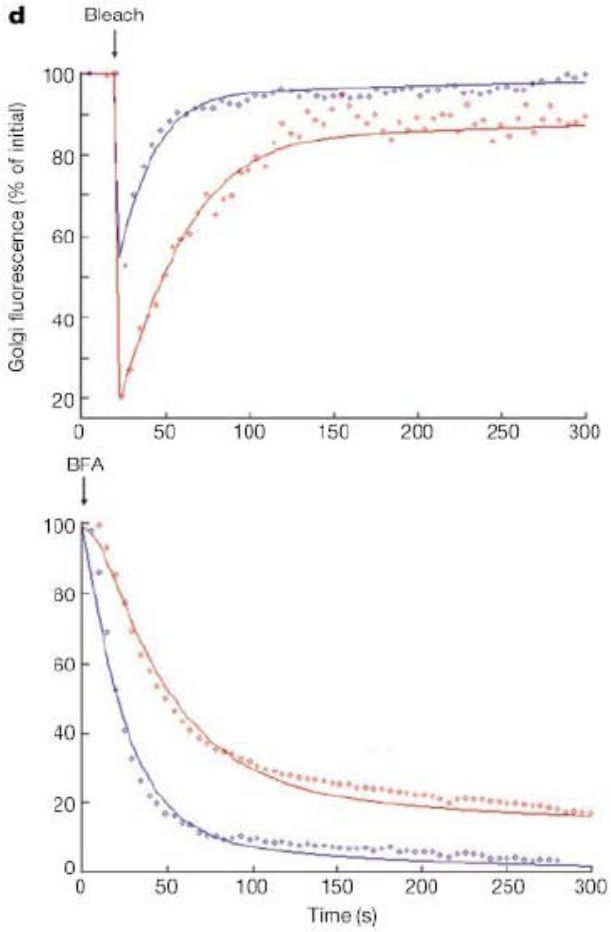
- P = process or flux
- S = state or species mass

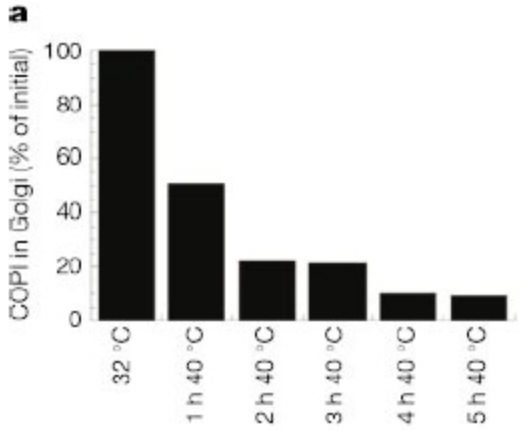
$$P^{tracer} = P^{ChemicalSystem} \frac{S^{tracer}}{S^{ChemicalSystem}}$$

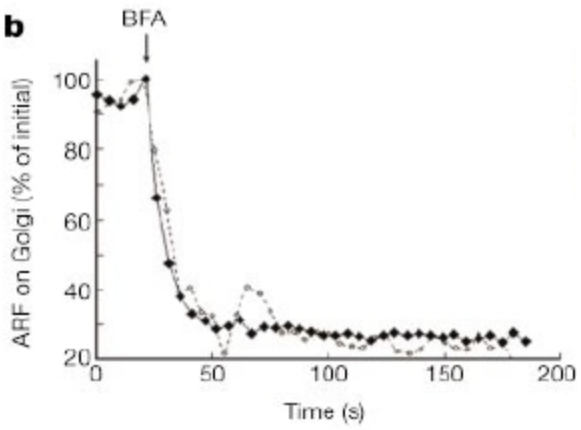
Then the tracer differential equations are written by applying conservation of mass to the P_{ji}^{tracer}.

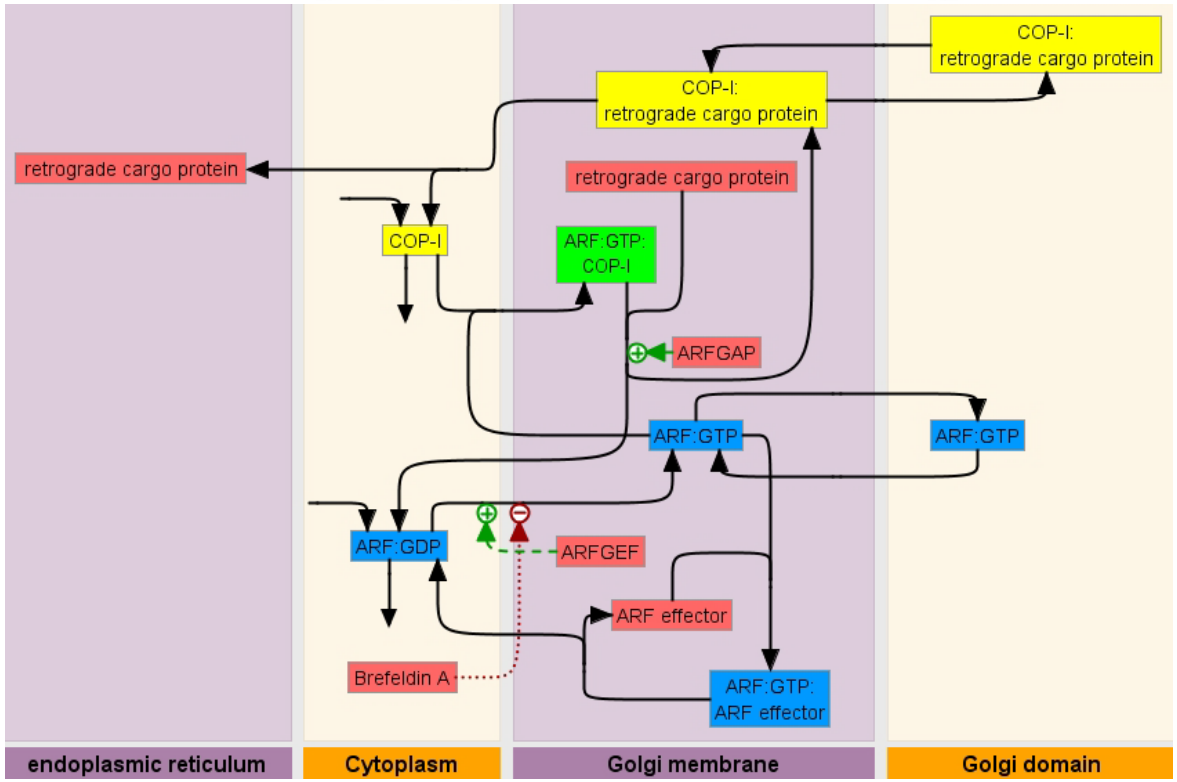
$$\frac{dS_j^{tracer}}{dt} = \sum_{i=1, i \neq j}^{n} P_{ji}^{tracer} - \sum_{i=1, i \neq j}^{n} P_{ij}^{tracer}$$

Physiological or pharmacological perturbations initiate non-steady states.









Presley JF, Ward TH, Pfeifer AC, Siggia ED, Phair RD, Lippincott-Schwartz J. ***Nature*** 2002

Other projects we've done using NSSTK

1. Patterson GH, Hirschberg K, Polishchuk RS, Gerlich D, Phair RD, Lippincott-Schwartz J.
Transport through the Golgi apparatus by rapid partitioning within a two-phase membrane system.
Cell 2008 Jun 13; 133(6): 1055-1067.

2. Kruhlak M, Crouch EE, Orlov M, Montano C, Gorski SA, Nussenzweig A, Misteli T, Phair RD, Casellas R.
The ATM repair pathway inhibits RNA polymerase I transcription in response to chromosome breaks.
Nature. **2007** Jun 7;447(7145):730-4.

3. Lin Y, Dueker SR, Follett JR, Fadel JG, Arjomand A, Schneider PD, Miller JW, Green R, Buchholz BA, Vogel JS, Phair RD, Clifford AJ.
Quantitation of in vivo human folate metabolism.
Am J Clin Nutr. **2004** Sep;80(3):680-91

4. Dundr M, Hoffmann-Rohrer U, Hu Q, Grummt I, Rothblum LI, Phair RD, Misteli T.
A kinetic framework for a mammalian RNA polymerase in vivo.
Science. **2002** Nov 22;298(5598):1623-6.

5. Presley JF, Ward TH, Pfeifer AC, Siggia ED, Phair RD, Lippincott-Schwartz J.
Dissection of COPI and Arf1 dynamics in vivo and role in Golgi membrane transport.
Nature. **2002** May 9;417(6885):187-93.

6. Phair RD, Misteli T.
High mobility of proteins in the mammalian cell nucleus.
Nature. **2000** Apr 6;404(6778):604-9.

7. Zaal KJ, Smith CL, Polishchuk RS, Altan N, Cole NB, Ellenberg J, Hirschberg K, Presley JF, Roberts TH, Siggia E, Phair RD, Lippincott-Schwartz J.
Golgi membranes are absorbed into and reemerge from the ER during mitosis.
Cell. 1999 Dec 10;99(6):589-601.

The NSSTK method has several advantages

- It permits an exact analysis of non-steady state experiments, even those involving tracers
- In steady state systems it reduces to the familiar linear ODEs
- Multiple tracer experiments can be analyzed simultaneously
- Enrichments are easily calculated
- Fluorescent protein tracers are handled in a straightforward way

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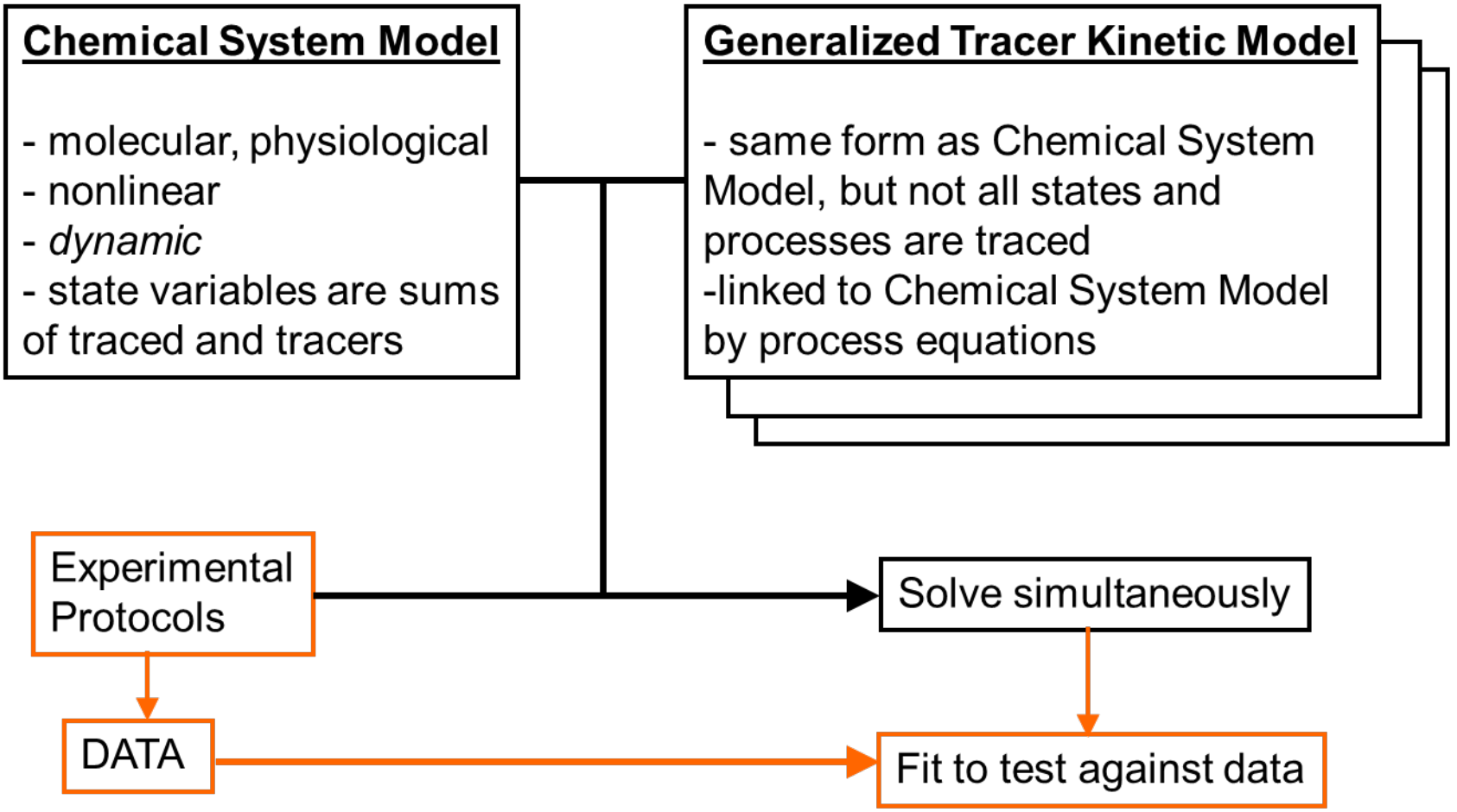
Summary of the NSSTK method



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Contact information

Our company, Integrative Bioinformatics Inc, is an independent consulting firm with strong academic roots.

We have an outstanding track record supporting high-profile research projects.

We are expensive, but our time is generally supported by jointly written NIH grant applications. Our success rate on such submissions is more than 40%.

We do not charge for our time in the grant-writing phase unless a full modeling project is required for preliminary results.

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- If you might like to add a professional modeling and software development team to your project without the usual difficulties of academic collaboration, please contact us to explore the possibilities.