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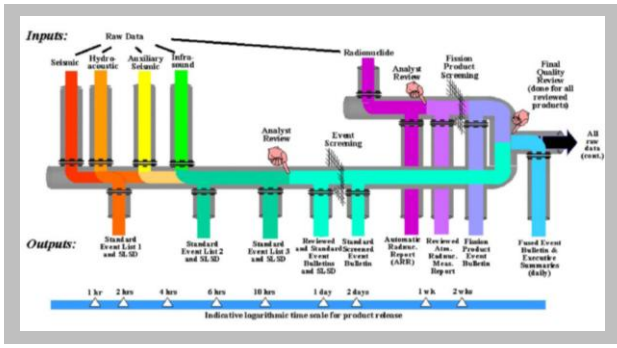


# System Dynamic Modeling to Predict Virus-Host Interactions


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U.S. DEPARTMENT OF ENERGY

National Nuclear Security Administration


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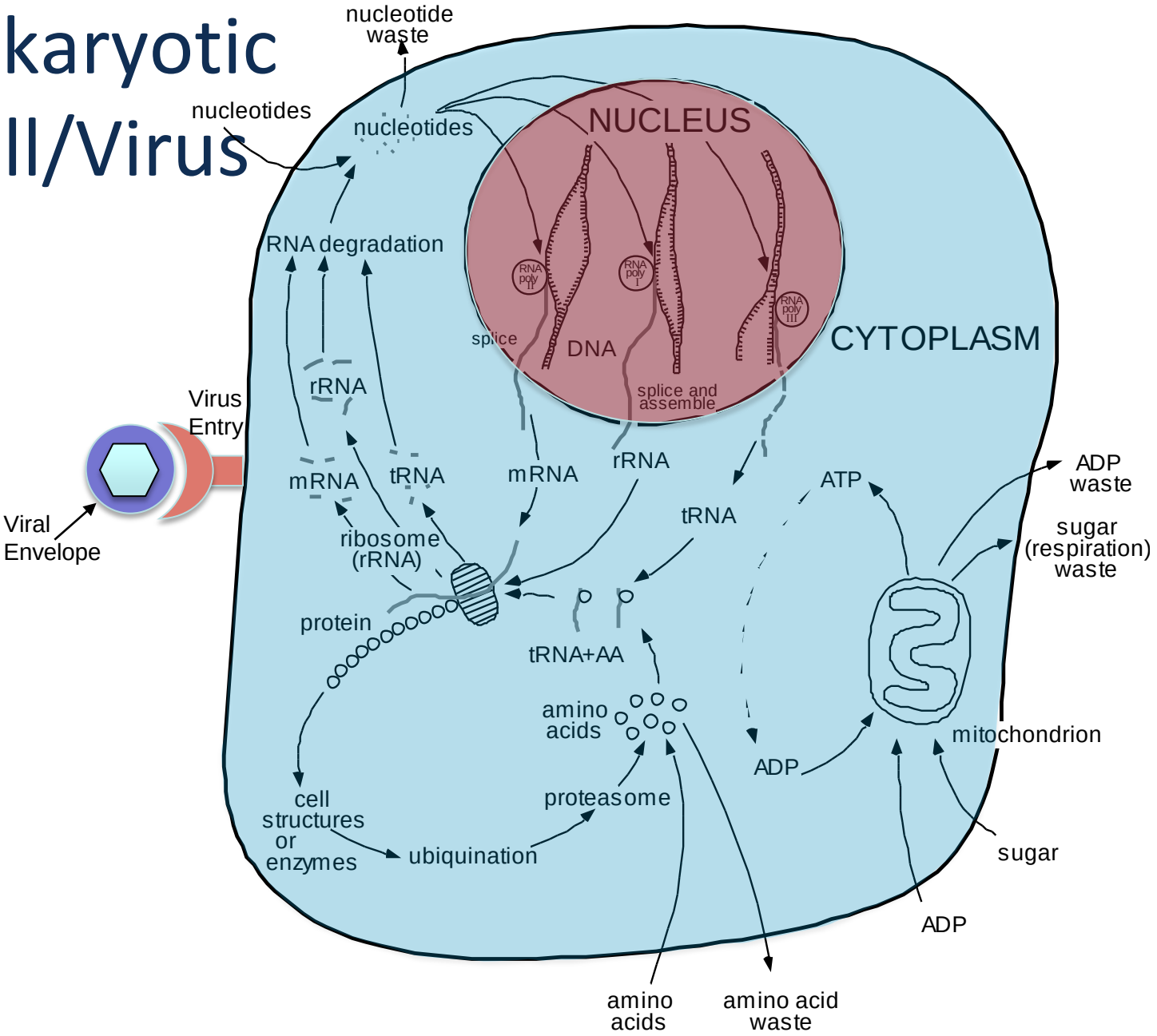
# Eukaryotic Cell/Virus



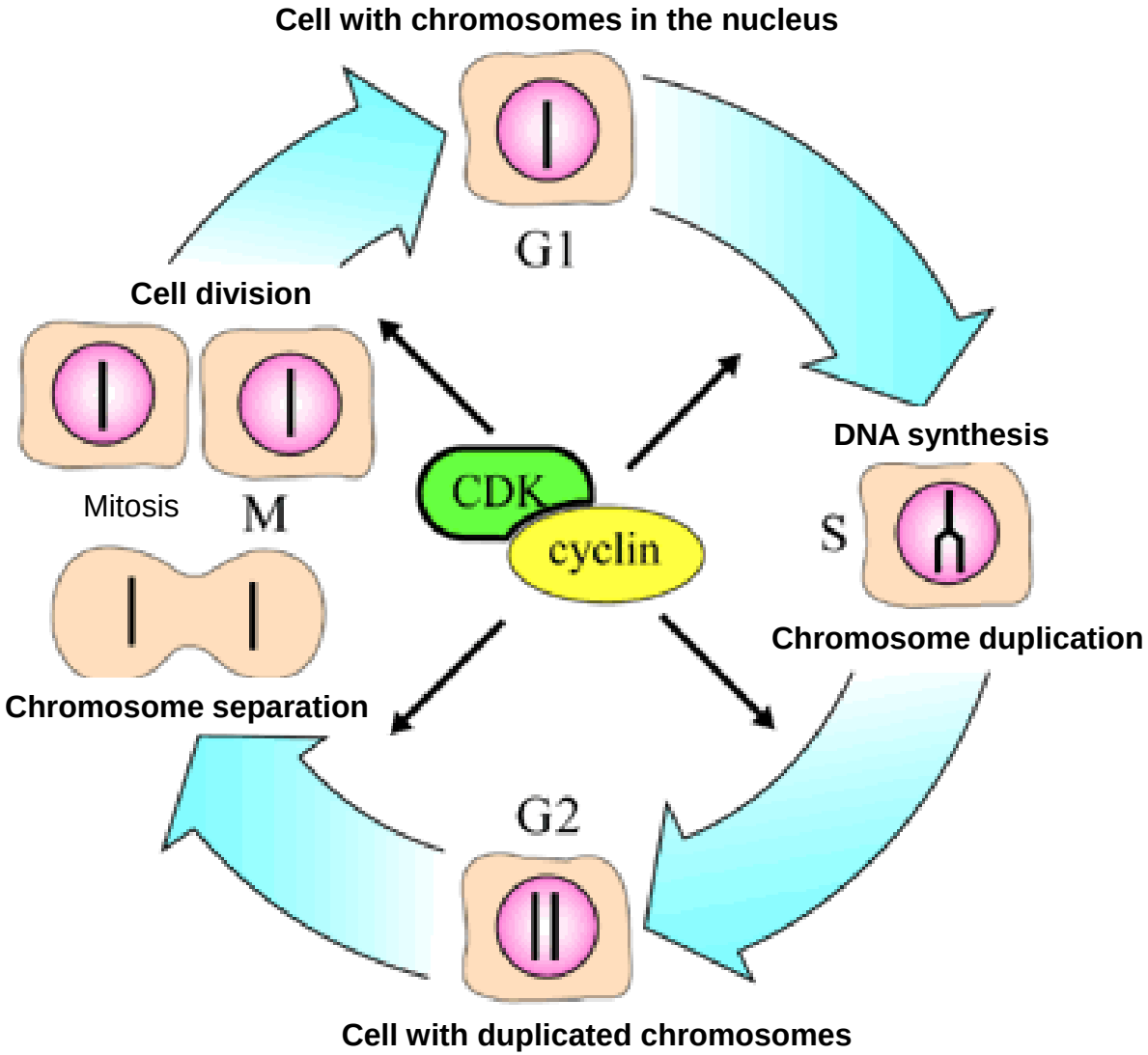
# Problem

- To understand complex viral pathways and their interaction with cells, we need mathematical models

- Most cell-virus modeling is handcrafted—no means to capture the necessarily increasing scope and detail
  - How do we deal with the problem of complexity?

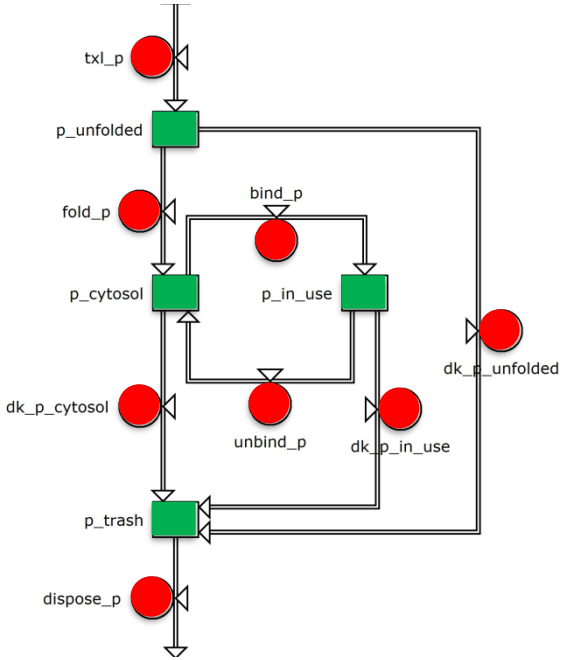
- Why Sandia?
  - capabilities in math, Mod/Sim, computation and systems analysis, + National Security

# The Cell Cycle




From: http://nobelprize.org/nobel_prizes/medicine/laureates/2001/eng.gif

# How to Build a Cell-Cycle Model



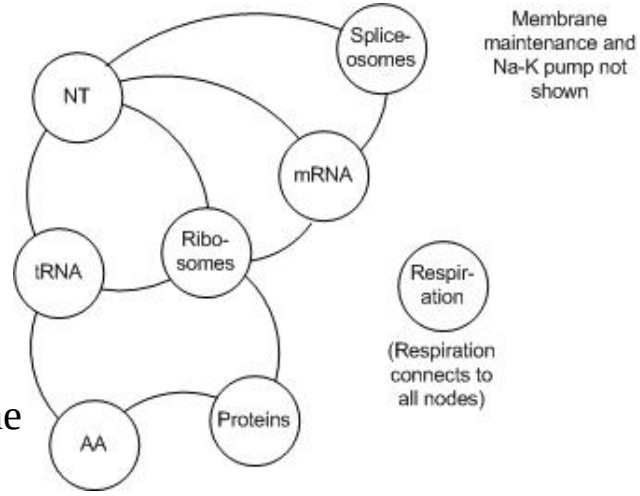

Use ***templates*** based on System Dynamics to describe molecule lifecycles.

squares represent states, ODEs typically of the form:

$$\frac{dC}{dt} = R_{in} - R_{out}$$

circles represent rates, typically of the form:

$$R = kC_1C_2\ldots$$




**Connect the templates using a relationship diagram.** (Typical modeling to date has been only of these networks without the consideration of the complete molecule lifecycles)

# Basic Cellular Machinery

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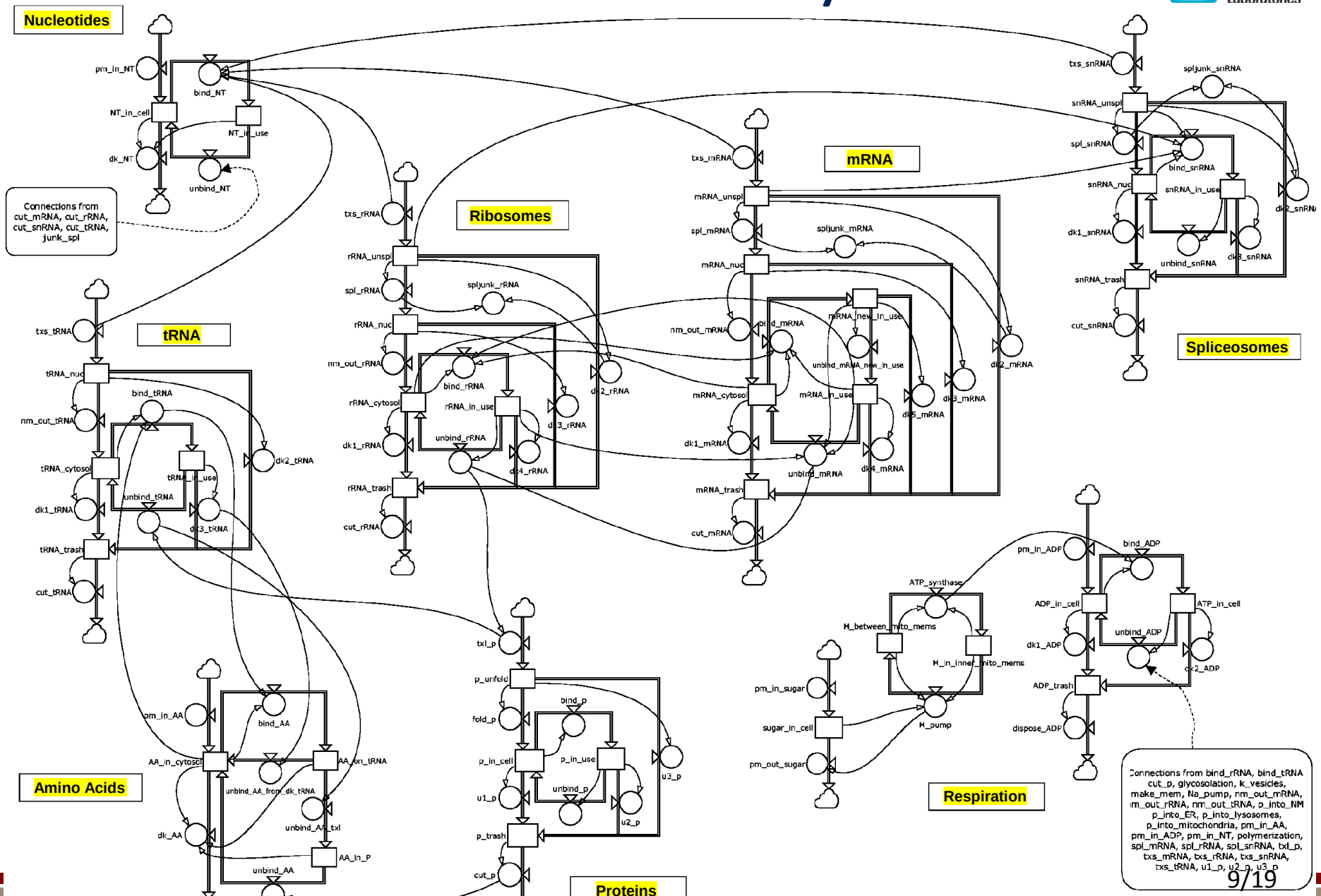

# How to Build a Cell-Cycle Model

Constrain the model (i.e., adjust parameter values so that the model results reflect observations)

- $10^9$ ATP, $10^7$ ATP/s, $10^{10}$ proteins, $5 \times 10^6$ rRNA, $3 \times 10^4$ RNA pol, 30 NT/RNApol/s, 50 NT/DNApol/s etc.
- HeLa cells divide in 1-1.5 days—G1 takes ~10 hr, S takes ~8 hr, G2 phase takes ~6 hr, M takes ~4 hr
- Other data can be inferred—e.g. 8-hr S phase needs ~5000 DNApol
- Concentration data for specific proteins usually ***not*** available! (Concentration and reaction rates can be adjusted together when data are available without significantly affecting results)
- Initial cell cycle when exiting G0 must be similar to continuous cell cycling, using same set of reaction rates

# How to Build a Cell-Cycle Model

## 428 Coupled ODEs

- Standard template used as basis for each molecule to track its lifecycle
- 41 ODEs in the base model track bulk AA, NT, ATP, RNAs, and proteins
- 387 ODEs track 33 cell-cycle specific proteins/complexes and mRNAs and DNA
- Stiff equation solver (ode15, in Matlab), as reaction timescales range from microseconds to months.
- With ode15, we solve for 10 days in less than an hour on a PC

# Proteins in Cell Cycle

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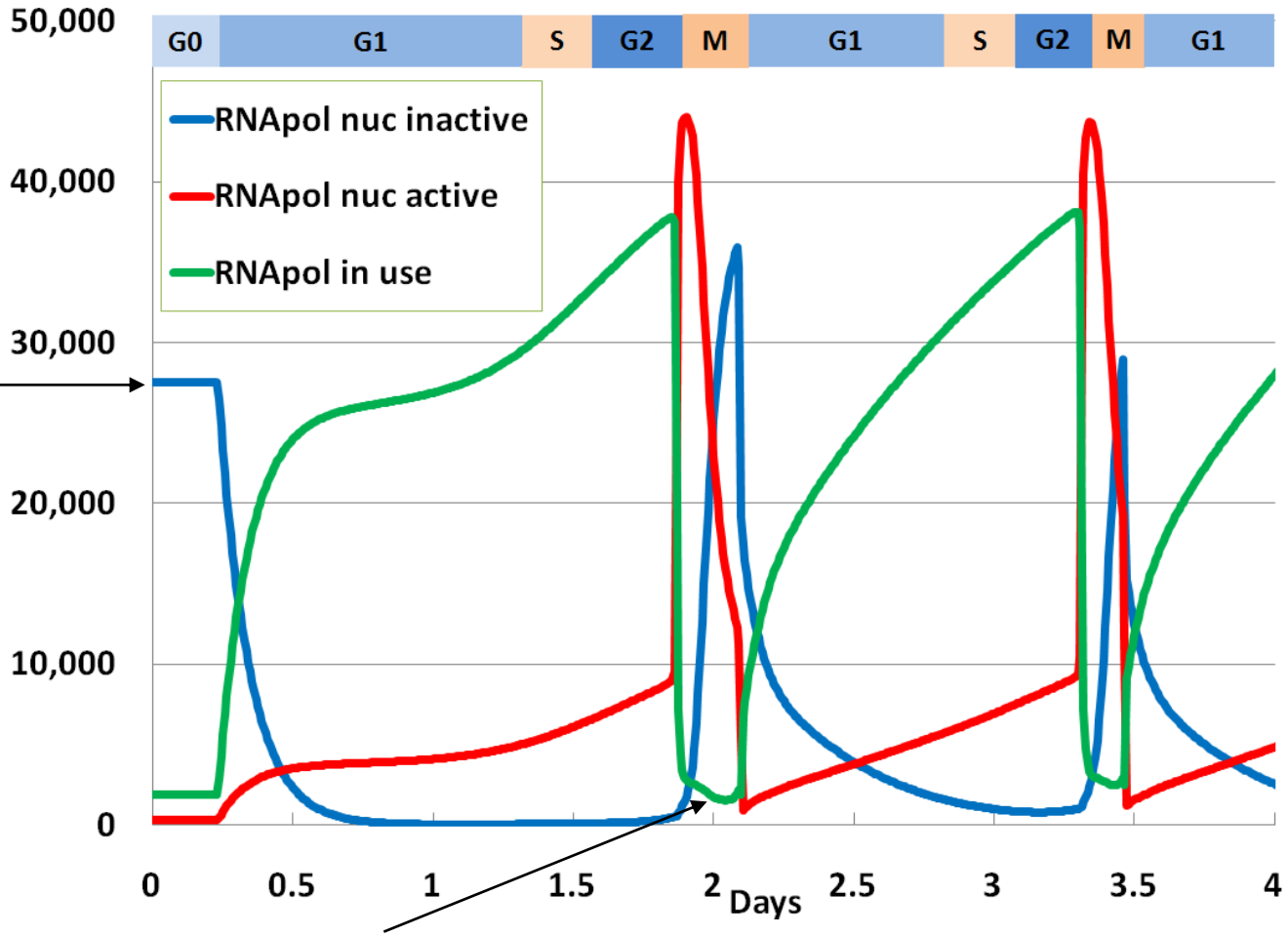
| | |
|---|---|
| p27 | inhibits cyclins--prevent S |
| Rb | inhibits E2F |
| cycE | activates Cdk2 kinase |
| NF-Y | TF for cycA, cycB, Cdk1, Cdc25C, TF-gro |
| cycA | activates Cdk2 and Cdk1 kinases |
| SCF | ubiq (requires subunit) |
| DNA poly | DNA polymerase |
| Wee1 | (Myt1) kinase-prevent Cdk1 activation |
| cycB | activate Cdk1 kinase |
| cycD | activates Cdk4or6 kinase; inactivates APC(Cdh1) |
| Cdk2 | kinase--phosphorylates RC--allows DNA replication |
| TF-grow | Transcription Factors (eg, c-myc, c-Jun, Notch) |
| RC | DNA replication complex (eg, hORC1, hCdc6) |
| B-Myb | TF for cycD, Cdk1, Plk1, DNA pol, B-Myb, TF-grow, others |
| E2F | TF for cycE, cycA, Rb, Cdk1, Cdk2, E2F, Cdk25A, DNA pol, others |
| Skp2 | ubiq subunit for p27, E2F, RC, TF-grow, B-Myb, free cycE, cycA, cycD |
| Btrc | ubiq subunit for Emi1, Cdc25A (sometimes), Wee1, others |
| Plk1 | kinase--activates Cdc25C; deacts Emi1, Wee1; translocs cycA/Cdk1, cycB/Cdk1, Cdc25C |
| Cdh1 | ubiq subunit--maintains G0, G1--ubiq Cdc20, cycA (free), cycB (free), Cdc25A, RC, Plk1 |
| Cdc20 | ubiq subunit for Securin, cycB, cycA |
| Cdc14 | phosphatase--ends M--activates p27, Wee1, Cdh1; deacts Cdc25A, Cdc25B, Cdc25C |
| Cdc25A | phosphatase--activates cycE/Cdk2, cycA/Cdk2, cycA/Cdk1*, cycB/Cdk1* |
| Cdc25B | phosphatase--activates cycA/Cdk1, cycB/Cdk1* |
| Securin | keeps separase from destroying chromotin cohesion proteins |

| | |
|---|---|
| Cdk1 | kinase--activates APC(Cdc20) |
| Cdc25C | phosphatase--activates cycB/Cdk1 |
| Emi1 | inhibits Cdh1, Cdc20 |
| APC | ubiquitinase (requires subunit) |
| cycC | inhibits RNA pol |
| KPC | ubiq for p27 |
| RNA poly | transcription |
| eIF-4 | initiates translation |
| Fbw7 | ubiq subunit for cycE, TF-grow, RC |

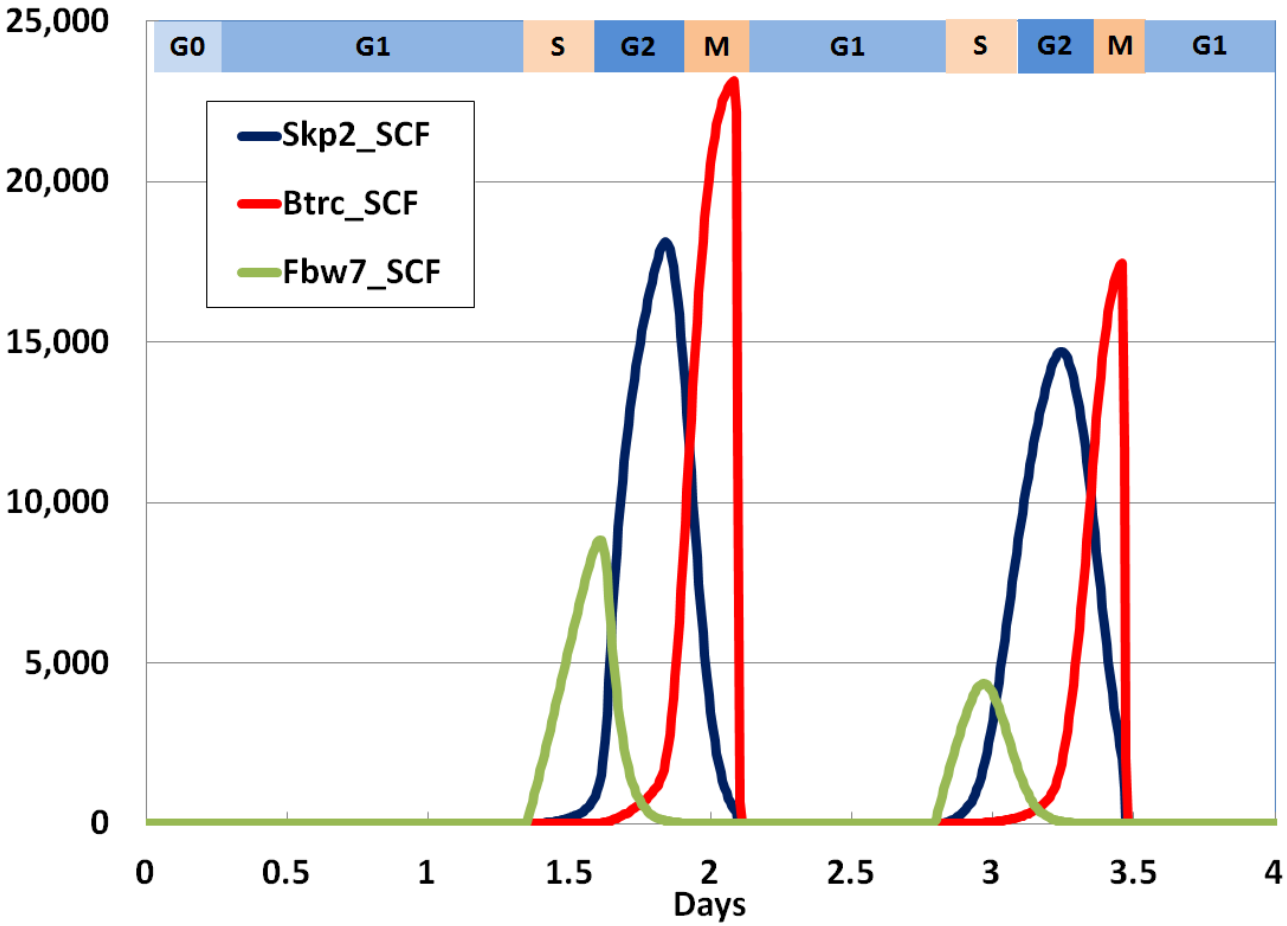
# Some Results

A large pool of RNA poly must be available immediately




Not all RNA polymerase dissociate from DNA during mitosis

# SCF bound to Fbw7, Skp2, Btrc




All present at same time SCF:

- binds Fbw7
- ubiquitinates Fbw7 substrates,
- Fbw7 autoubiquitinates,
- binds Skp2
- ubiquitinates Skp2 substrates (esp. RC)
- Skp2 autoubiquitinates,
- binds Btrc

# Virus-Cell Link

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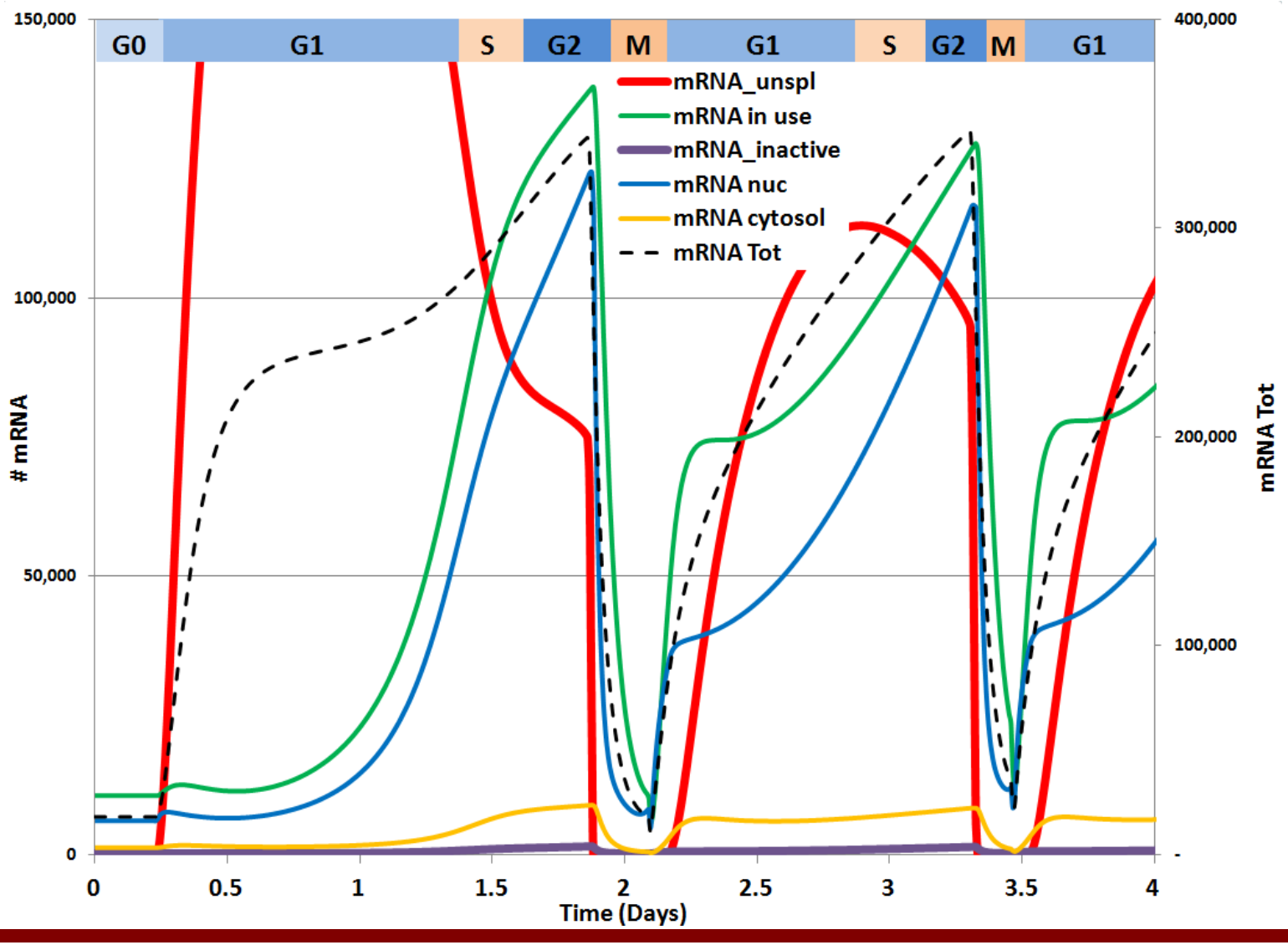
**Entry via**

- **Membrane Fusion -**
  - Examples: HIV, Herpes simplex virus or influenza.
- **Endocytosis -**
  - Examples: poliovirus, Hepatitis C virus and Foot-and-mouth disease virus.
- **Genetic Injection -**
  - Example: the phages.
- Once a virus enters a cell, replication is not immediate (seconds to hours).
- A series of molecular pathways are initiated dependant on virus or VLP cargo.

# mRNA

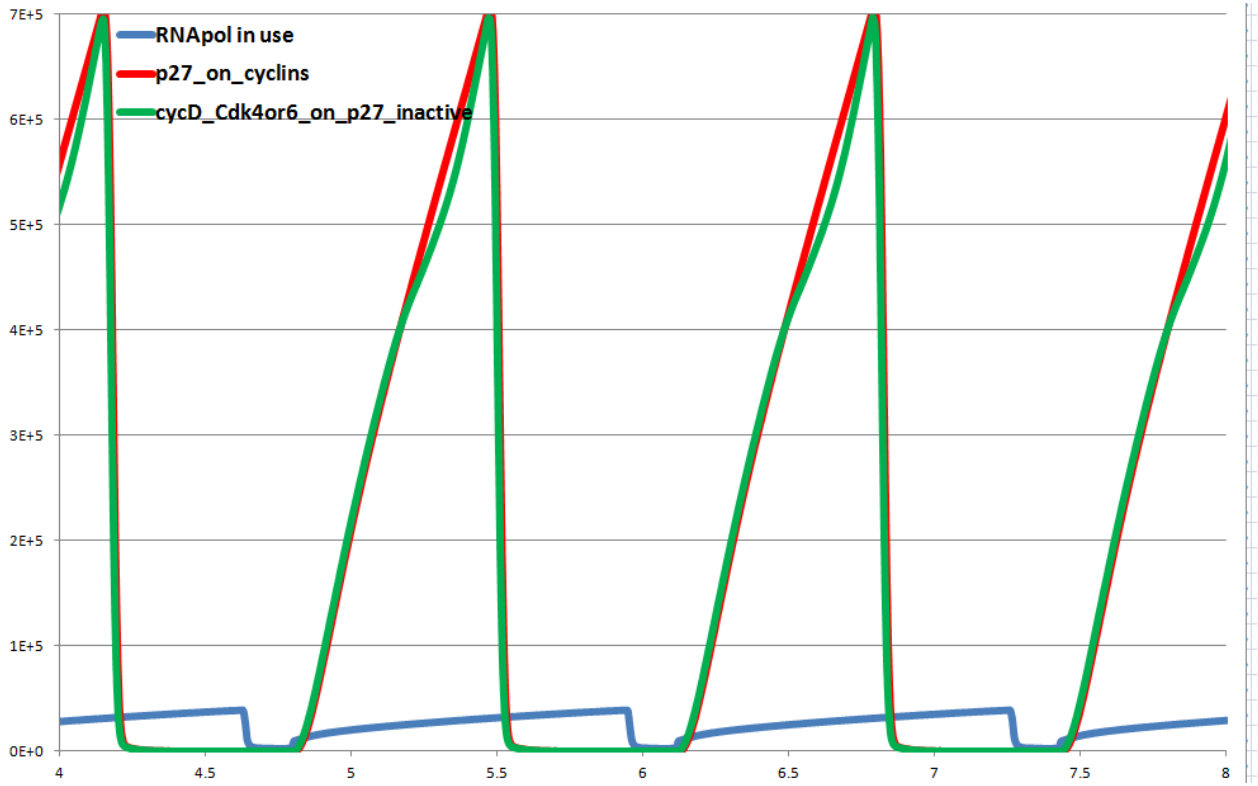

G0
G1
S
G2
M
G1
S
G2
M
G1
mRNA_unspl
mRNA in use
mRNA_inactive
mRNA nuc
mRNA cytosol
mRNA Tot
150,000
100,000
50,000
0
400,000
300,000
200,000
100,000
-
# mRNA
mRNA Tot
0
0.5
1
1.5
2
2.5
3
3.5
4
Time (Days)

# Testable Viral Entry Hypothesis

**How does Viral-Entry impact RNA-poly concentration & activity?**

Bethal (2006) found that Rifamycin binds the beta subunit of RNA polymerase, thereby blocking mRNA synthesis. Antiviral compounds inhibit the transcription of DNA by several mechanisms or by inhibition of viral entry into host cells.

There are several effects: (1) mitogen deactivates cycC/Cdk8, thus releasing inhibition of RNA polymerase; (2) mitogen activates KPC (not shown), which ubiquitinates p27; and (3) mitogen allows transcription of several proteins, including cycD



# Some Predictions

## Molecular level

- p27 is important for maintaining G0 and timing duration of G1 in the first cell cycle after G0.
  - does not have a significant role in timing G1 in continuous cell cycling.
  - G1 duration in subsequent cell cycles is timed by APC(Cdh1).
- p27 does not inhibit cycD/Cdk4 or cycD/Cdk6 activity
- Not all RNA polymerase are removed from condensed DNA during mitosis; the number attached ~the number active during G0.

## System level

- When redundant pathways can activate a time critical process, one preferred pathway must inactivate the others.
  - Important in considering knockout experiments
- No explicit pre-translation mRNA regulation is needed
  - other than splicing or the number of RNA polymerase
- In G0, a supply of inactive RNA poly must be present in cells capable of dividing

# What is New and Unique

- Use of System Dynamics techniques to deal with arbitrarily complex cellular pathways and their interactions
- The modeling framework uses 2 constructs
  - *Basic-cellular-machinery construct*
  - *Molecule-lifecycle construct*
- Features
  - Molecules tracked from birth to death (a necessary step toward rigor)
  - RNAs modeled
  - Compartmentalization modeled
  - Internal consistency maintained—allows calibration to cellular-scale observations (mitigates data needs)
- Useful predictions are developed

# Conclusions

- We can handle large complex bio pathways. (will be exciting to port bigger models to massively parallel machine!)

- Proposed applications:
  - Basic biology research
  - Bio-threat reduction
  - Disease understanding
  - Radiation-effects research

- Future: epigenetics (enzymes that control DNA exposure and gene expression), receptor-to-expression cascades involved in cancer and t-cell functioning.