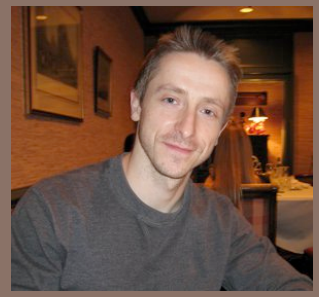
TO LYSE OR NOT TO LYSE: TRANSIENT-MEDIATED STOCHASTIC FATE DETERMINATION IN CELLS INFECTED BY BACTERIOPHAGES

w/Richard Joh (GA Tech, left), Yuriy Mileyko (GA Tech/Duke, right) & others



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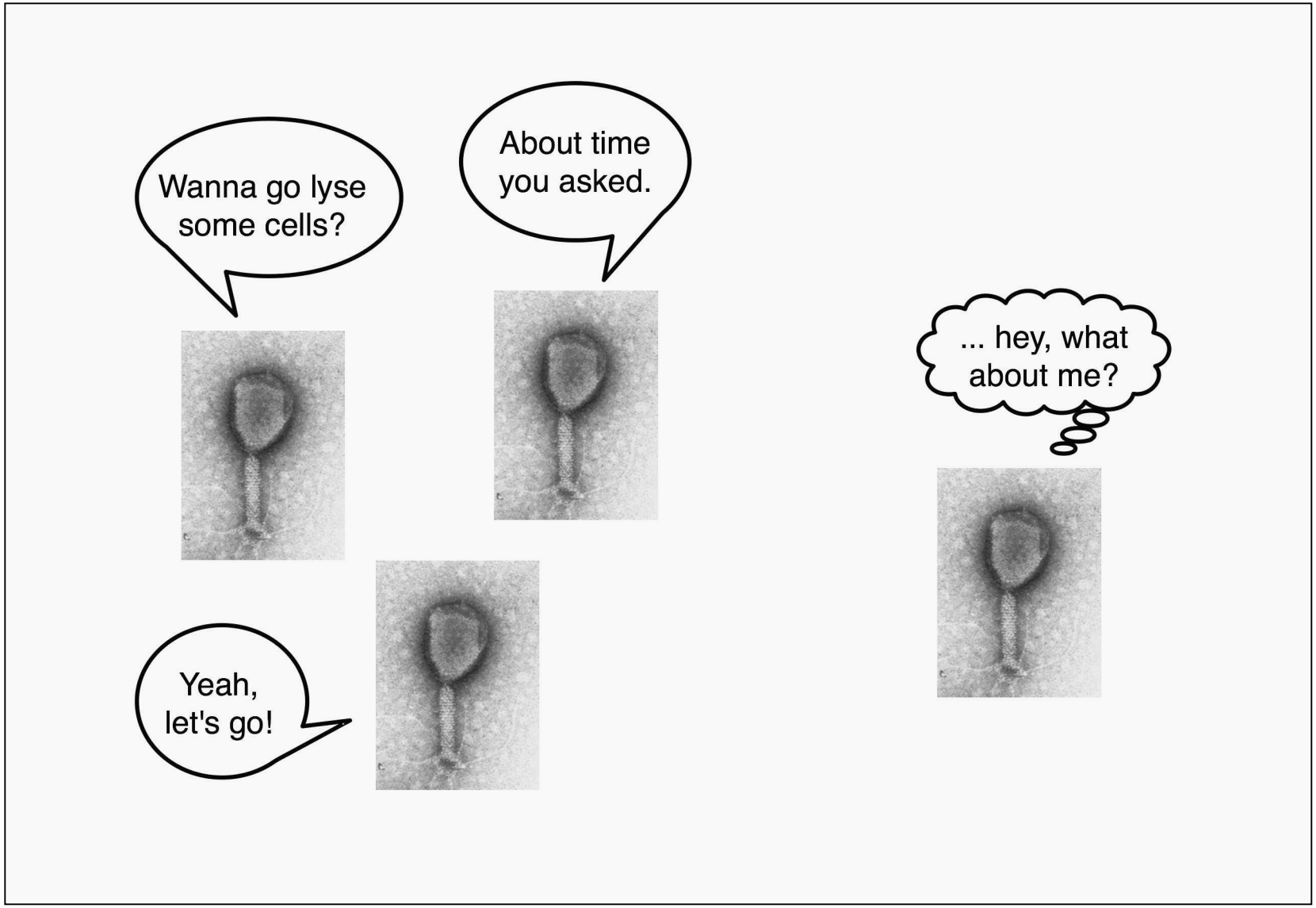
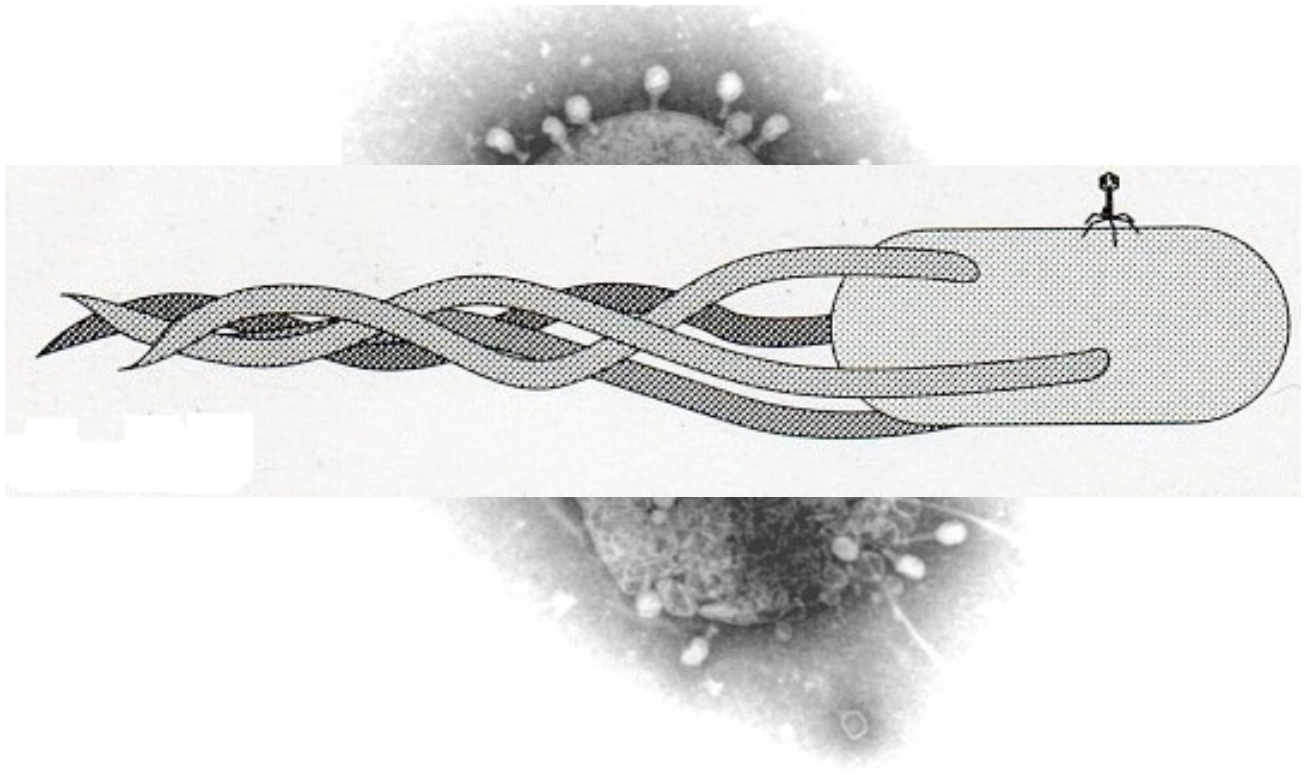
Email: jsweitz@gatech.edu,
Web: http://ecotheory.biology.gatech.edu, KITP, March 1, 2011


Wanna go lyse some cells?
About time you asked.
... hey, what about me?
Yeah, let's go!



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Central Questions in Today's Talk

1) Does co-infection alter cell fate, and if so, in what way?

   Yes, lysis and lysogency depend on co-infection

2) How can viral genomes interact intracellularly to collectively determine cell fate?

   Coupling of transcription with viral protein pool

3) How can a simple model of fate determination be reconciled with observed single-cell level data?

   Stochastic dynamics & a gene dosage compensation mechanism provide an alternative explanation for cell fate determination in co-infected hosts

Experimental work: Kourilsky (Mol Ge. Genet, 1973), Kobiler et al. (PNAS, 2005), St. Pierre and Endy (PNAS, 2008), Zeng et al. (Cell 2010)

Theoretical work: Weitz et al. (Biophys J., 2008), Mileyko et al. (PNAS 2008), Joh & Weitz (PLoS Comp Biol, in press)

From intracellular mechanisms to traits

Ecological Model

$$\frac{dV}{dt} = \overbrace{\beta\phi NV}^{\textit{Phage production}} - \overbrace{mV}^{\textit{Phage decay}}$$

Trait space

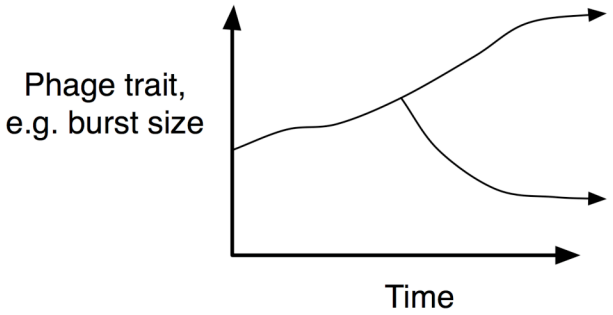
$$\{..., \beta_1, \beta_2, \beta_3, ...\}$$

Biophysics

&

Gene regulation

Evolutionary model

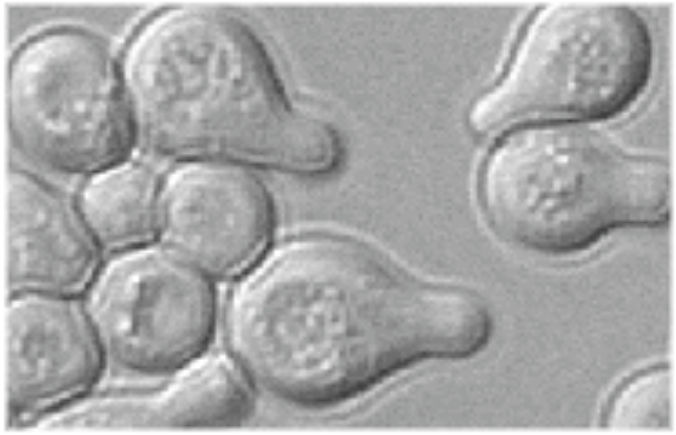


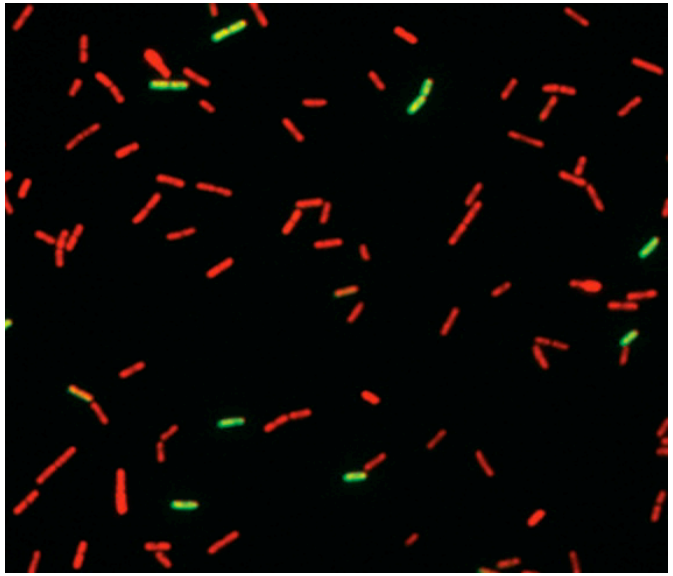
Outline

- Determination of alternative cell fates
- Quantitative model of lysis-lysgoeny decisions
- Heterogeneity of decisions: gene dosage effect on lysis-lysogeny

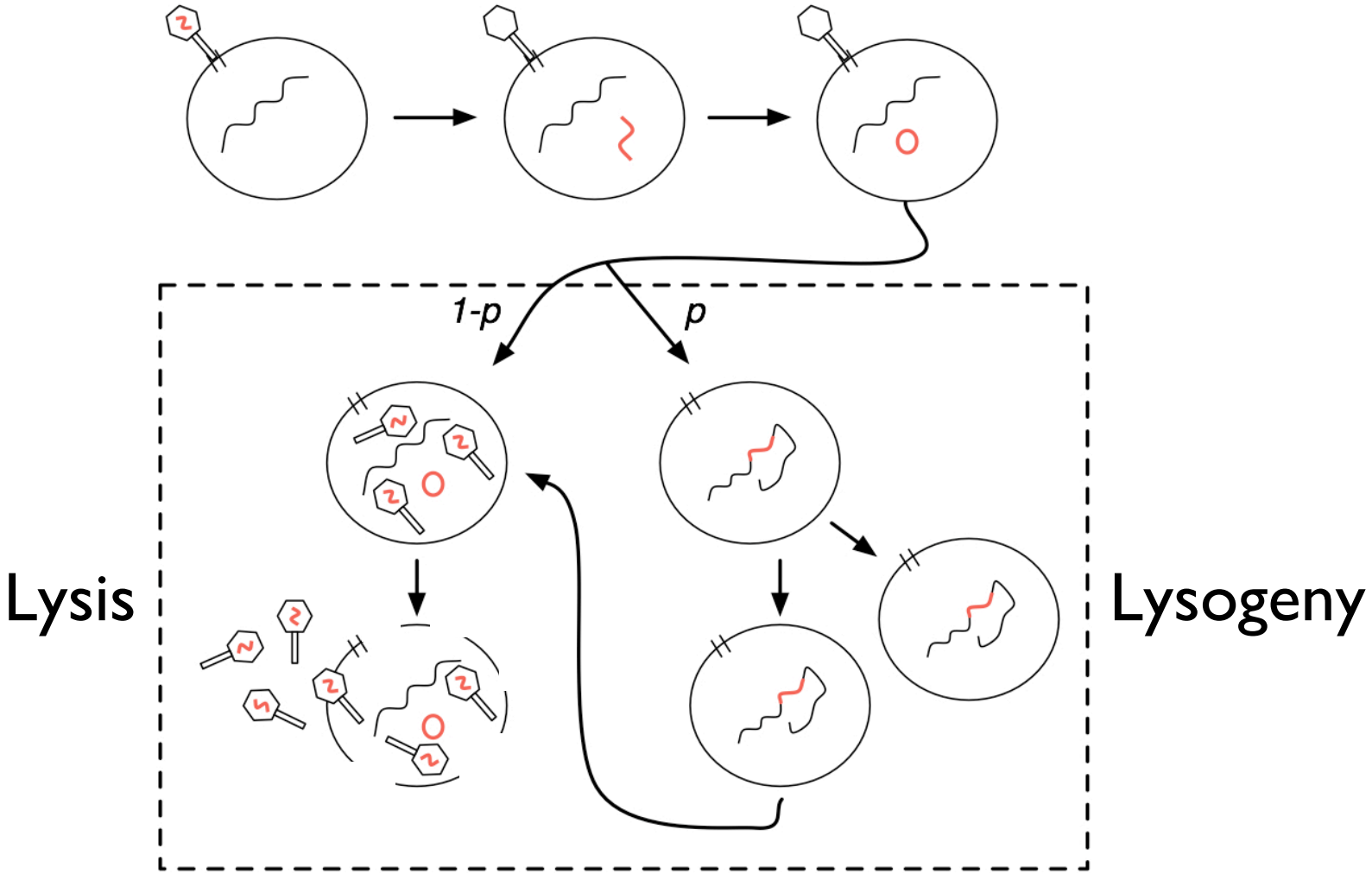
Stochastic cell fates in unicellular organisms

- Eukaryotes
  - *S. cerevisiae* mating
- Bacteria
  - Bacterial persistence to antibiotics
  - Competence: *Bacillus subtilis*
- Viruses?

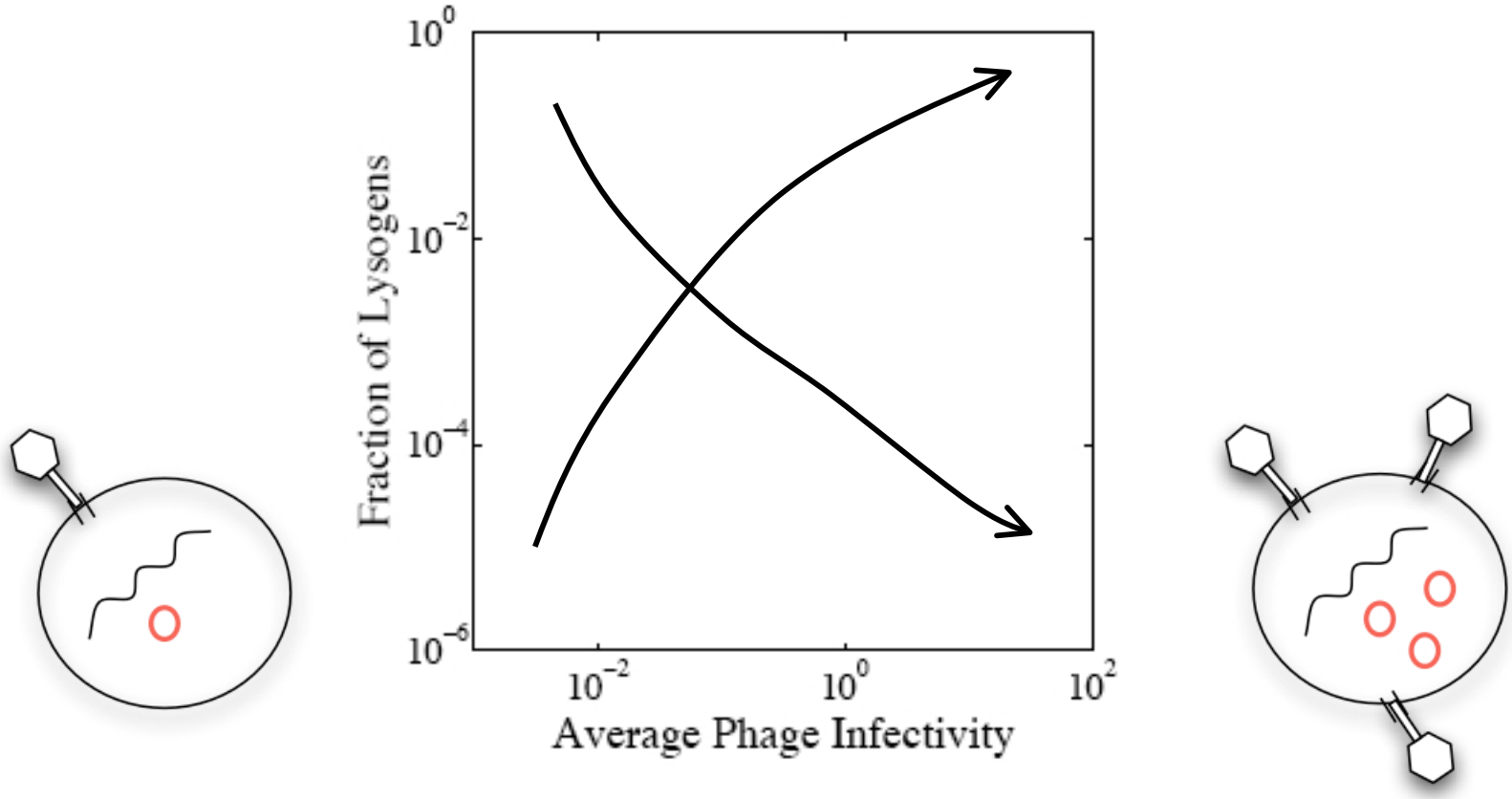




Viruses also drive alternative cell fates: lysis or lysogeny

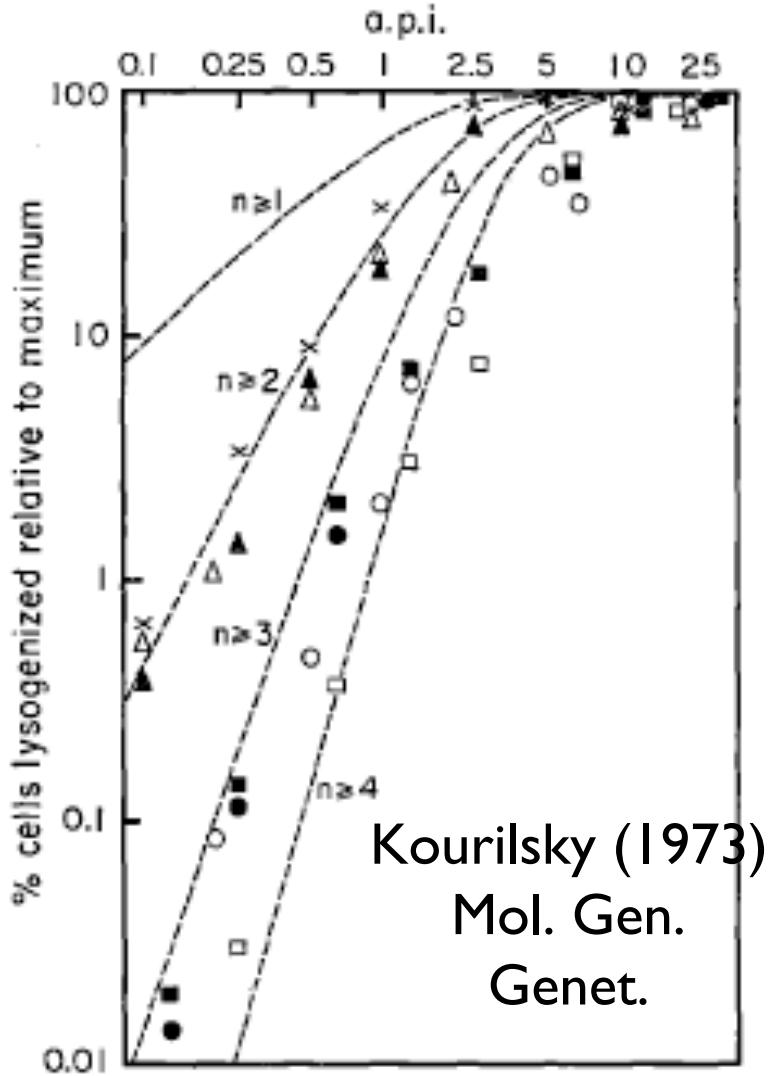


Strategies and the Cellular Multiplicity of Infection



Which of these is what phages really do?

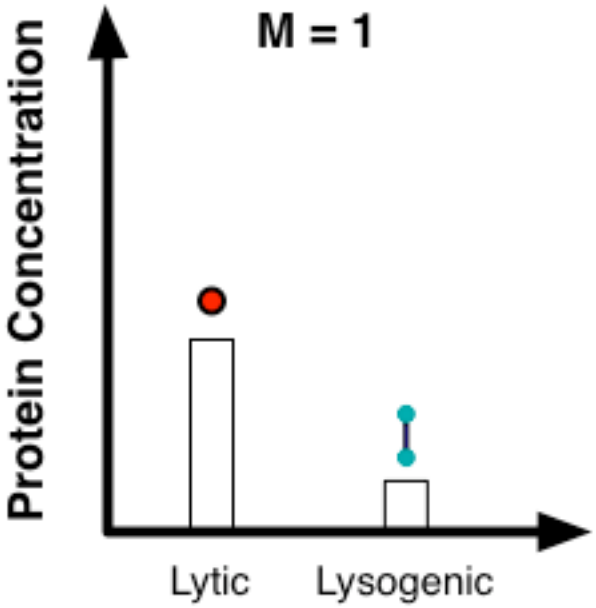
Chance of lysogeny critically depends on cellular multiplicity of infection (MOI)

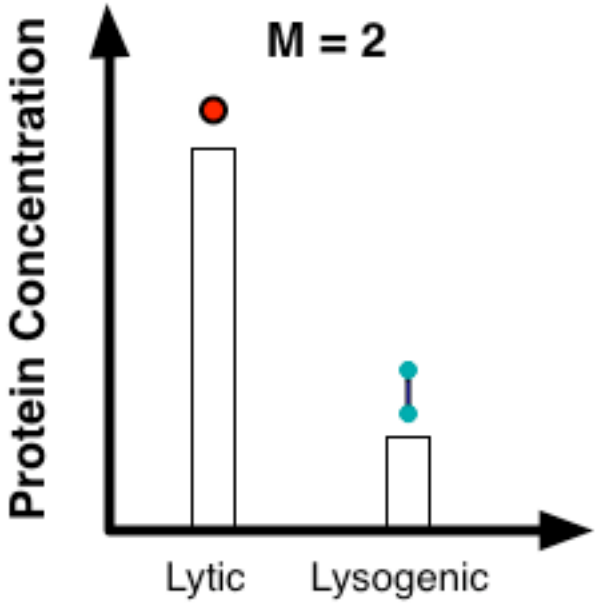


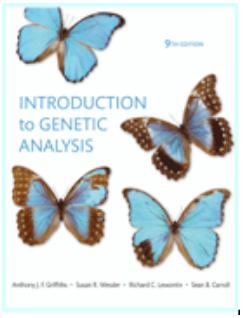
Kourilsky (1973) Mol. Gen. Genet.

- API = # phage / # host
- Lysogeny is more likely when a host cell is multiply infected

Intuition from Classic Gene Dosage Expectation: Expression is Linearly Related to Viral Copy Number



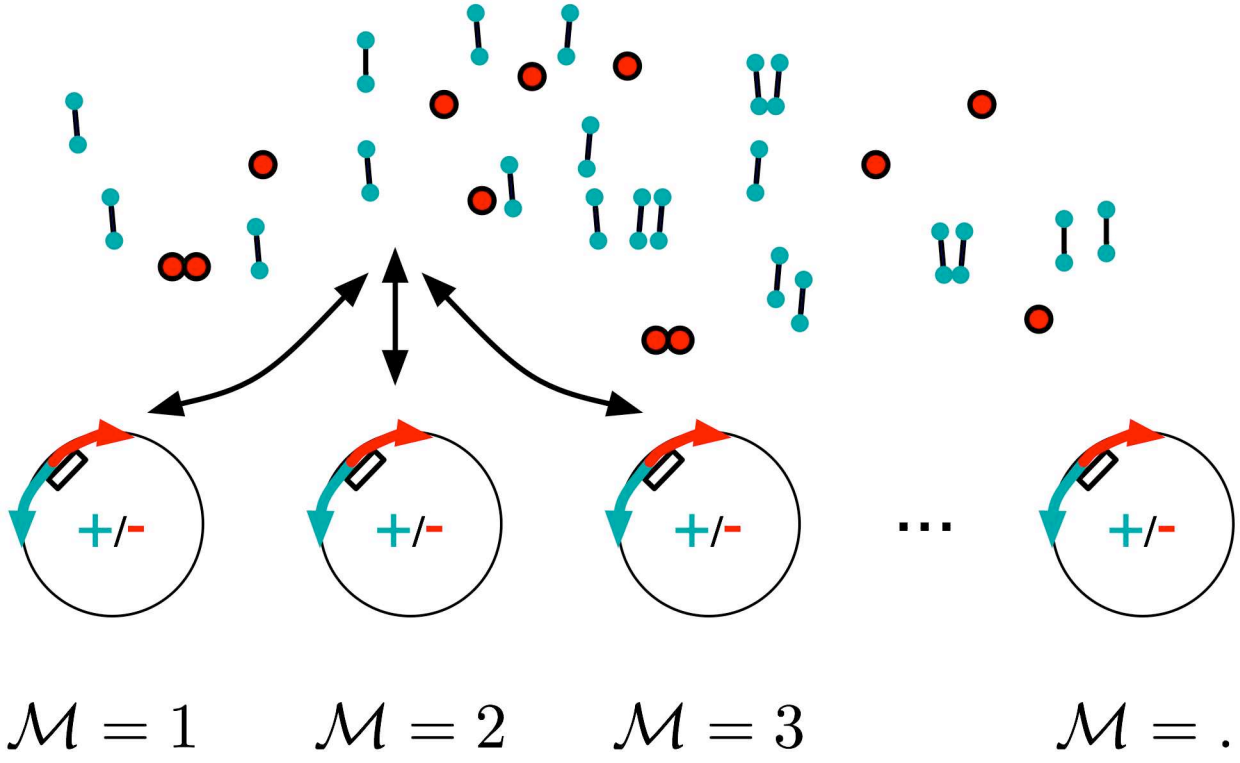




"In general, the amount of transcript produced by a gene is directly proportional to the number of copies of that gene in a cell."

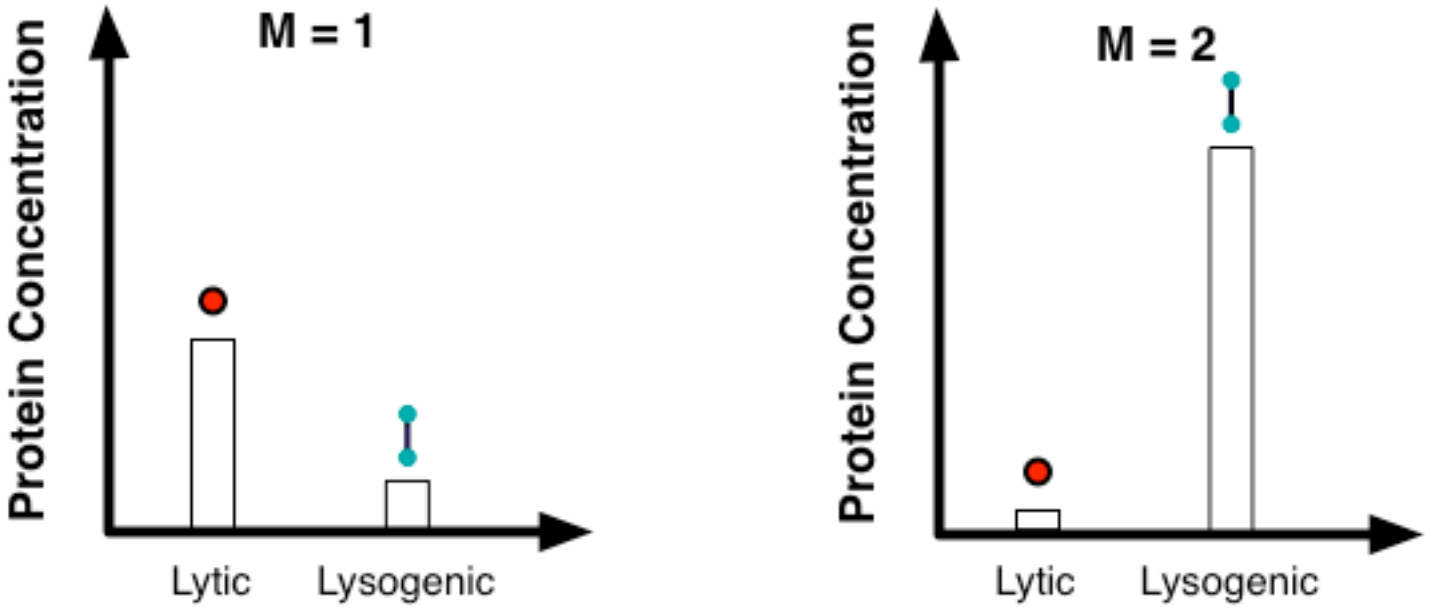
Griffiths et al., Intro. to Genetic Analysis

A “Simple” Decision Switch: In Reality, a Coupled Set of Regulatory Networks

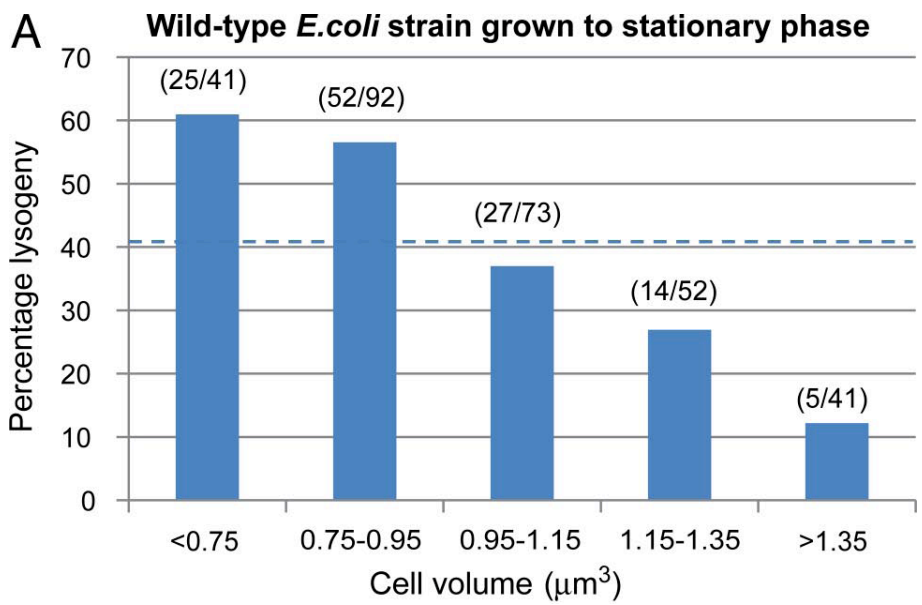


Communal gene products form the basis for “viral communication”

SO, Gene Expression May be Nonlinearly Related to Gene Copy Number:



Cell volume is another critical parameter

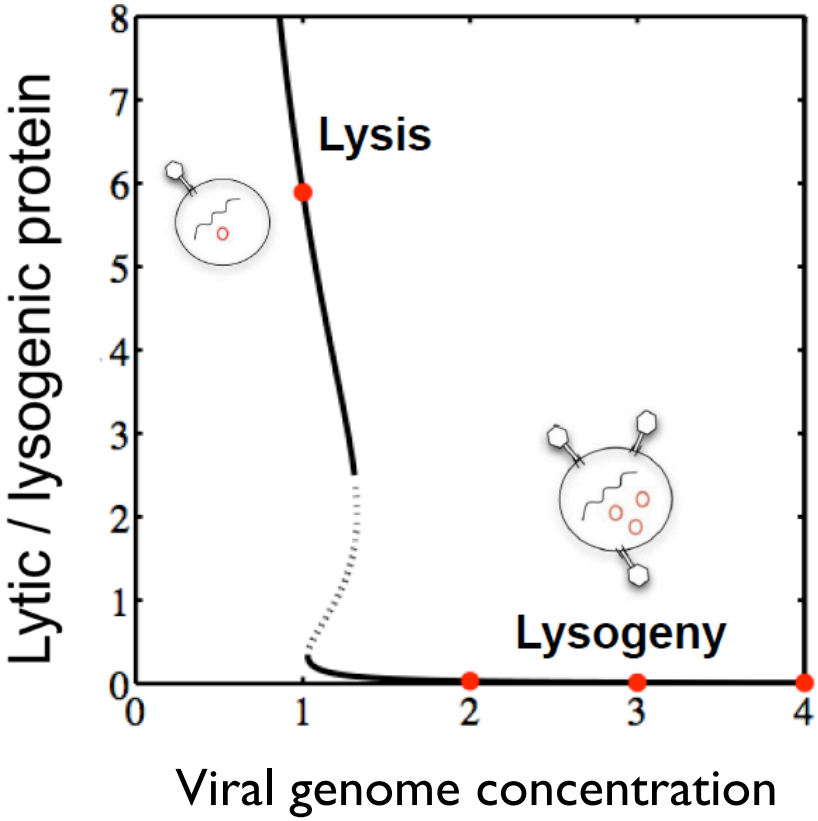


St. Pierre and Endy (2008) PNAS

- Lysis-lysogeny is stochastic
- Cell fate determination depends on concentration of viral genome

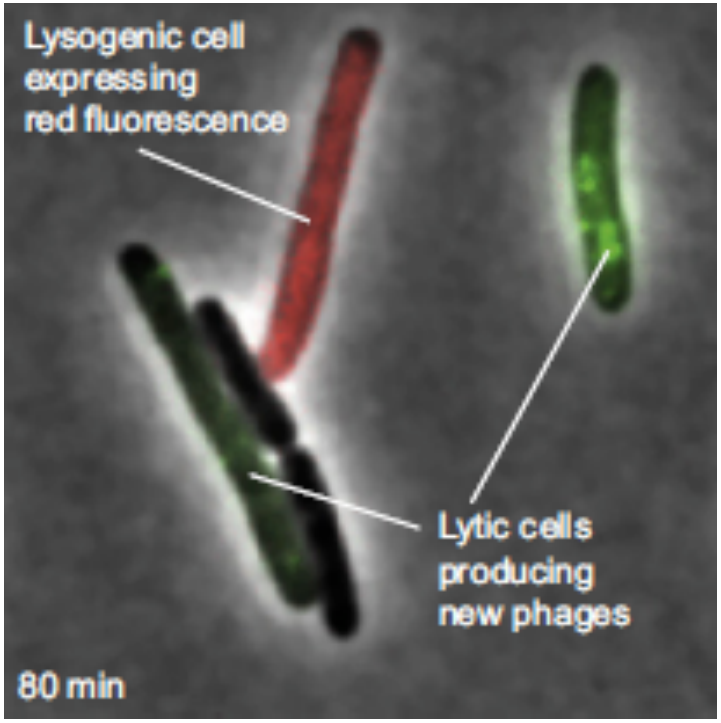
Viral concentration is key to lysis-lysogeny switch

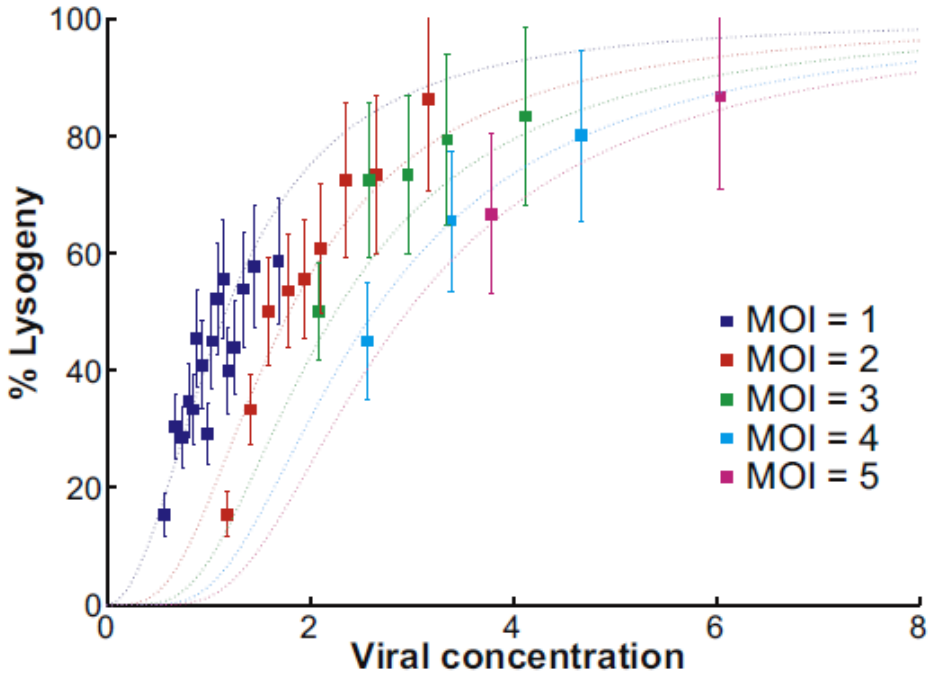
- Explicit consideration of viral genome concentration
- Based on asymptotic behaviors of gene regulation
- Small change in viral copy number can shift between lysis and lysogeny



Weitz, Mileyko, Joh and Voit (2008) Biophys J

Experiments confirmed concentration dependence





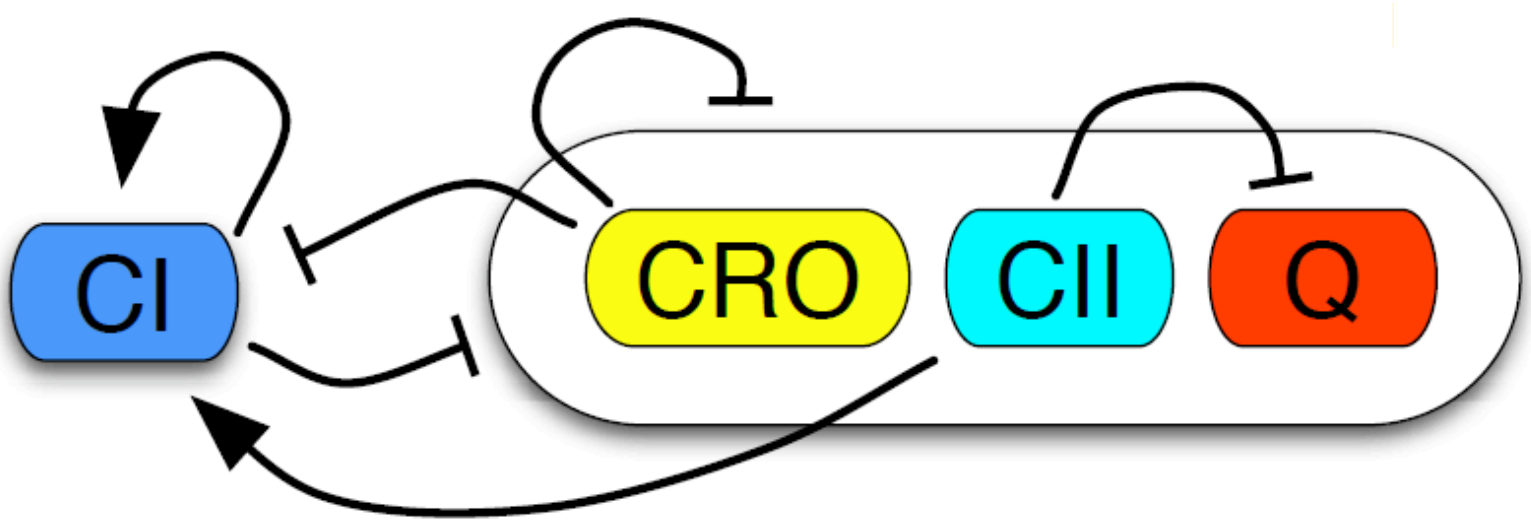
Zeng et al (2010) Cell

A stochastic model of phage lambda is necessary

Outline

- Determination of alternative cell fates
- Quantitative model of lysis-lysgoeny decisions
- Heterogeneity of decisions: gene dosage effect on lysis-lysogeny

Core GRN for lysis-lysogeny



- CI (repressor): lead to and maintain lysogeny
- CRO: control repressor
- CII: transcription activator
- Q: activate late lytic genes

Cumulative work of many (Ptashne, Kobiler, Oppenheim, and many more).

Quantitative model of lysis-lysogeny

$$[cro\ mRNA] \qquad \frac{dm_y}{dt} = \underbrace{\frac{\mathcal{M}}{V}\alpha_y f_R}_{\text{Transcription}} - \underbrace{\gamma_m m_y}_{\text{Degradation}}$$

$$[\mathrm{CRO}] \qquad \frac{dY}{dt} = \underbrace{\sigma m_y}_{\text{Translation}} - \underbrace{\gamma_y Y}_{\text{Degradation}}$$

Full model of lysis-lysogeny

# of coinfecting phages

$$
\begin{aligned}
[cI\ mRNA] \quad \frac{dm_x}{dt} &= \frac{\mathcal{M}}{V}\alpha_x f_{RM}^{basal} + \frac{\mathcal{M}}{V}\beta_x f_{RM}^{act} + \frac{\mathcal{M}}{V}\delta_x f_{RE} - \gamma_m m_x\,, \\
[cro\ mRNA] \quad \frac{dm_y}{dt} &= \frac{\mathcal{M}}{V}\alpha_y f_R - \gamma_m m_y\,, \\
[cII\ mRNA] \quad \frac{dm_z}{dt} &= \frac{\mathcal{M}}{V}\alpha_z f_R - \gamma_m m_z\,, \\
[Q\ mRNA] \quad \frac{dm_Q}{dt} &= \frac{\mathcal{M}}{V}\alpha_Q f_R - \gamma_m m_Q - \zeta m_Q m_{aQ}\,, \\
[aQ\ mRNA] \quad \frac{dm_{aQ}}{dt} &= \frac{\mathcal{M}}{V}\delta_{aQ} f_{aQ} - \gamma_m m_{aQ} - \zeta m_Q m_{aQ}\,, \\
[\mathrm{CI}] \quad \frac{dX}{dt} &= \sigma m_x - \gamma_x X\,, \\
[\mathrm{CRO}] \quad \frac{dY}{dt} &= \sigma m_y - \gamma_y Y\,, \\
[\mathrm{CII}] \quad \frac{dZ}{dt} &= \sigma m_z - \gamma_z Z\,, \\
[\mathrm{Q}] \quad \frac{dQ}{dt} &= \sigma m_Q - \gamma_Q Q\,,
\end{aligned}
$$

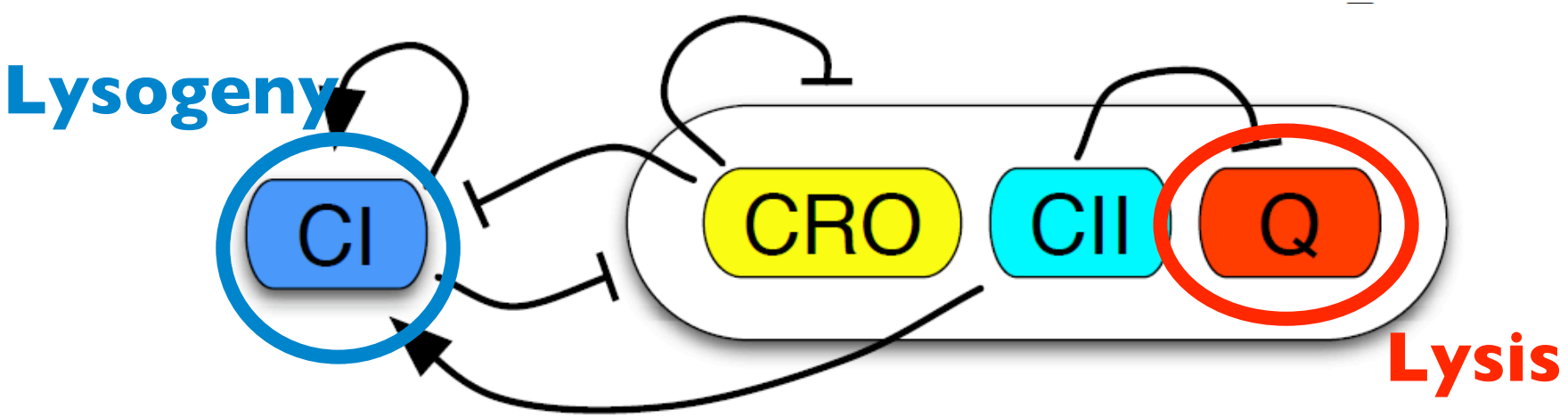
Host cell volume

Parameters for lysis-lysogeny

Parameter	Reference value	Reference
γ_x	0.01 (min^{-1})	≈ 0 [62], 0.042 [25]
γ_y	0.06 (min^{-1})	0.016 [63]
γ_z	0.10 (min^{-1})	0.16 w/o CIII [64]
γ_q	0.01 (min^{-1})	
γ_m	0.1 (min^{-1})	0.12 [65]
α_x	0.06 (min^{-1})	0.06 [36]
α_y	0.84 (min^{-1})	0.84 [36], 3 [62]
α_z	0.8 (min^{-1})	$< \alpha_y$
α_q	0.75 (min^{-1})	$< \alpha_z$
β_x	0.66 (min^{-1})	0.66 [36], 3.42 [66]
δ_x	0.9 (min^{-1})	0.9 [25]
δ_{aQ}	2 (min^{-1})	
c_d^x	0.05 (nM^{-1})	0.05 [67], 0.18 [68]
c_d^y	5.8 (nM^{-1})	5.8 [69], 307 [70]
c_d^z	0.05 (nM^{-1})	
c_t^z	0.05 (nM^{-1})	
c_p^{aQ}	0.2 (nM^{-1})	
σ	0.5 (min^{-1})	
ζ	0.1 $(nM^{-1}min^{-1})$	0.02 [71]
V	1 (μm^3)	0.5$\sim$2.0

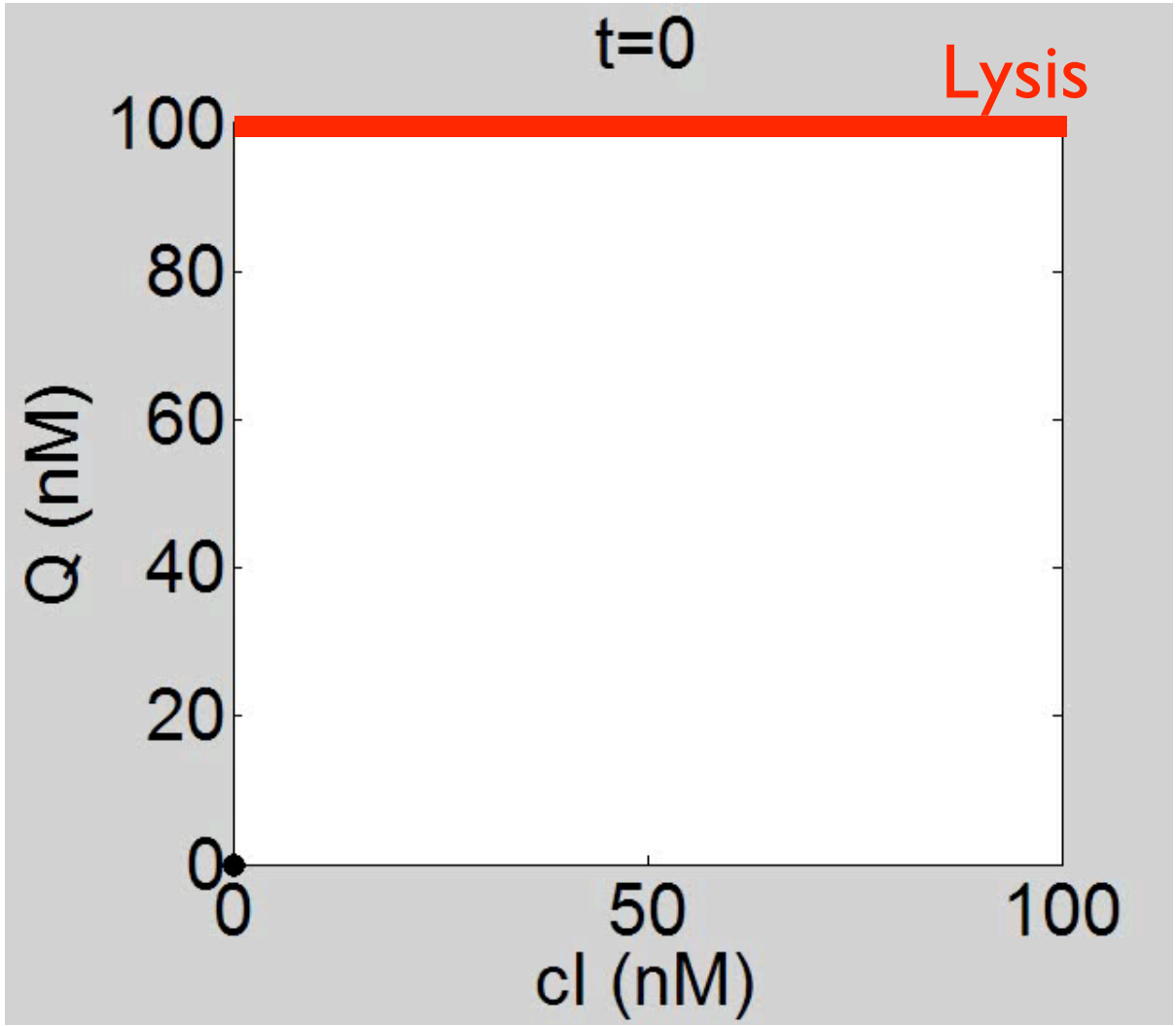
Stochastic simulations by Gillespie algorithm

- Stochastic simulation: time evolution trajectory using Gillespie algorithm

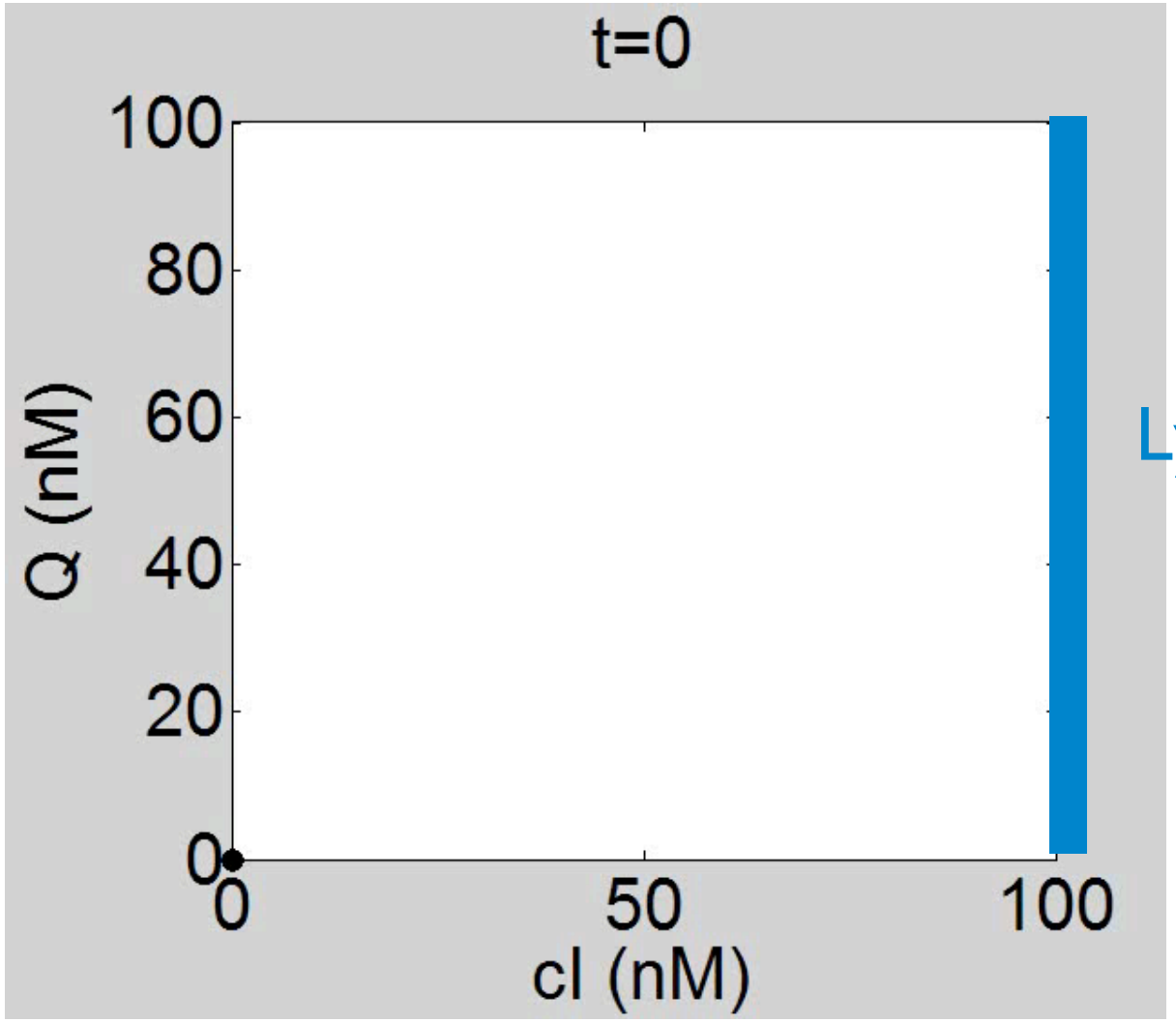


- Threshold concentrations for decisions
  - Reaching CI threshold: lysogeny
  - Reaching Q threshold: lysis

First passage: lysis

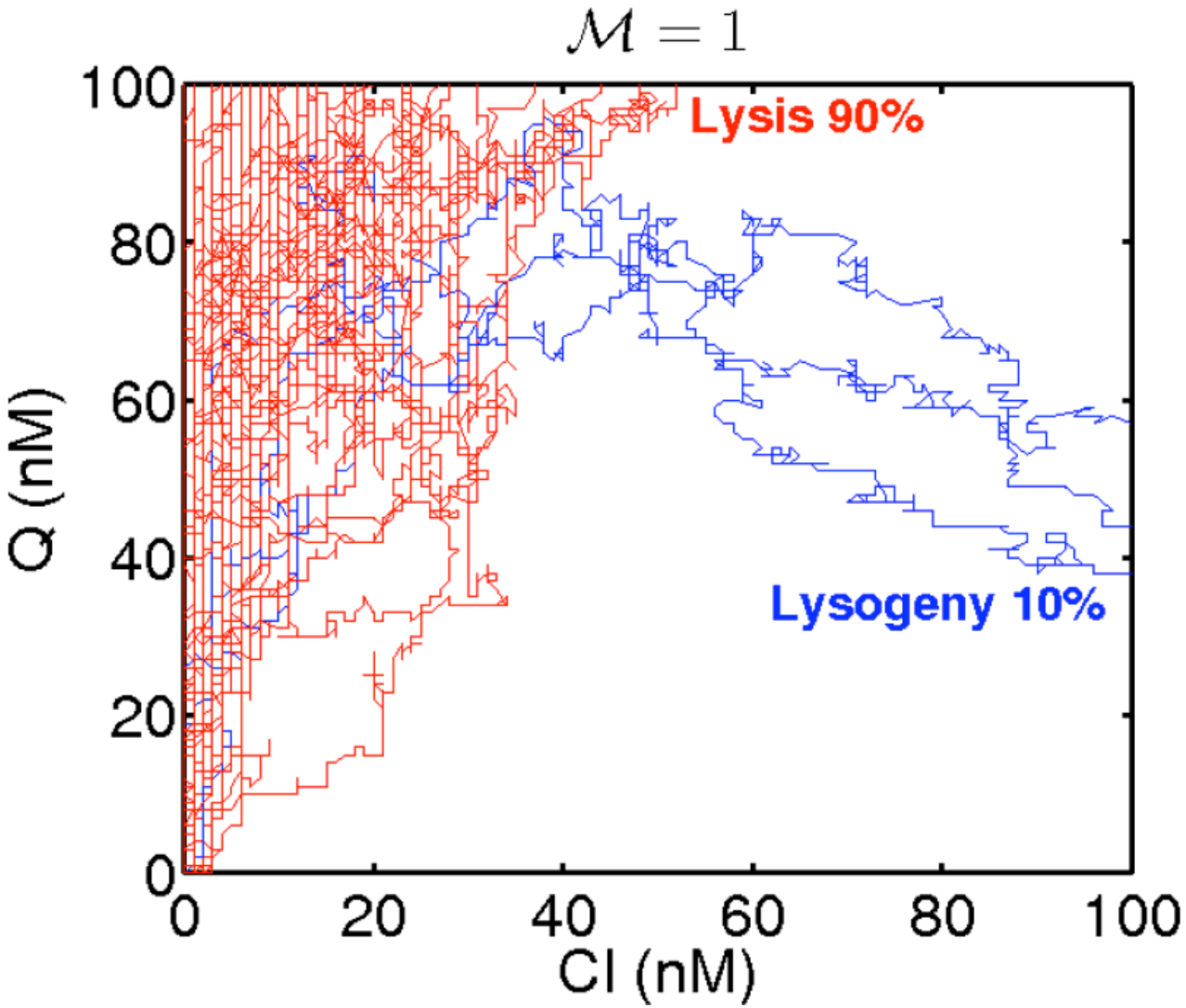


First passage: lysogeny

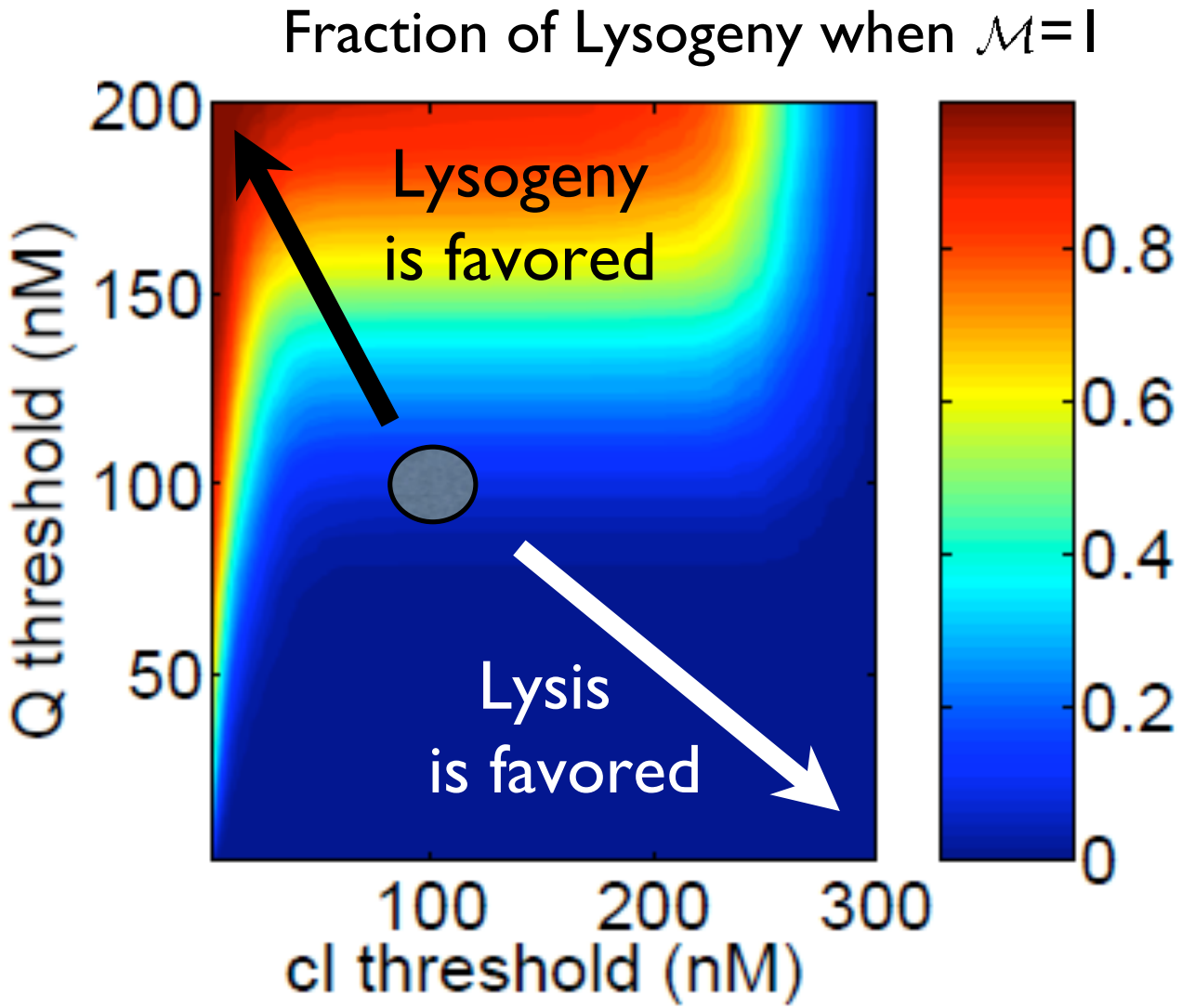


Lysogeny

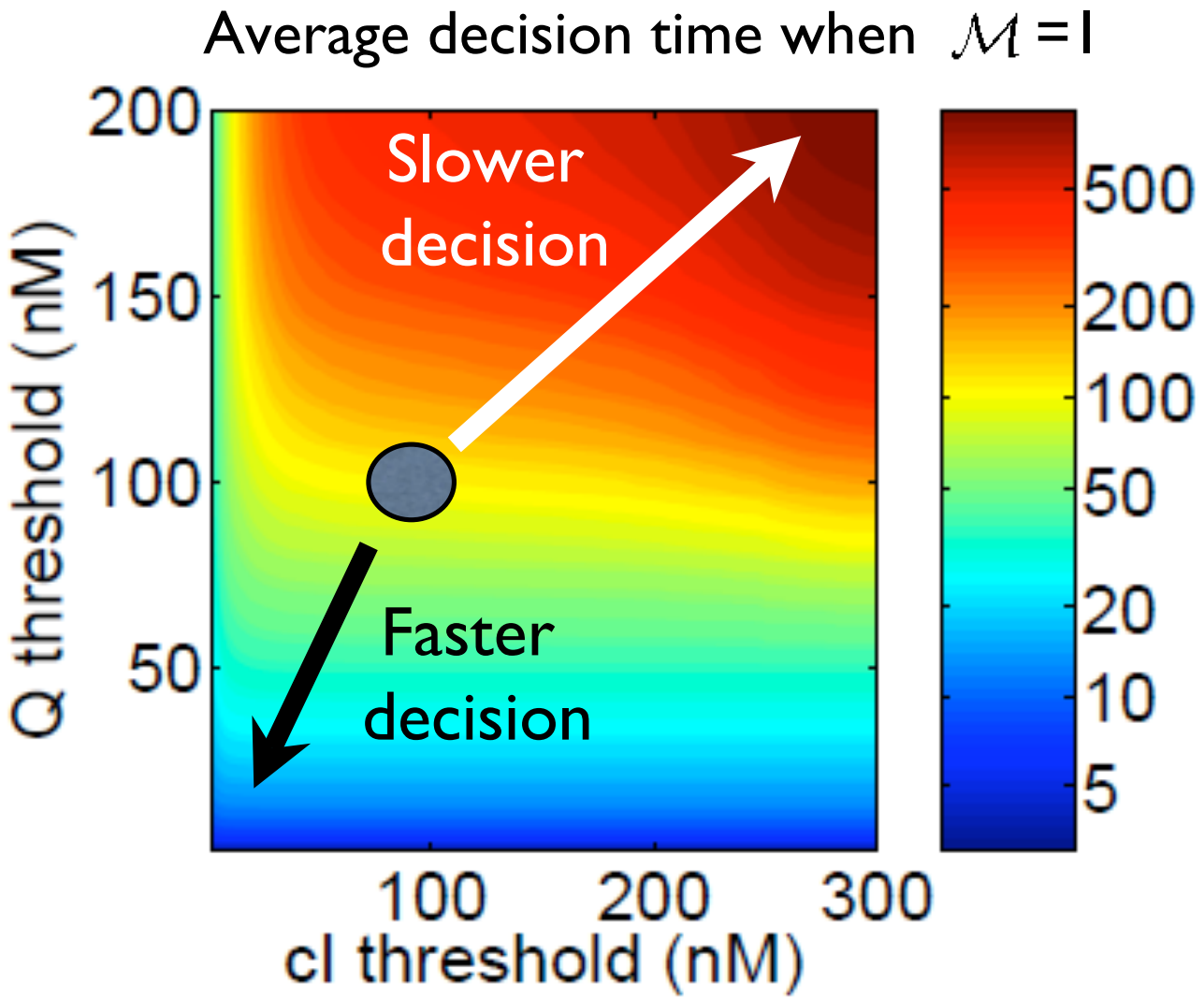
Fraction of lysogeny determined by multiple stochastic simulations



Thresholds as evolvable traits



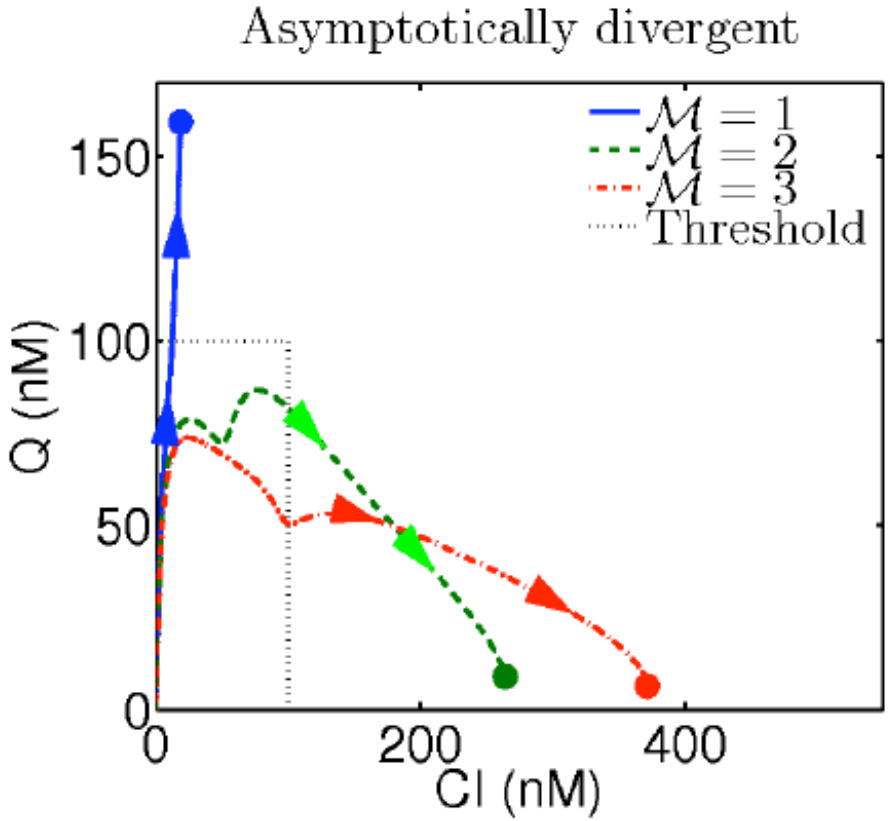
Average decision time can also be tuned by thresholds

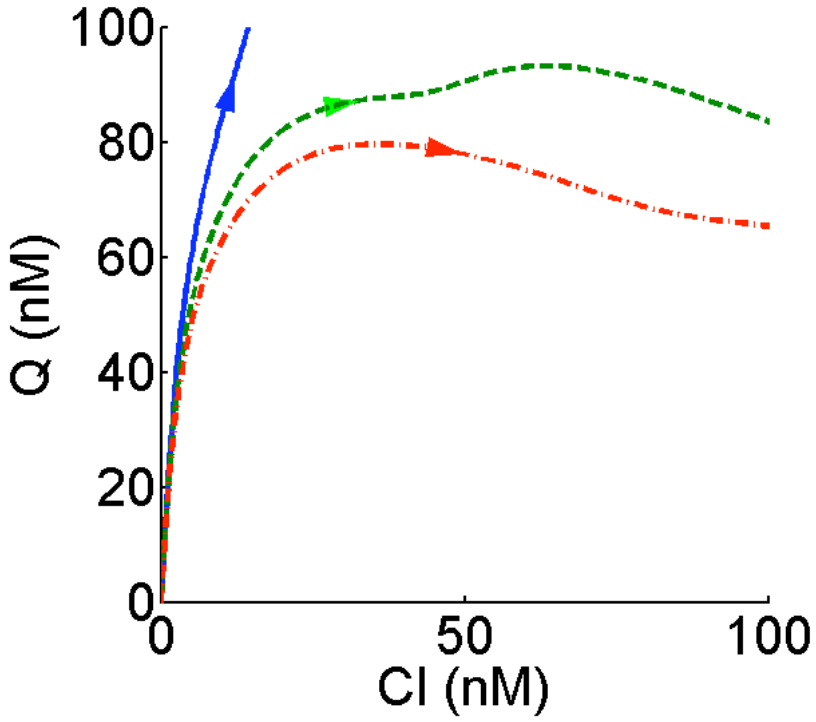


What are essential features of alternative decisions?

- Is bistability necessary for alternative decisions?
- Can two systems behave the same way due to similarity of transient dynamics?

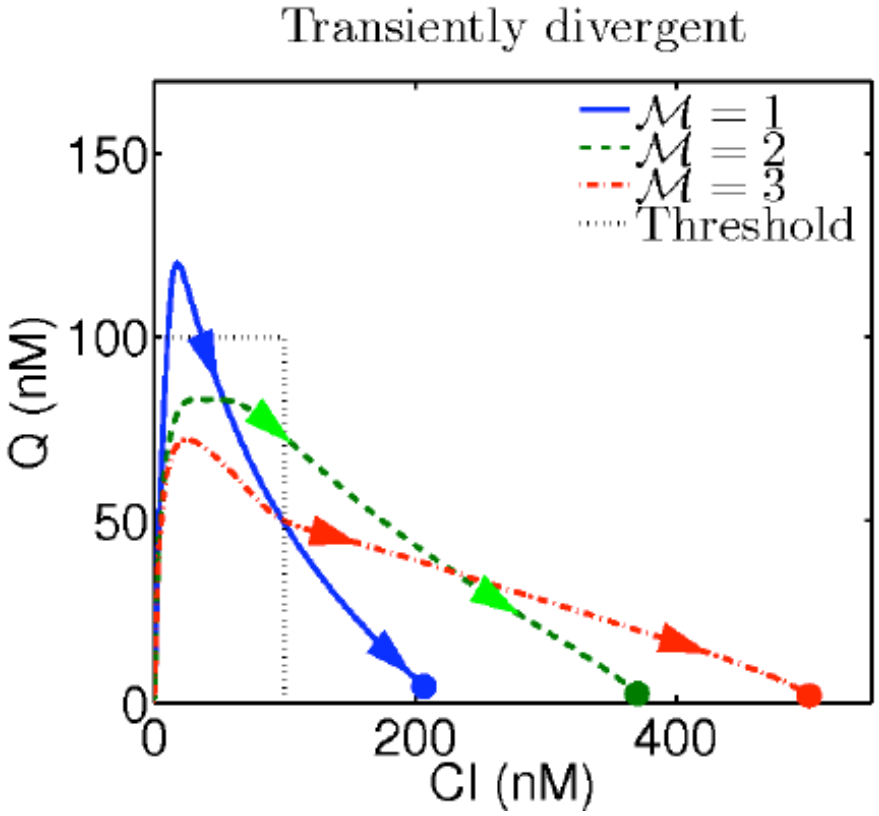
Asymptotically divergent GRN

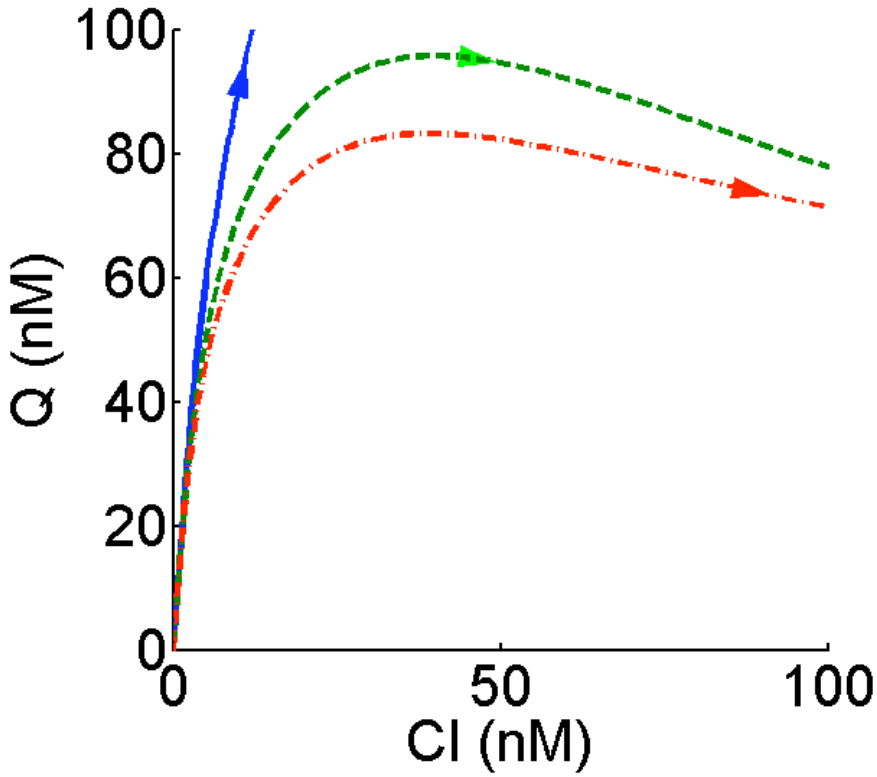




- $\mathcal{M}$=1: Q expression level stays high.
- $\mathcal{M}$ >1: Q→0.

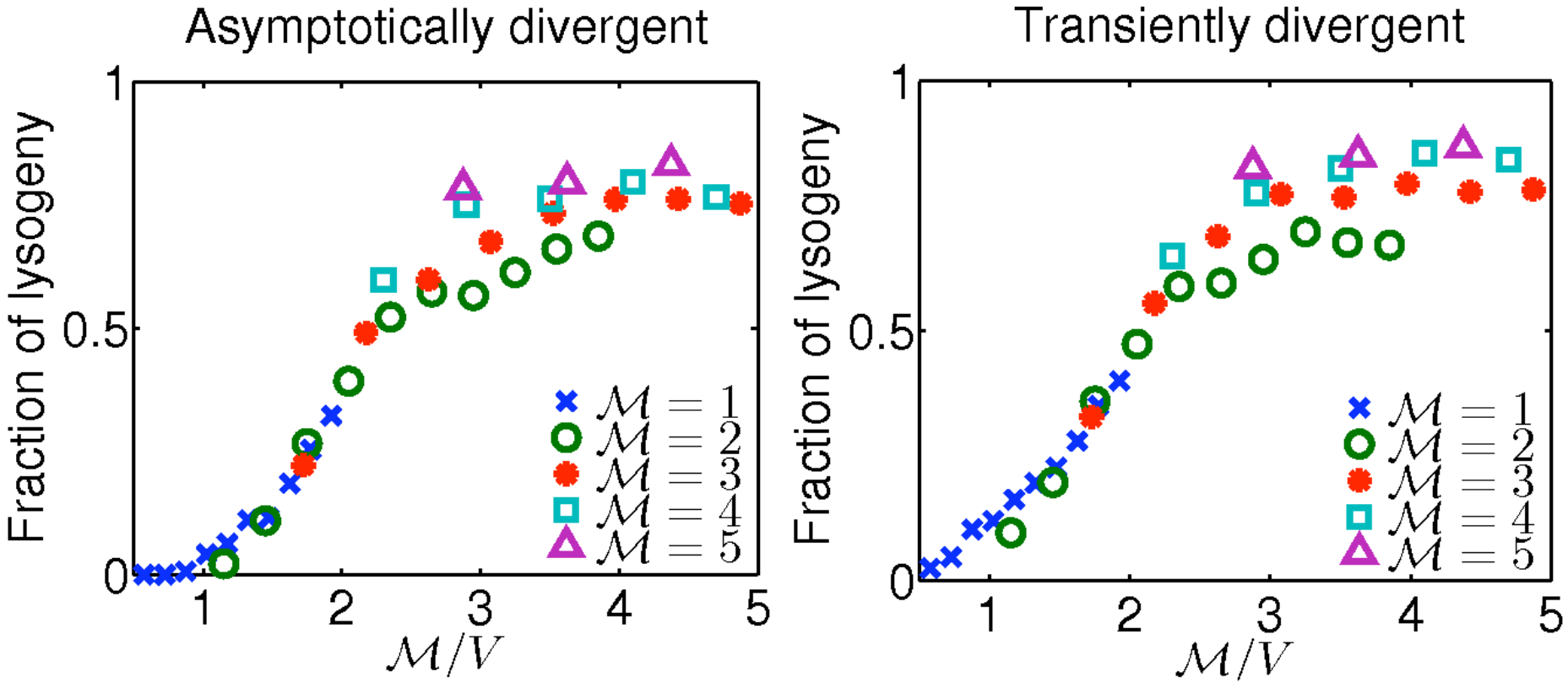
Transiently divergent GRN





- At all $\mathcal{M}$s, Q→0.
- Maximum transient levels of Q are different.

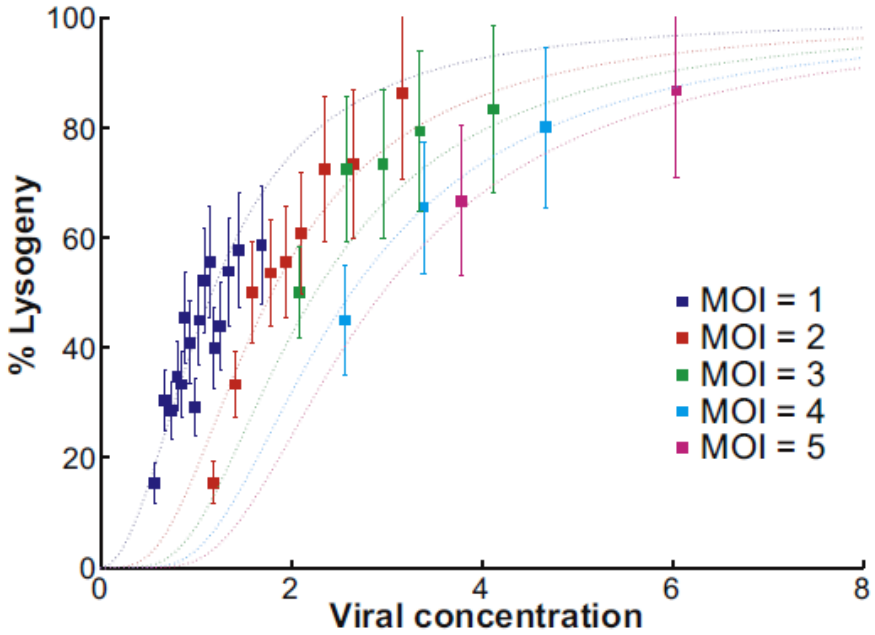
Similarity of transient dynamics leads to same response



- Responses can be very similar even if steady-state behaviors are qualitatively distinct

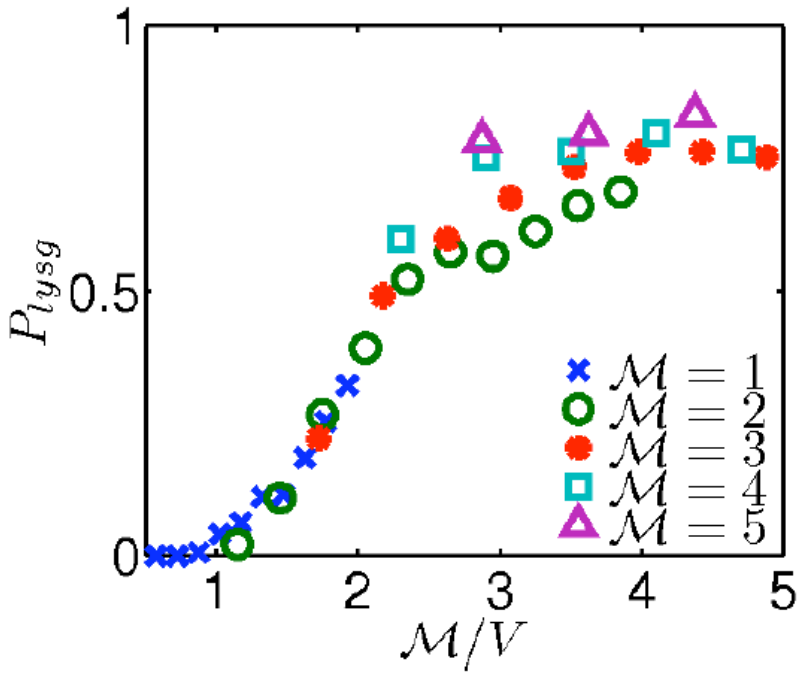
Experiments show much more heterogeneity than simulations

Data



Simulation

Asymptotically divergent



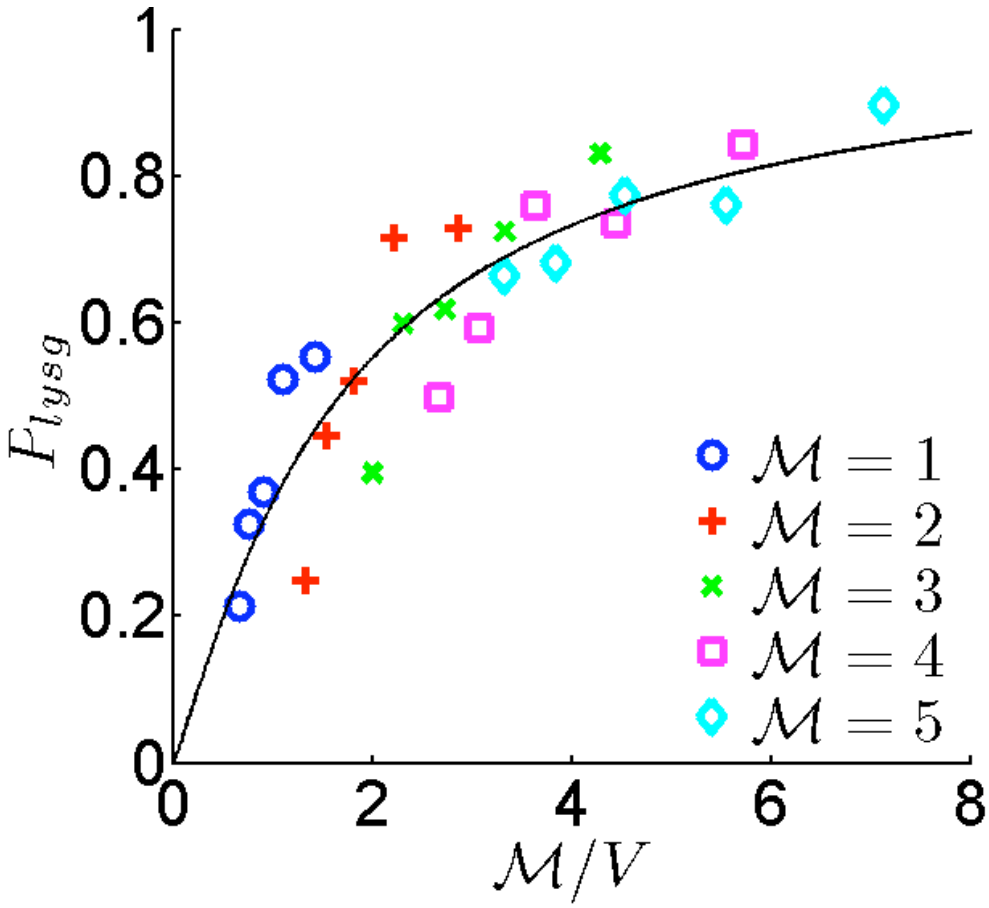
Zeng et al (2010) Cell

How can we explain this discrepancy?

Outline

- Determination of alternative cell fates
- Quantitative model of lysis-lysgoeny decisions
- Heterogeneity of decisions: gene dosage effect on lysis-lysogeny

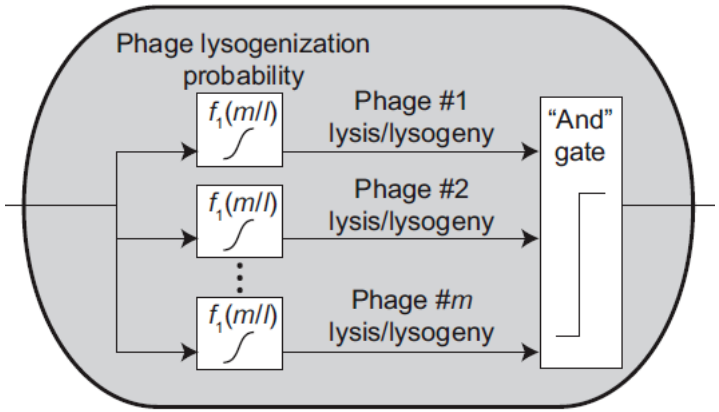
What can explain the variance of lysogeny?



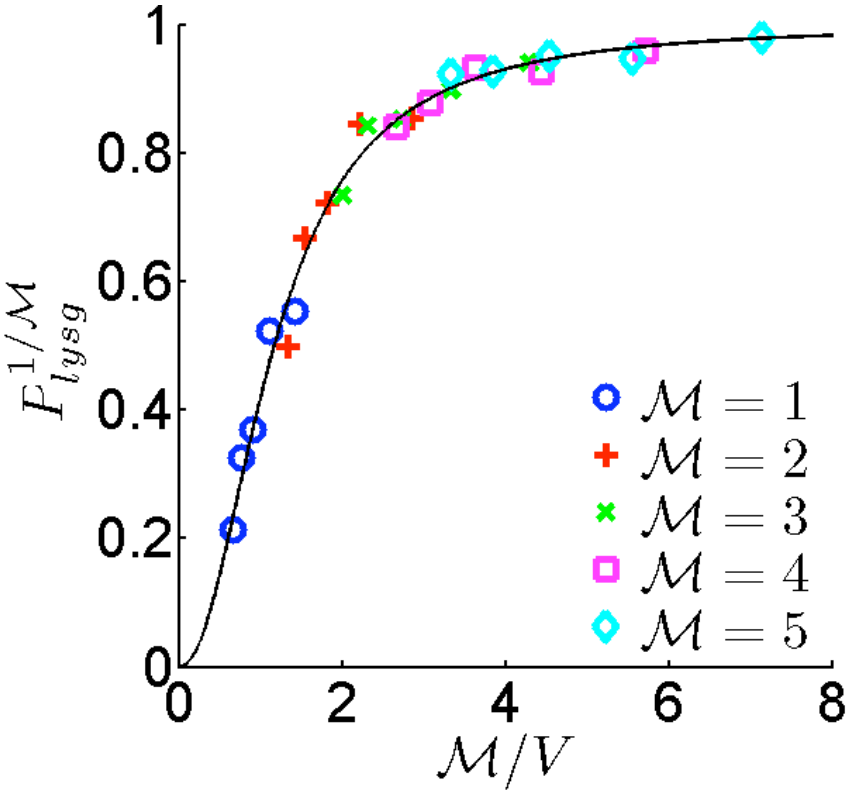
- At same $\mathcal{M}/V$, a singly infected cell has much higher probability of lysogeny than a doubly infected cell

Data taken from Zeng et al (2010) Cell

Quasi-independent decision proposed by Zeng et al (2010)



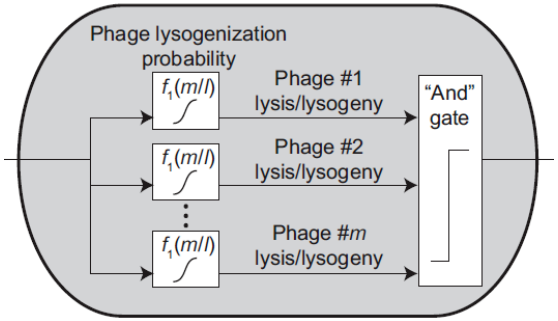
$$P_{lysg} = (P_{1\,phage})^{\mathcal{M}}$$



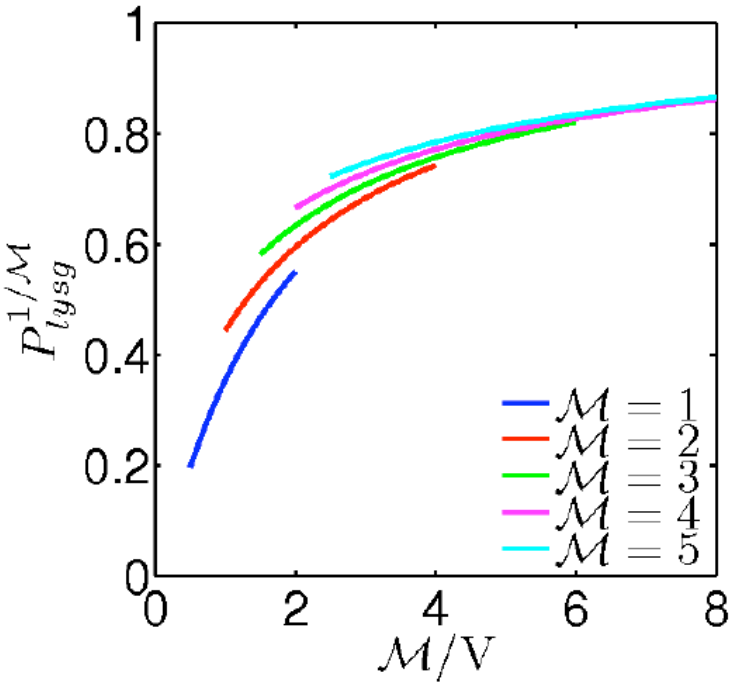
- Phages are independent
- Phages know the presence of other phages

When phages are totally independent

- Assume phages have no way of detecting other phages within a host

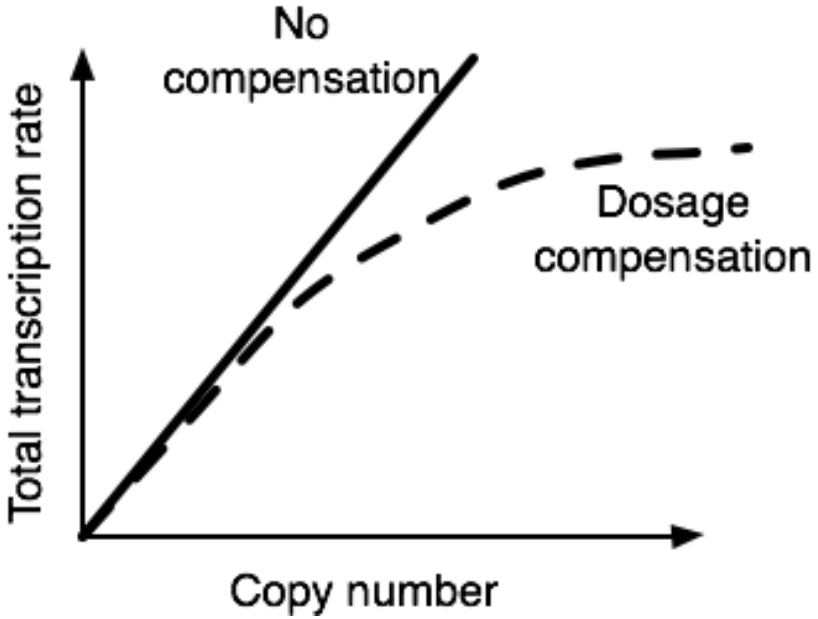


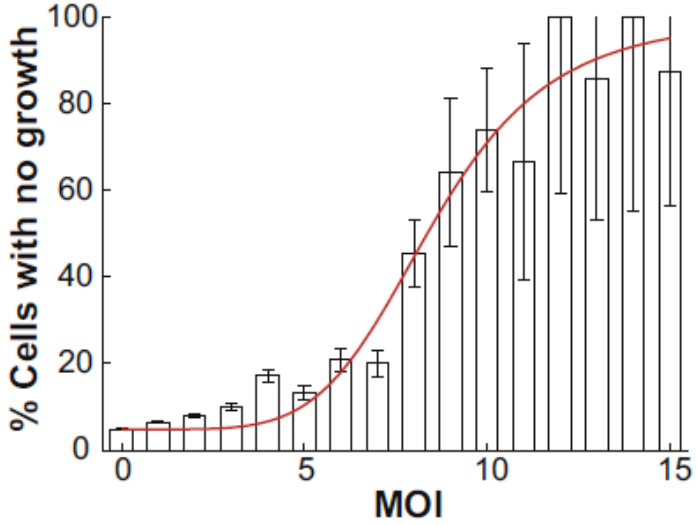
$$f\left(\frac{\mathcal{M}}{V}\right) \rightarrow f\left(\frac{1}{V}\right)$$



Gene dosage compensation

- Gene expressions is not always proportional to copy number



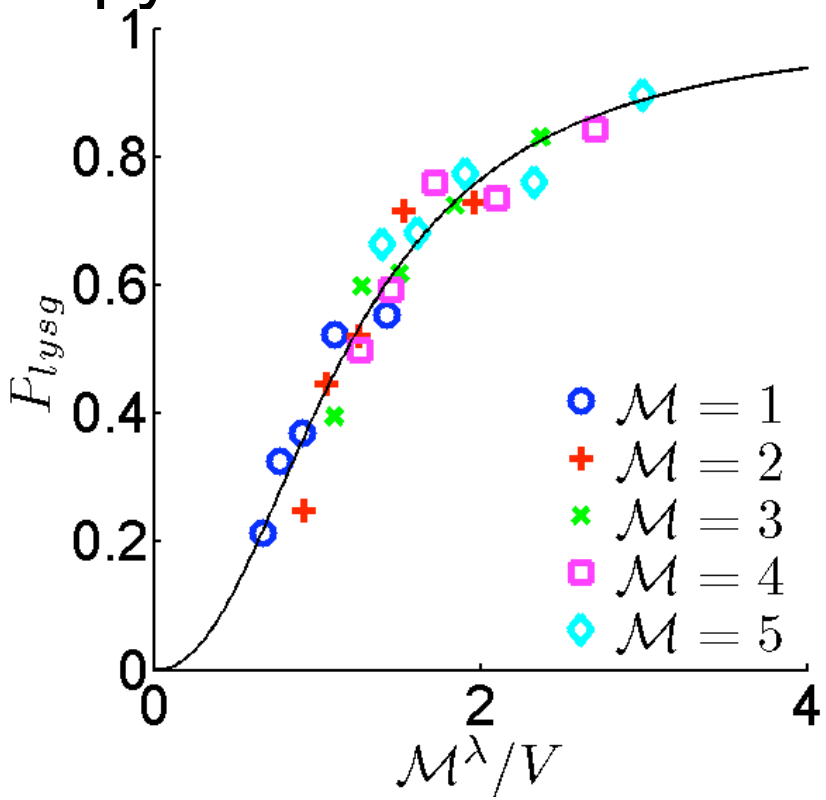


Zeng et al (2010) Cell

Tot transcription = $\mathcal{M}^{\lambda}$ transcription/copy

Data can also be collapsed by partial dosage compensation

- Effective copy number is smaller than actual copy number



Data supports $\lambda = 0.5$

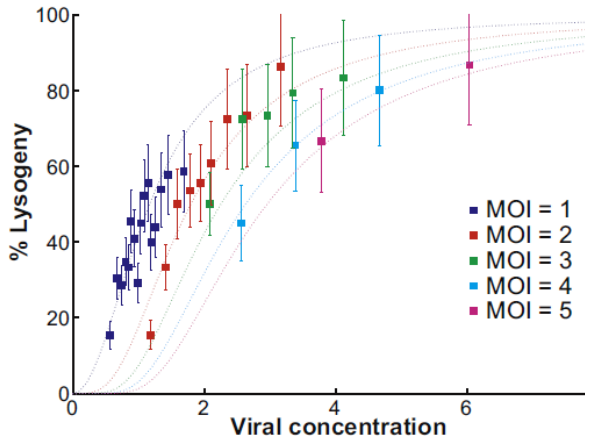
Comparison of different rescaling schemes

- Quasi-independent decisions:
  - Decision for each phage is independent
  - However, the decision rule for each phage depends on the concentration of all phages

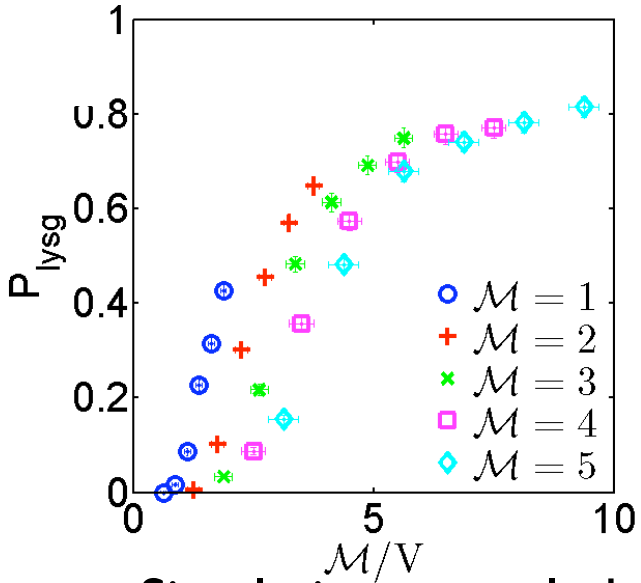
- Gene dosage compensation
  - Mechanism by which resource limitation impacts viral gene production
  - Effective number of viral genomes is predicted to be less than the actual number

Stochastic simulations support effect of dosage compensation

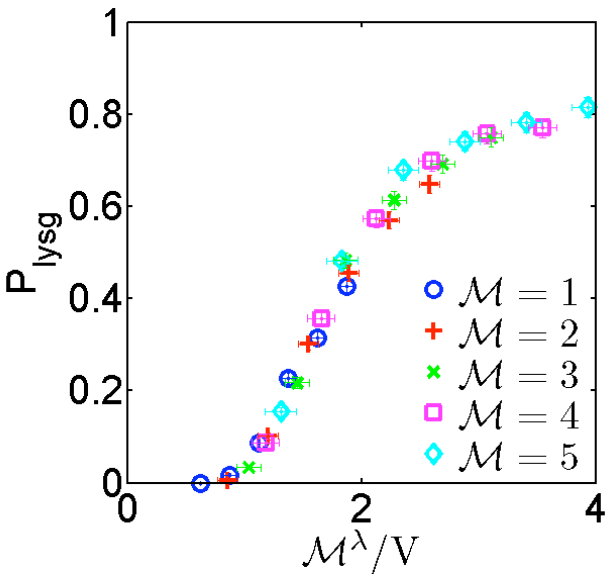
- Replace $\mathcal{M}$ with $\mathcal{M}^{\lambda}$ in our simulation



Data



Simulation, unscaled



Simulation, rescaled

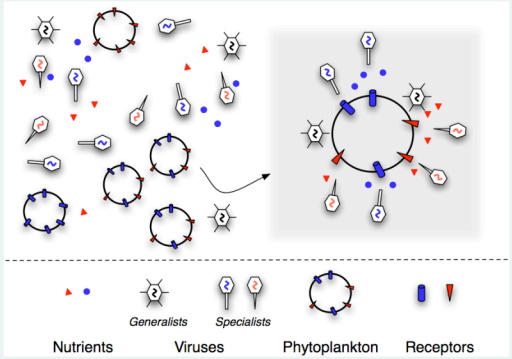
Stochastic simulations supports partial gene dosage compensation

Conclusions

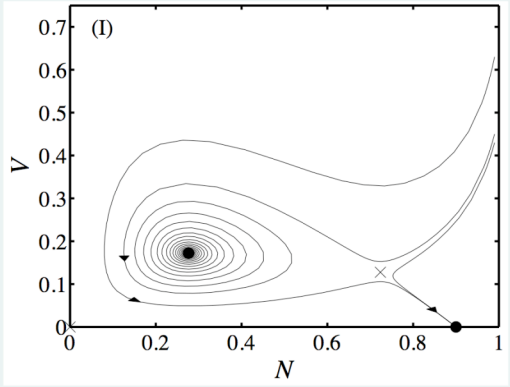
- Feedback and transient dynamics of gene regulation are sufficient for lysis and lysogeny
- Systems with qualitatively distinct steady state behaviors might lead to similar decisions if their transient dynamics are similar
- Gene dosage compensation can explain observed variation of MOI dependence
- Future work involves predicting cell fate based on partial information of gene regulatory state

Other Things We Do

Eco-evolutionary dynamics of phages and their hosts

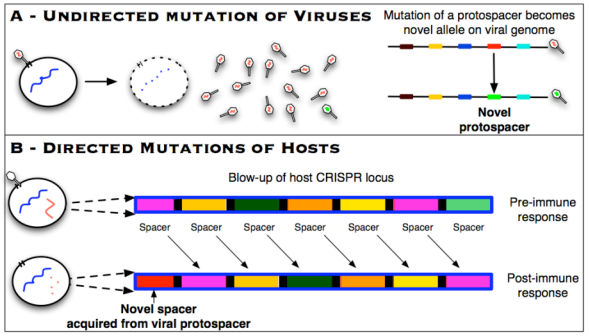


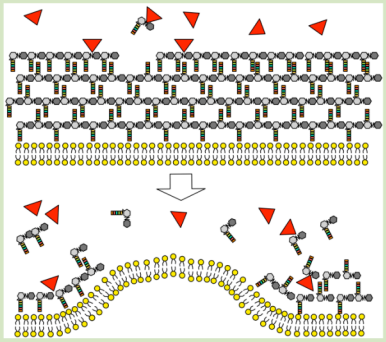
Dangerous nutrients: top-down vs. bottom up forces impact evolution of resource uptake
Menge & Weitz (2009) JTB 257: 104



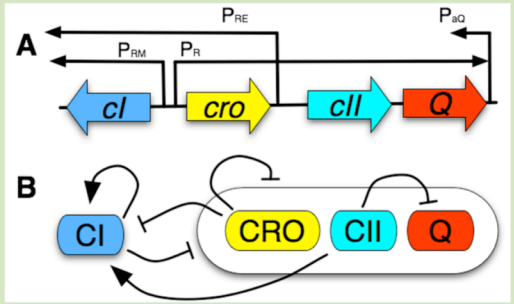
Host-state impacts phage effectiveness and subsequent host-phage dynamics
Dushoff & Weitz (2008). Theor Ecol 1:13

CRISPR-induced co-evolutionary dynamics
Weitz & collaborators (in prep)





Quantifying enzymatic lysis
Mitchell et al. (2010) Physical Biology 7:046002



Cell fate determination by viruses
Joh & Weitz (in press) PLoS Comp Biol

Systems biology and biophysics of phage traits

Acknowledgments

Weitz Lab
- Richard Joh
- Tae Lee
- Yuriy Mileyko (Duke)

Russell Monds, Stanford
Selwyn Quan, Stanford
Harold Kim, GATech
Eberhard Voit, GATech

Ido Golding, Baylor College of Medicine
Lanying Zeng, UIUC
NESCENT Working Group

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Mileyko, Joh and Weitz(2008) *Small-scale copy number variation and large-scale changes in gene expression*, **PNAS**, 105:16659-16664.

Gudelj, Weitz et al (2010). *An integrative approach to understanding microbial diversity: from intracellular mechanisms to community structure.* **Ecology Letters**. 13:1073-1084.

Joh and Weitz (in press) *To lyse or not to lyse: transient-mediated stochastic fate determination in cells infected by bacteriophages.* **PLoS Computational Biology**

BURROUGHS WELLCOME FUND

FUNBIO LAWS

James S. McDonnell Foundation

Questions?

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