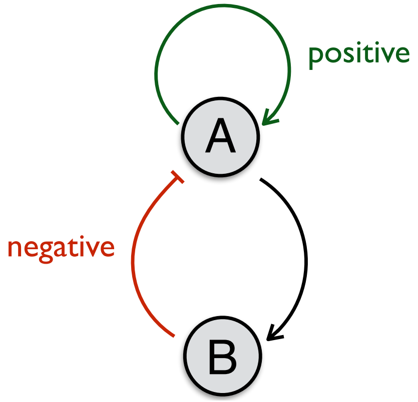
Understanding a dual feedback oscillator



Modelling positive feedback: auto-regulation of protein A on gene *A*

transcription

$$\frac{dM}{dt} = \frac{u_{\mathrm{basal}} + u_{\mathrm{max}} \frac{A^2}{K_2^2}}{1 + \frac{A^2}{K_A^2}} - d_M M$$

translation

$$\frac{dA}{dt} = vM - d_A A$$

quasi-steady state

$$\frac{dM}{dt} \simeq 0$$

$$\Longrightarrow$$

$$\frac{dA}{dt} = \frac{v}{d_M} \left[\frac{u_{\mathrm{basal}} + u_{\mathrm{max}} \frac{A^2}{K_A^2}}{1 + \frac{A^2}{K_A^2}} \right] - d_A A$$

Reducing the number of parameters by rescaling

Rescale **times by a time** in the model and **concentrations by a concentration** in the model to give dimensionless units:

$$\frac{dA}{dt} = \frac{v}{d_M}\left[\frac{u_{\text{basal}} + u_{\max}\frac{A^2}{K_A^2}}{1+\frac{A^2}{K_A^2}}\right] - d_A A$$

choose

$$\tilde{t} = d_A t$$

$$\tilde{A} = \frac{A}{K_A}$$

Then

$$\frac{1}{d_A}\cdot\frac{d}{dt}\left(\frac{A}{K_A}\right) = \frac{v}{d_A d_M K_A}\left[\frac{u_{\text{basal}} + u_{\max}\frac{A^2}{K_A^2}}{1+\frac{A^2}{K_A^2}}\right] - \frac{A}{K_A}$$

or

$$\frac{d\tilde{A}}{d\tilde{t}} = \alpha\left[\frac{b+\tilde{A}^2}{1+\tilde{A}^2}\right] - \tilde{A}$$

with

$$\alpha = \frac{u_{\max} v}{d_A d_M K_A}$$

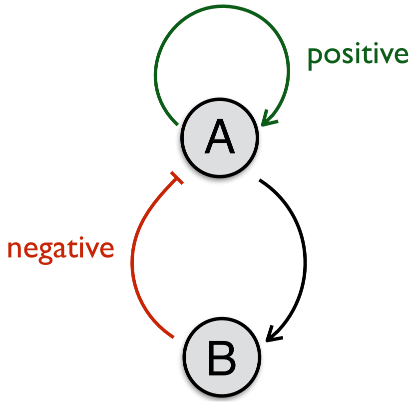
$$b = \frac{u_{\text{basal}}}{u_{\max}}$$

Two parameters instead of six!

Including both positive and negative feedback

$$\frac{dA}{dt} = \boxed{\frac{1}{1+\left(\frac{B}{K}\right)^2}} \times \boxed{\alpha\frac{b+A^2}{1+A^2}} - A$$

$$\frac{dB}{dt} = \kappa A - d_B B$$



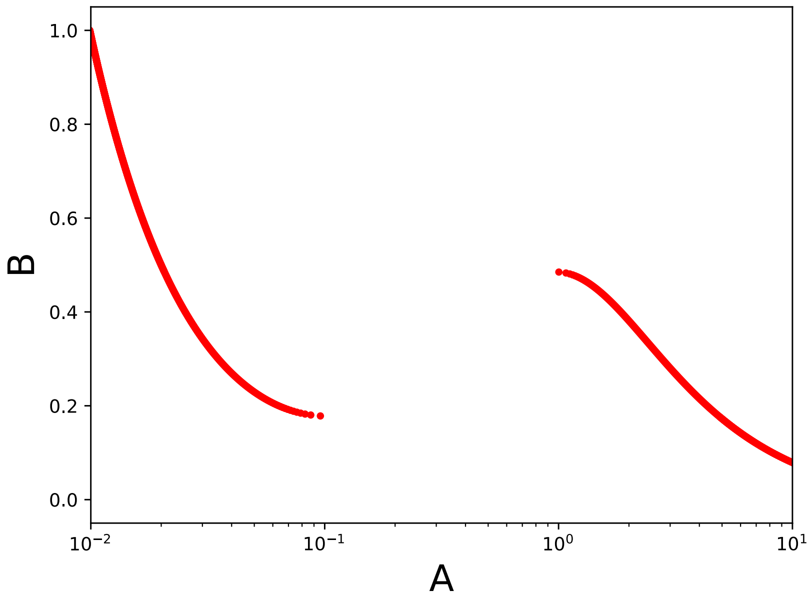
For a constant B, the positive feedback can generate bistability

At steady state

$$\frac{dA}{dt} = 0 \qquad \text{so} \qquad \tilde{\alpha}(b + A^2) = A(1 + A^2) \qquad \text{or} \qquad A^3 - \tilde{\alpha}A^2 + A - \tilde{\alpha}b = 0$$

$$\text{with} \qquad \tilde{\alpha} = \frac{\alpha}{\left[1 + \left(\frac{B}{K}\right)^2\right]}$$

There are two stable fixed points for each constant value of B.

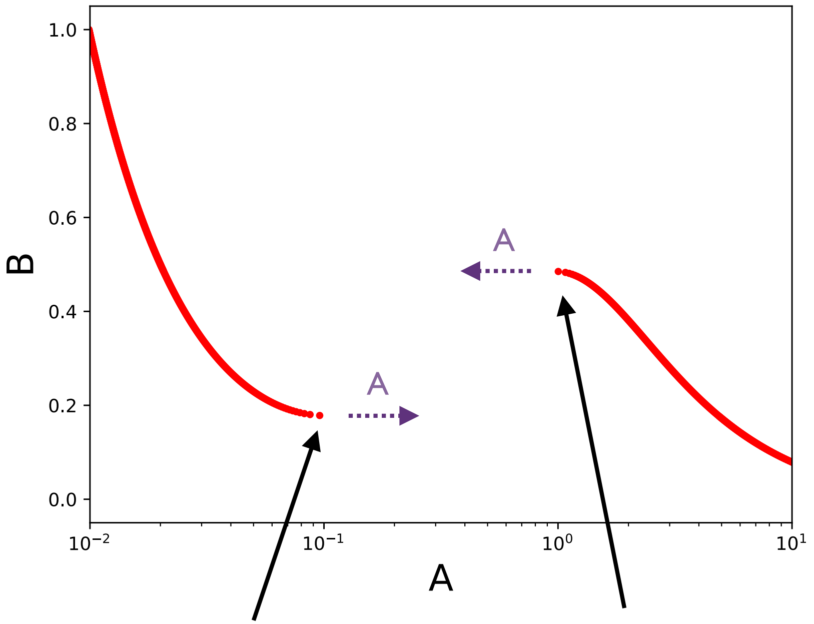


A bifurcation diagram not a phase-plane plot.

The negative feedback via B should destabilise the system to generate oscillations.

B should encourage A to move between the two branches of steady states

$$\frac{dA}{dt} = \frac{\alpha\left[b+A^2\right]}{\left[1+\left(\frac{B}{K}\right)^2\right]\left[1+A^2\right]} - A$$



B should act to increase A, so less repression of gene *A* by B

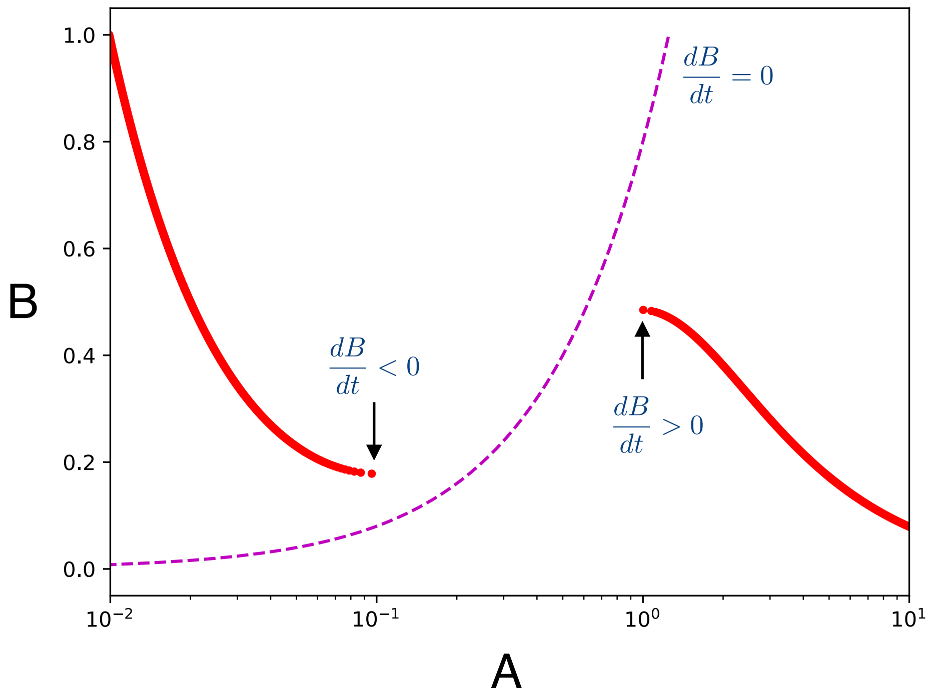
$$\frac{dB}{dt} < 0$$

B should act to decrease A, so more repression of gene *A* by B

$$\frac{dB}{dt} > 0$$

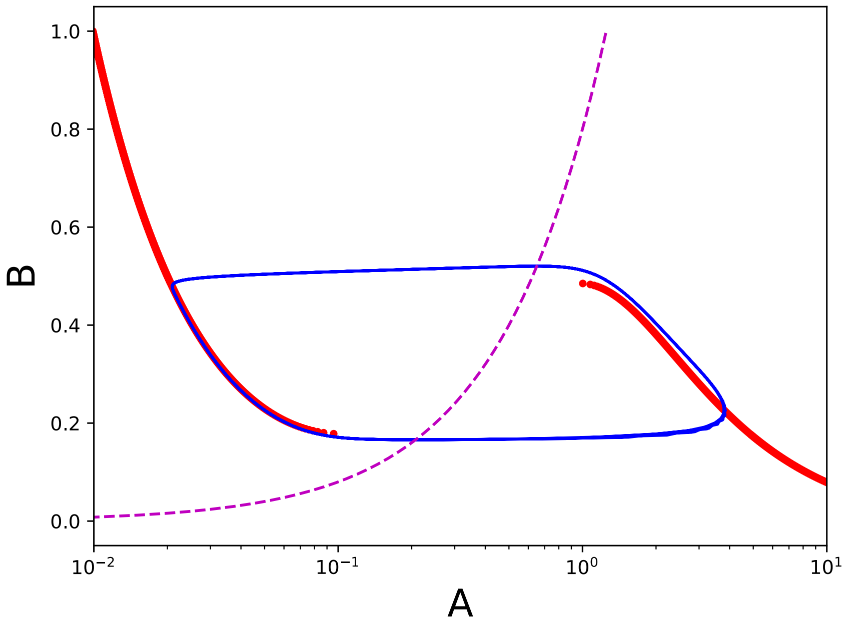
If B’s nullcline is between the two branches of steady states, B is destabilising

$$\frac{dB}{dt} = \kappa A - d_B B$$



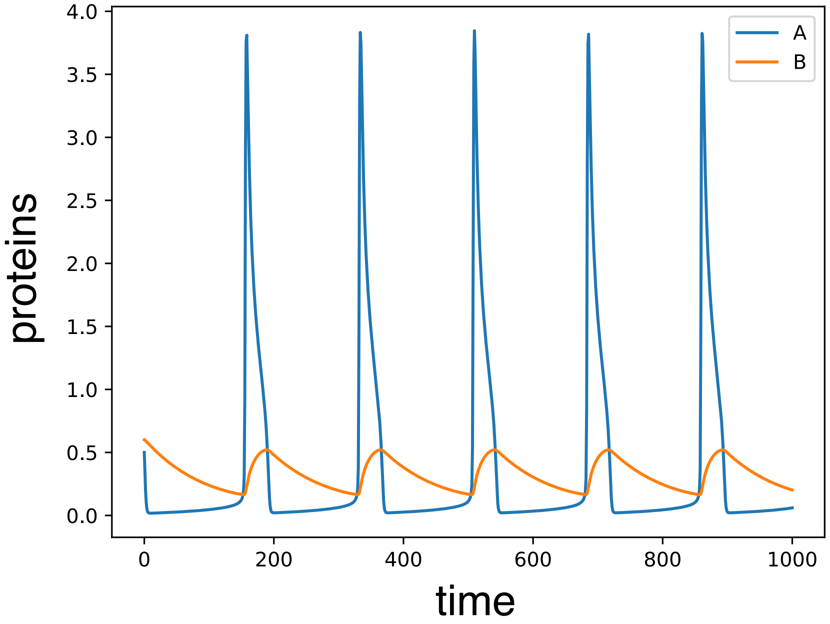
Krishna, Semsey, Jensen 2009

With a slowly varying B, there is a limit cycle.

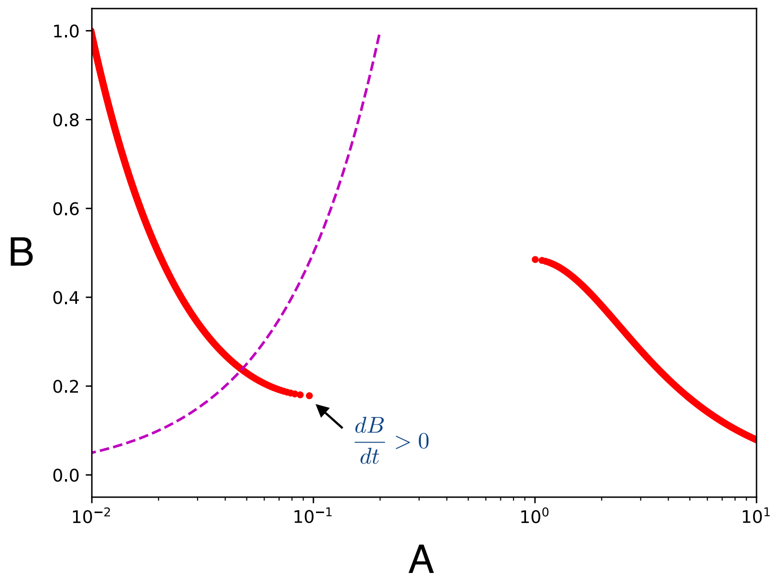


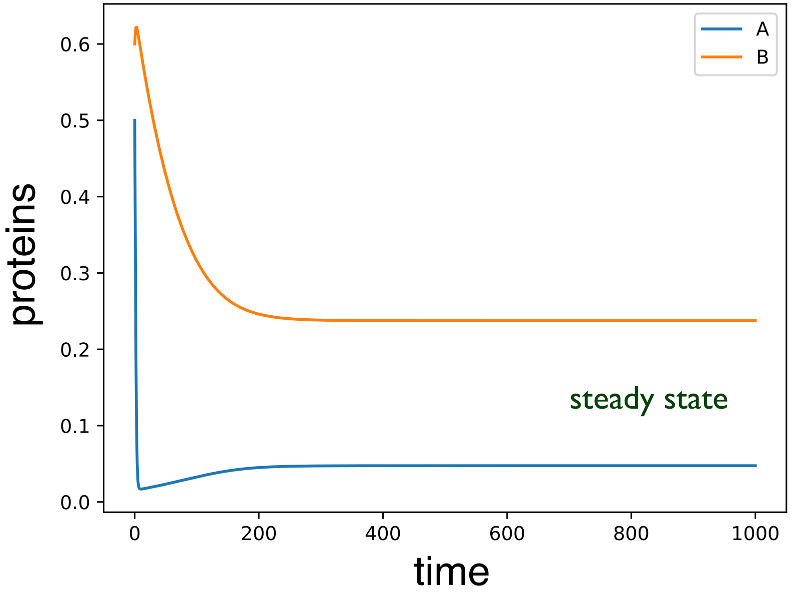
Relaxation oscillations:
Near the former low steady states, B's slower lifetime drives the dynamics;
near the former high steady states, A's faster lifetime drives the dynamics.

$$d_B \ll d_A$$



When B does not destabilise, the attractor is a steady state.





Stochastic gene expression

All chemical reactions are affected by thermal fluctuations and so are stochastic

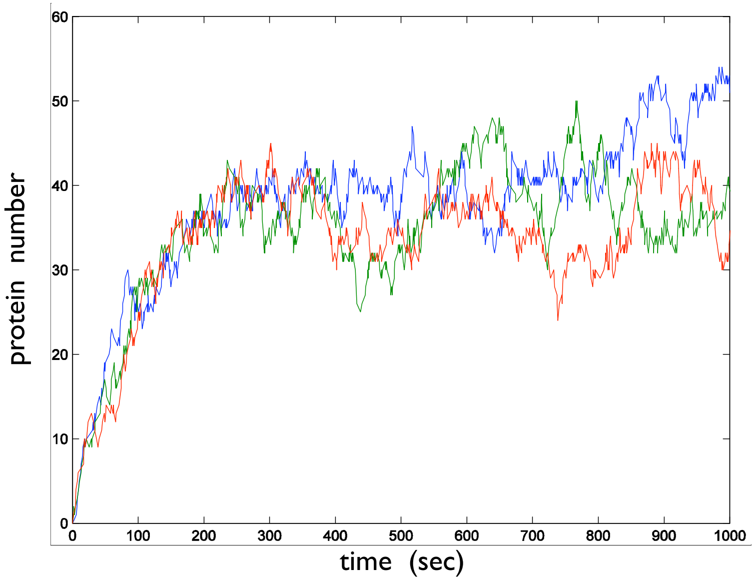
$$A + B \longrightarrow C$$

1. Reactants diffuse to find each other in solution
2. They must overcome the energy barrier of the reaction

Both events are randomly affected by thermal fluctuations – collisions with other molecules.

If the numbers of molecules are sufficiently large, then the mean number of molecules, or more correctly the mode, approximately obeys the appropriate chemical rate equations.

How should we quantify stochasticity?

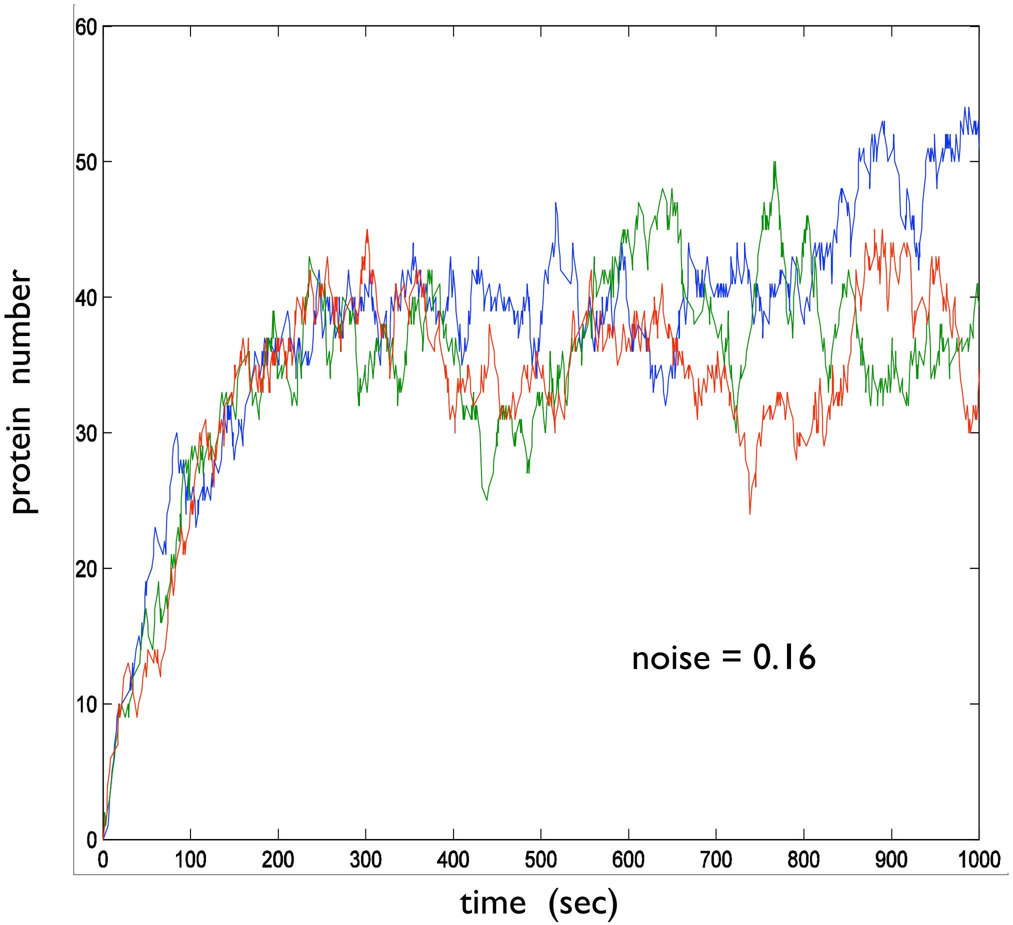


Noise is often defined as the coefficient of variation – the typical size of a fluctuation relative to the mean:

$$\boxed{\text{noise} = \frac{\text{standard deviation}}{\text{mean}}}$$

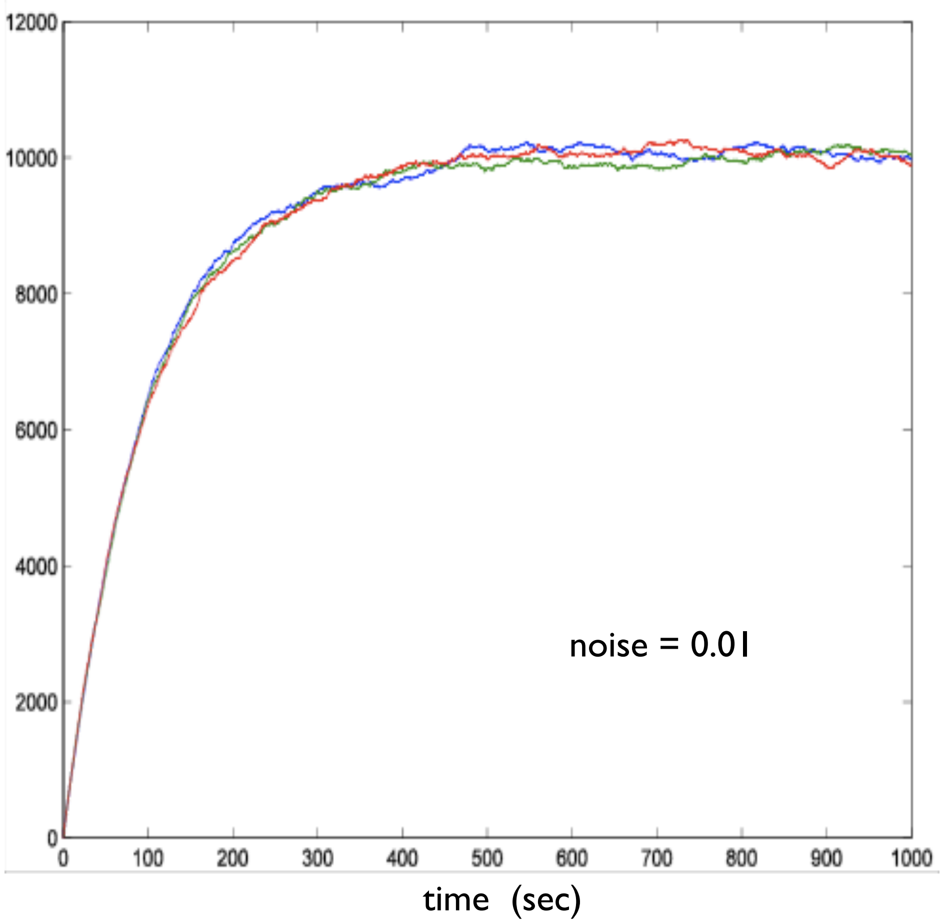
Stochasticity is more substantial at low numbers. Why?

mean 40



noise = 0.16

mean 10,000



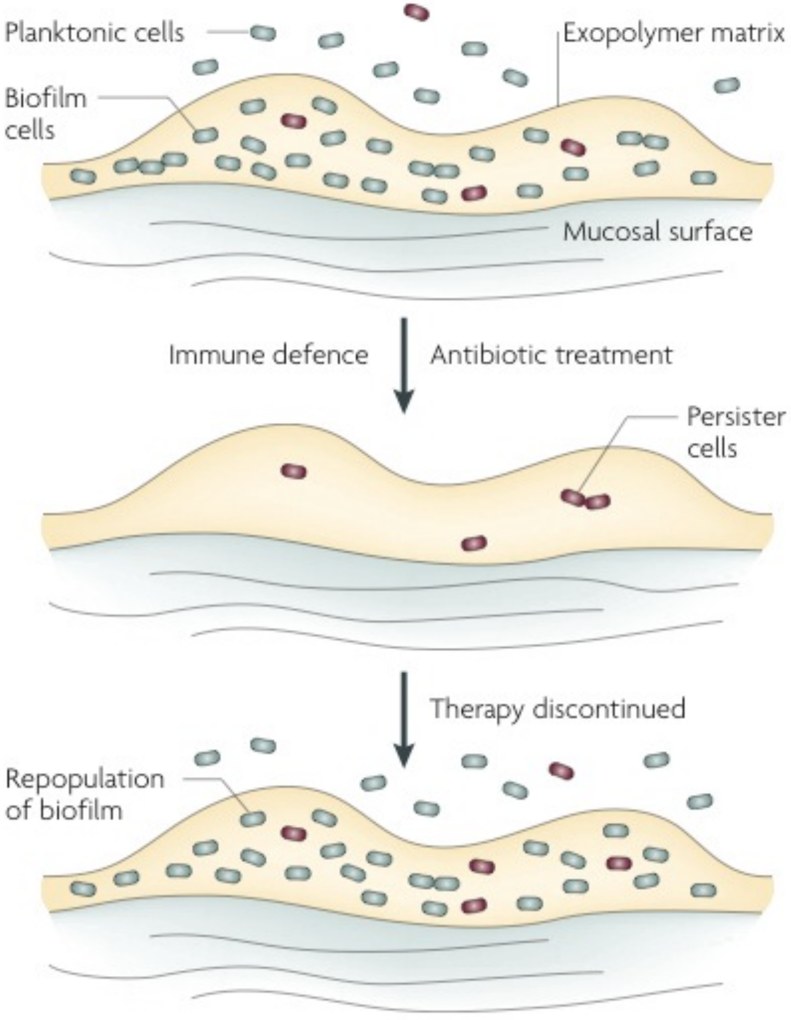
noise = 0.01

Why is stochasticity only substantial when typical numbers of molecules are low?

As a reaction changes the number of molecules by one or two, it is only when numbers are small that stochasticity – the random timing of individual reactions – matters.

Stochasticity can be exploited: persister cells enable a population to be both invasive and tolerant to drugs

probability of
being a
persister is less
than 10^{-5}
for *E. coli*

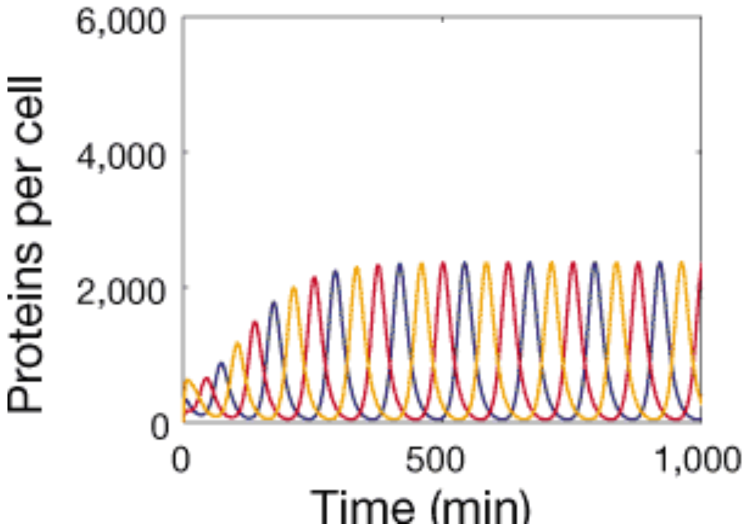


Lewis, *Nat Rev Microbiol* 2007

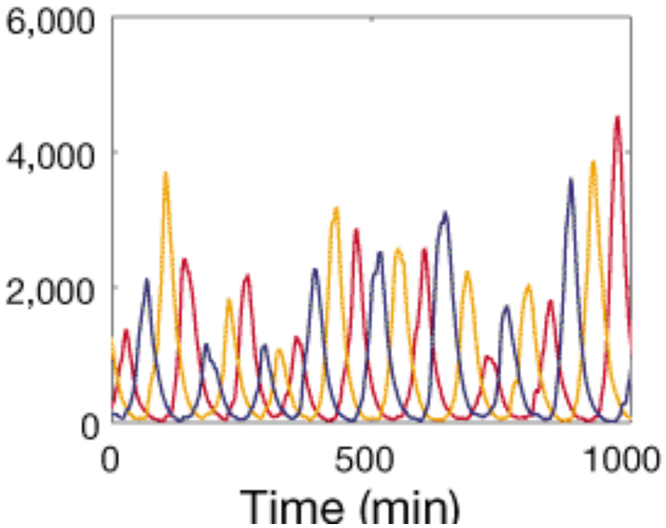
Stochasticity affects the reliability of biochemical networks by affecting timing and is therefore regulated away

e.g. biological rhythms

deterministic



stochastic



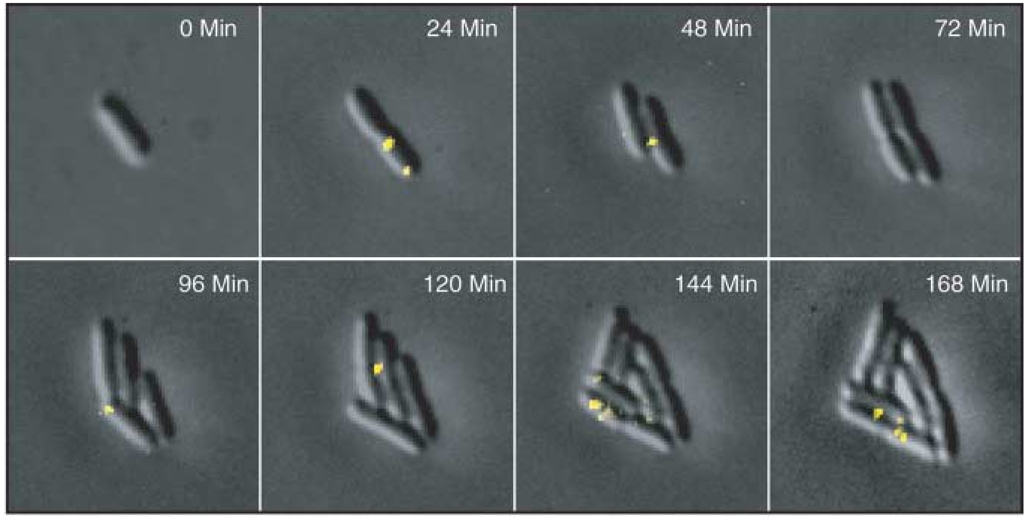
Elowitz & Leibler, *Nature* 2000
Barkai & Leibler, *Nature* 2000

Stochasticity is generated during gene expression: translation can occur in bursts

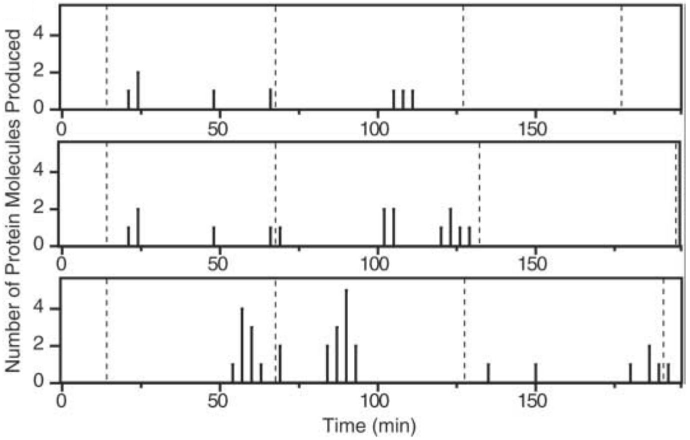
Probing Gene Expression in Live Cells, One Protein Molecule at a Time

Ji Yu,[1*] Jie Xiao,[1*] Xiaojia Ren,[1] Kaiqin Lao,[2] X. Sunney Xie[1†]

Following expression of a fluorescent membrane protein in bacteria over time.



Occasionally, one mRNA is transcribed.



Bursts of translated protein.

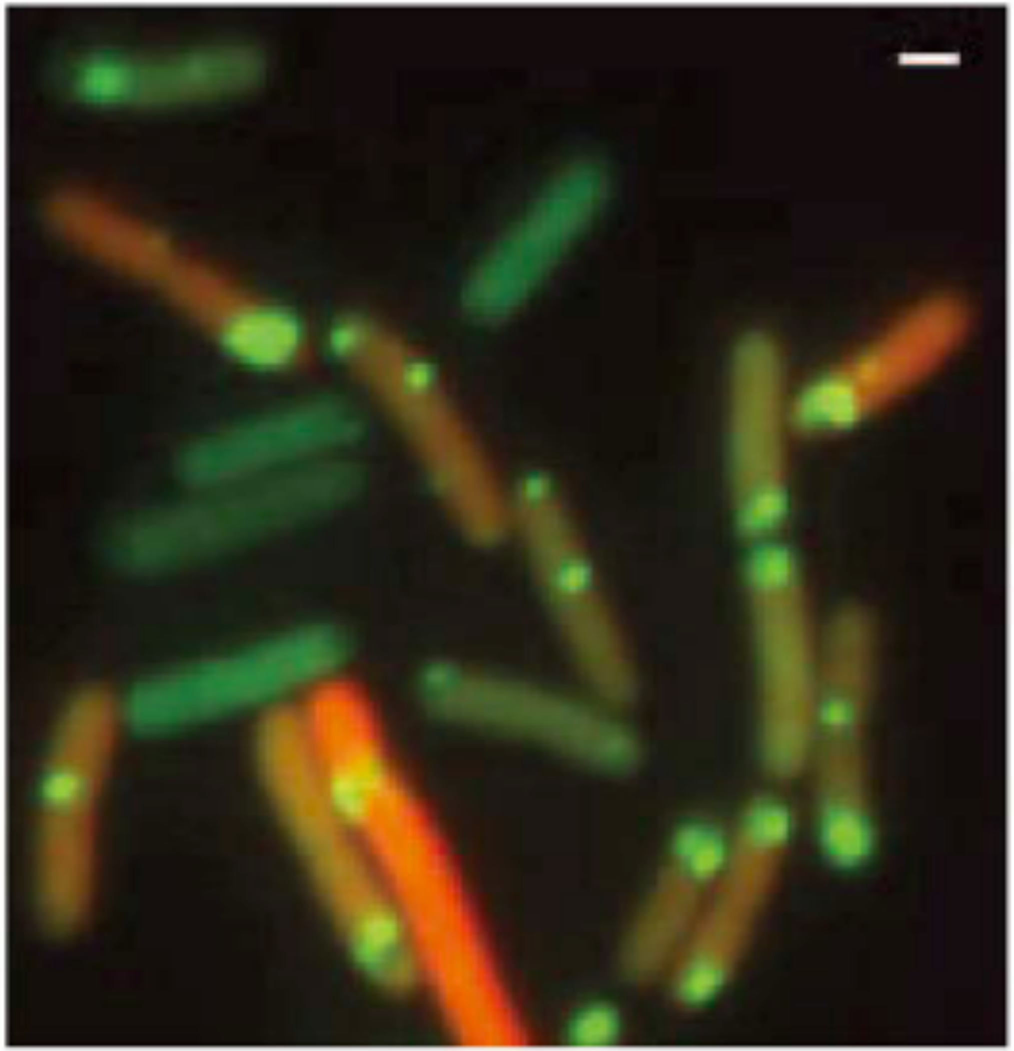
Yu *et al.*, Science (2006)

Transcription can also occur in bursts

Real-Time Kinetics of Gene Activity in Individual Bacteria

Ido Golding,[1,*] Johan Paulsson,[2,3] Scott M. Zawilski,[1] and Edward C. Cox[1,*]

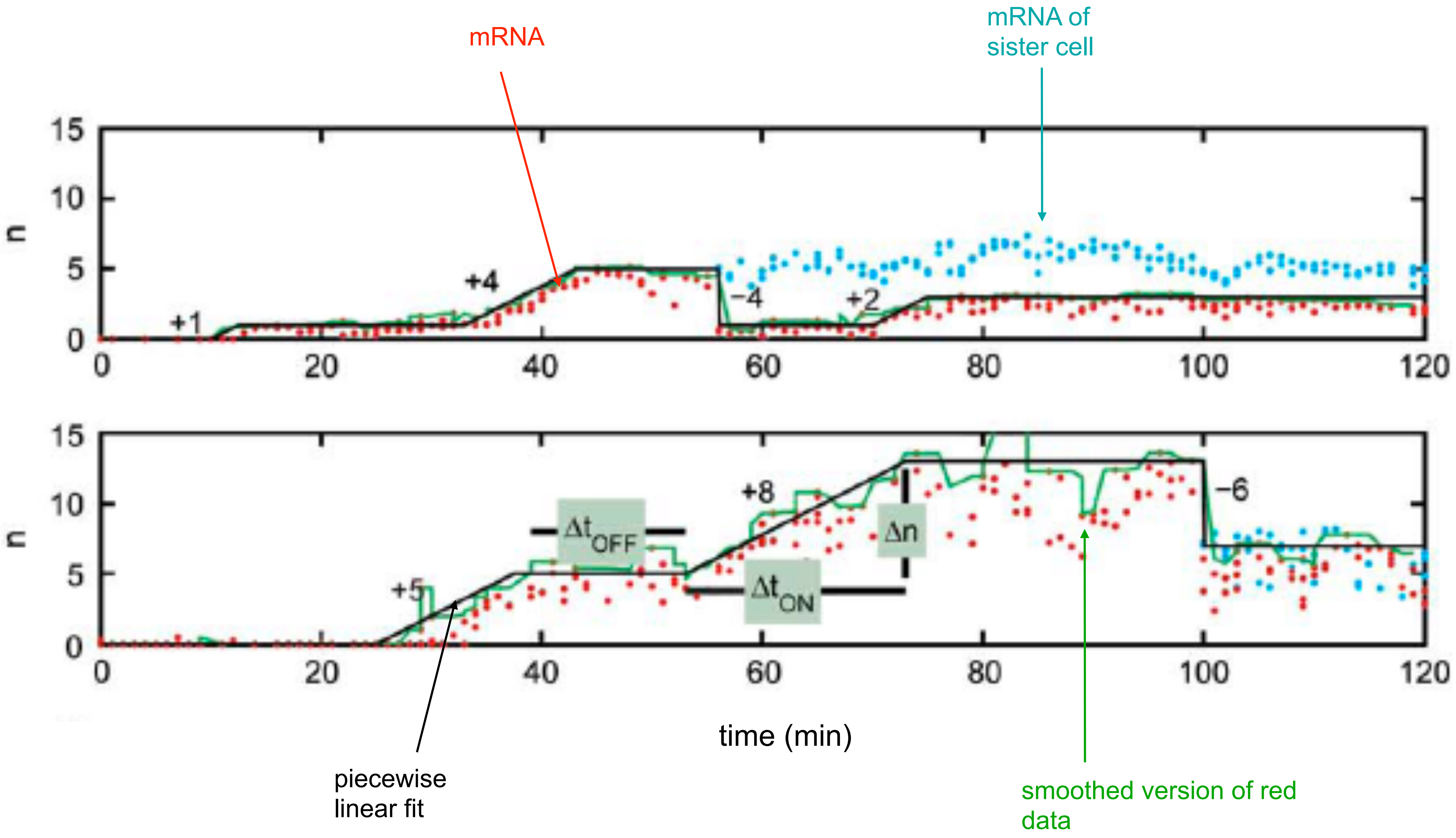
Cell *123*, 1025–1036, December 16, 2005



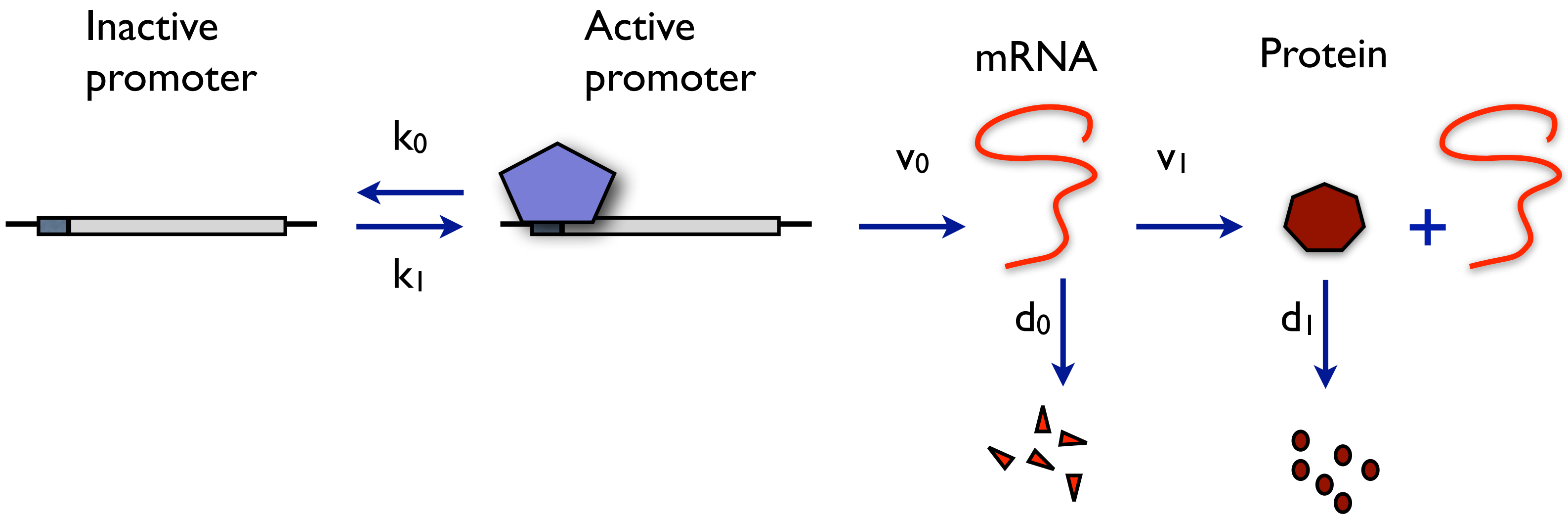
Red: protein
Green spots: mRNA

scale bar: 1 µm

Time course of mRNA numbers: mRNA is produced in bursts



The most common model of gene expression has both bursts in transcription and translation



Inactive promoter

k_0

k_1

Active promoter

v_0

mRNA

d_0

v_1

Protein

d_1

+

Kaern et al., *Nat Rev Genetics* 2005

To perform stochastic simulations, we typically use the Gillespie, or stochastic simulation, algorithm

Step 1: choose which reaction will occur

Step 2: choose when that reaction will occur

Example: an elementary model of gene expression

1: $\xrightarrow{k} A$

2: $A \xrightarrow{d} \phi$

probability of a reaction in time δt

$$a_1 \delta t = k\, \delta t$$

propensity of reaction 1

$$a_2 \delta t = dA\, \delta t$$

propensity of reaction 2

probability of no reaction

$$P_0(t + \delta t) = P_0(t)\Big[1 - (a_1 + a_2)\delta t\Big] \quad \text{hence} \quad P_0 \sim \mathrm{e}^{-(a_1 + a_2)t}$$

probability of a reaction *i* at time $t + \delta t$

$$P_i(t)\delta t = P_0(t) a_i \delta t$$

Gillespie, *J Phys Chem* 1977