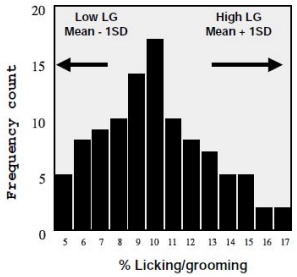
dynamic processes in cells (a systems approach to biology)

jeremy gunawardena
department of systems biology
harvard medical school

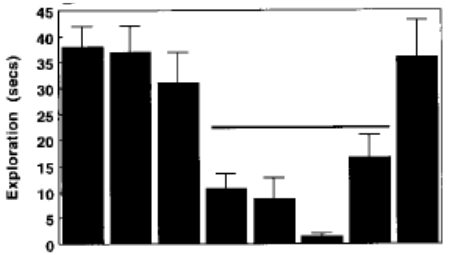
lecture 8

1 october 2015

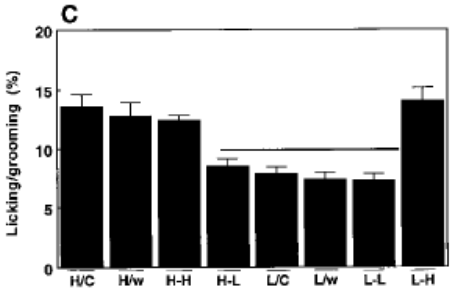
cellular identity can be re-programmed – III



adult female offspring

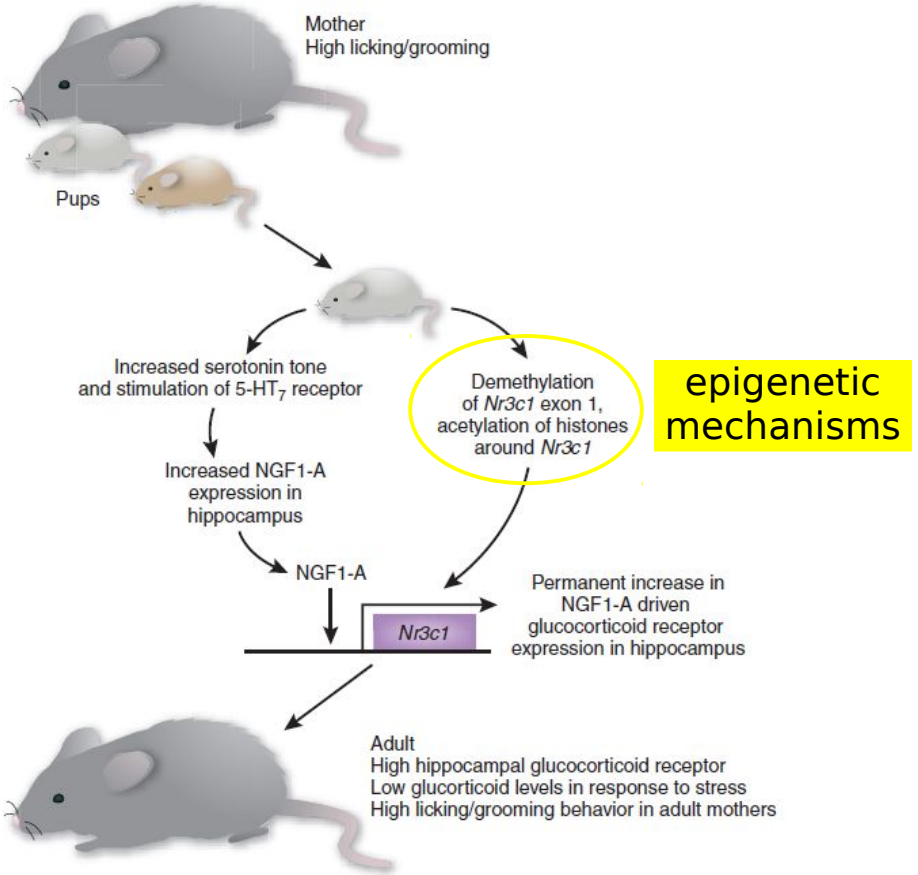


open field exploration



licking/grooming

cross-fostering group

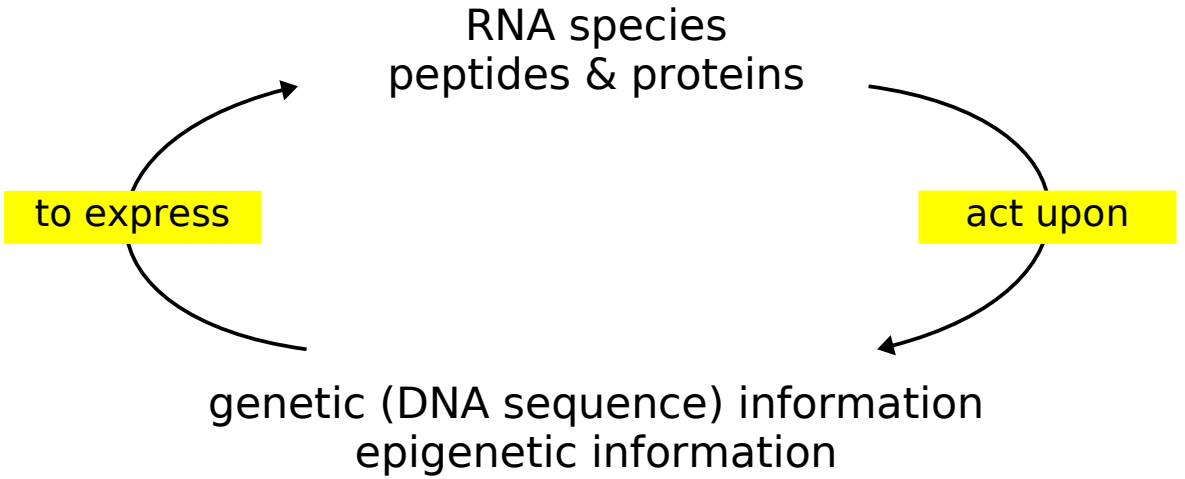


Francis, Diorio, Liu, Meaney, *"Nongenomic transmission across generations of maternal behaviour and stress responses in the rat"*, Science **286**:1155-8 1999; Szyf, Weaver, Champagne, Diorio, Meaney, *"Maternal programming of steroid receptor expression and phenotype through DNA methylation in the rat"*, Frontiers in Neuroendocrinology **26**:139-62 2005

molecular basis of cellular identity

how does a single genome give rise to many different cellular identities?

cells create different stable states of feedback between inherited genetic and epigenetic information and patterns of molecular expression from DNA

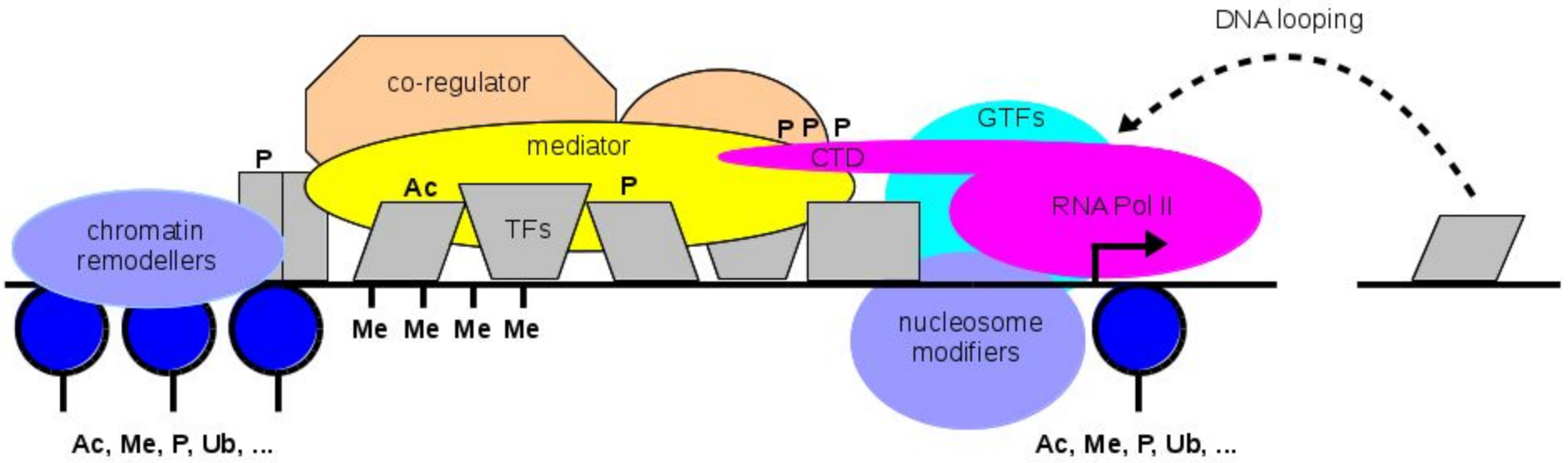


and these states can be stably inherited by daughter cells

Jacques Monod, Francois Jacob, *"Genetic regulatory mechanisms in the synthesis of proteins"*, J Mol Biol **3**:318-56 1961

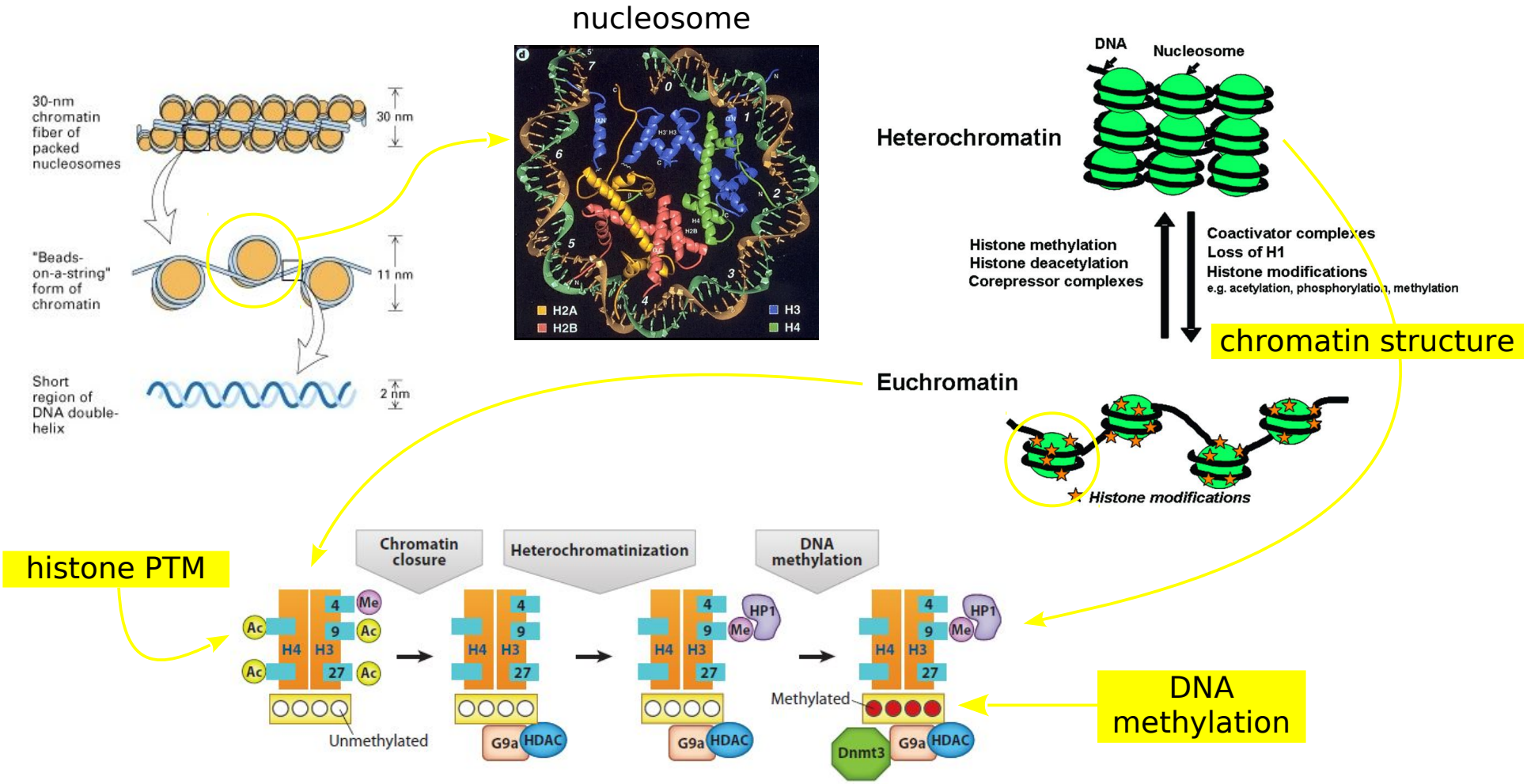
gene expression in eukaryotes

expression of genetic information is regulated by **weak linkage**



Ptashne & Gann, **Genes & Signals**, Cold Spring Harbor Lab Press, 2002;
Levine, Cattoglio & Tjian, "*Looping back to leap forward: transcription enters a new era*", Cell **157**:13-25 2014

epigenetic information in eukaryotes



Bergmann, *"Programming of DNA methylation patterns"*, Annu Rev Biochem, **81**:97-117, 2012

gene regulation in the linear framework

Ahsendorf *et al. BMC Biology* (2014) 12:102
DOI 10.1186/s12915-014-0102-4

BMC Biology

RESEARCH ARTICLE Open Access

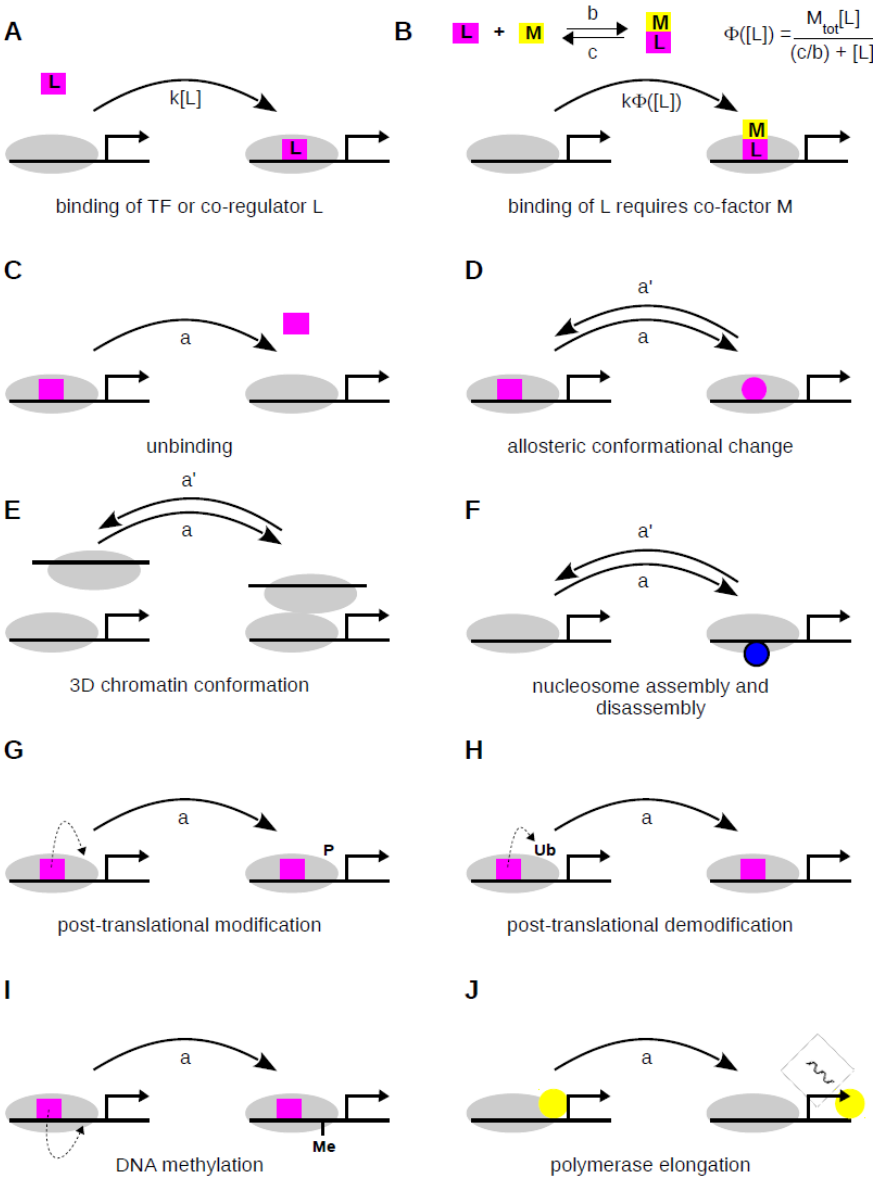
A framework for modelling gene regulation which accommodates non-equilibrium mechanisms

Tobias Ahsendorf[1,3†], Felix Wong[2,3†], Roland Eils[1,4] and Jeremy Gunawardena[3*]

BMC Biology **12**:102 2014

vertex = "snapshot" of DNA state or "microstate"

edge = transition between states



calculating gene expression

$$\rho \in \ker \mathcal{L}(G) \qquad \frac{\rho_\mu}{\sum_\mu \rho_\mu}$$ probability of microstate μ

each microstate has a characteristic rate of gene expression and the overall rate is proportional to the average over all microstates

$$\frac{d[\mathrm{mRNA}]}{dt} = \frac{\sum_\mu r_\mu \rho_\mu}{\sum_\mu \rho_\mu}$$

this assumption separates regulation from the stochasticity ("bursting", etc) of RNA polymerase

regulation and transcription should really be treated together (*)

Ackers, Johnson, Shea, PNAS, **79**:1129-33 1982

(*) Brown, Mao, Falkovskaia, Jurica, Boeger, "*Linking stochastic fluctuations in chromatin structure and gene expression*", PLoS Biol **11**:e1001621 2013;
Sanchez, Garcia, Jones, Phillips, Kondev, "*Effect of promoter architecture on the cell-to-cell variability in gene expression*", PLoS Comp Biol **7**:e1001100 2011

detailed balance at thermodynamic equilibrium

if the system reaches **thermodynamic equilibrium**, then **detailed balance** holds and ρ can be calculated in a particularly simple way

principle of detailed balance (in the linear framework): every edge in the graph has a complementary reverse edge and, in any steady state, each pair of such reversible edges is independently at steady state, irrespective of any other edges reaching those vertices

> *"For if this were not the case we could add a minute amount of some catalyst which would increase the rate of the reaction and its inverse along one of the paths, without affecting the two rates in the other path. This would disturb the existing equilibrium, contrary to the results of observation and of thermodynamics."* (*)

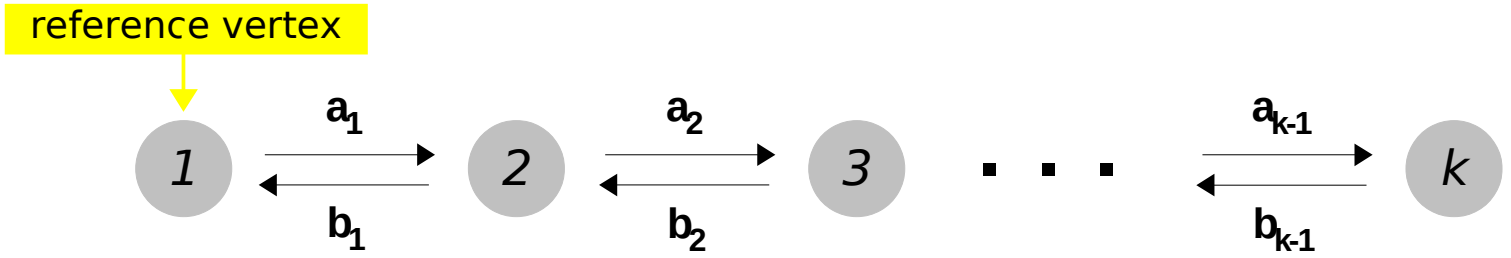
detailed balance is a consequence of **microscopic reversibility**: the fundamental laws of physics, whether classical newtonian mechanics or quantum mechanics, exhibit time-reversal symmetry (**+**)

(*) Gilbert Lewis, *"A new principle of equilibrium"*, PNAS **11**:179-83 1925

(**+**) Bruce Mahan, *"Microscopic reversibility and detailed balance"*, J Chem Edu **52**:299-302 1975

calculating ρ at equilibrium

choose any path of reversible edges from a reference vertex to a given vertex, k



suppose given a steady state of the Laplacian dynamics $x^* \in \ker \mathcal{L}(G)$

then, by detailed balance applied to each reversible pair of edges,

$$x_2^* = \left(\frac{a_1}{b_1}\right) x_1^* \qquad x_3^* = \left(\frac{a_2}{b_2}\right) x_2^* \qquad \cdots \qquad x_k^* = \left(\frac{a_{k-1}}{b_{k-1}}\right) x_{k-1}^*$$

$$x_k^* = \left(\frac{a_1}{b_1}\right)\left(\frac{a_2}{b_2}\right)\cdots\left(\frac{a_{k-1}}{b_{k-1}}\right) x_1^*$$

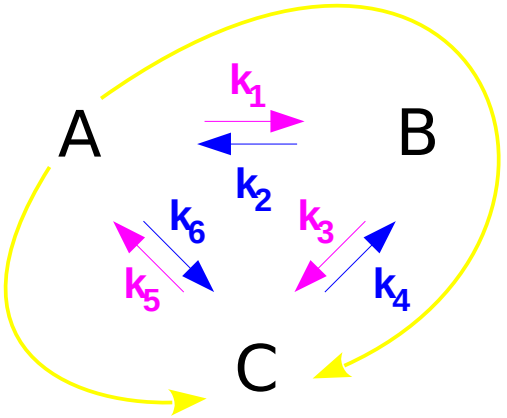
so we can take

$$\rho_k = \left(\frac{a_1}{b_1}\right)\left(\frac{a_2}{b_2}\right)\cdots\left(\frac{a_{k-1}}{b_{k-1}}\right) \qquad \rho \in \ker \mathcal{L}(G)$$

the cycle condition

for this construction to work, the result must be independent of the chosen path

$$[C] = \left(\frac{k_6}{k_5}\right)[A]$$



$$[C] = \left(\frac{k_1}{k_2}\right)\left(\frac{k_3}{k_4}\right)[A]$$

$$k_1 k_3 k_5 = k_2 k_4 k_6$$

the rate constants of a system that can reach equilibrium cannot be chosen arbitrarily but are constrained by the cycle condition: in any cycle, the product of the rate constants going clockwise equals the product going counterclockwise.

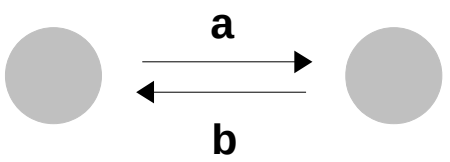
Gunawardena, *"A linear framework for time-scale separation in nonlinear biochemical systems"*, PLoS ONE **7**:e36321 2012

equivalent to equilibrium statistical mechanics

the **van't Hoff equation** links the two methods

1852-1911

change in Gibbs free energy



$$\ln\left(\frac{a}{b}\right) = -\frac{\Delta G}{RT}$$

so, for any microstate, k

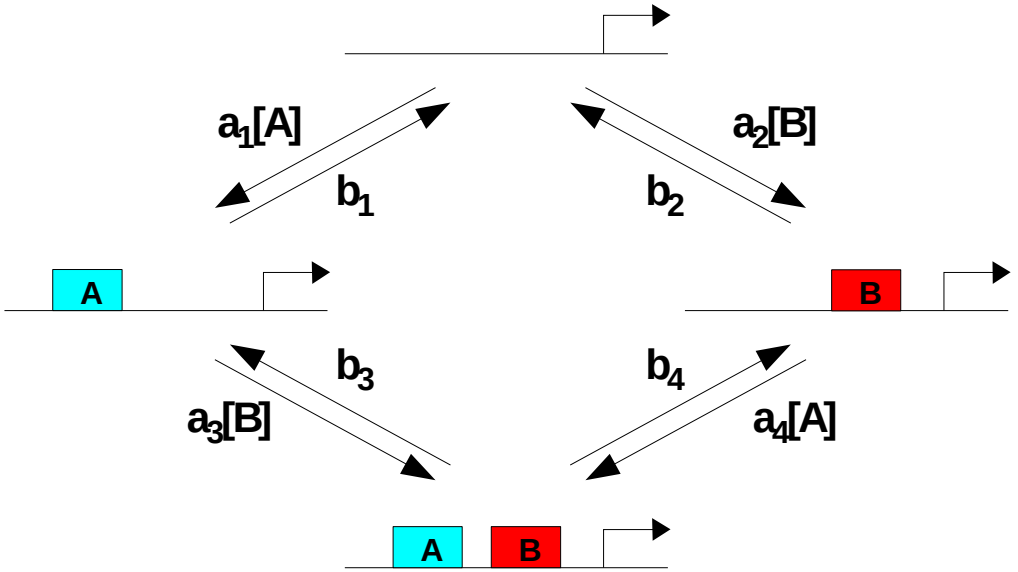
$$\ln \rho_k = -\sum \text{(interaction energies)/RT}$$

TF-DNA, TF-TF, TF-RNAP, ...

the partition function is given by $\sum_{\mu} \rho_{\mu}$

Bintu et al, *"Transcriptional regulation by the numbers"*, Curr Opin Gen Dev, **15**:116-24 & 125-35, 2005; Segal, Widom, *"From DNA sequence to transcription behaviour"*, Nat Rev Genetics **100**:443-56 2009; Sherman, Cohen, *"Thermodynamic state-ensemble models of cis-regulation"*, PLoS Comp Biol **8**:e1002407 2012.

example – two transcription factors



$$K_1 = \left(\frac{a_1}{b_1}\right) \qquad K_2 = \left(\frac{a_2}{b_2}\right)$$

$$K_3 = \left(\frac{a_3}{b_3}\right) \qquad K_4 = \left(\frac{a_4}{b_4}\right)$$

$$a_1[A]a_3[B]b_4b_2 = a_2[B]a_4[A]b_3b_1$$

cycle condition

$$K_1K_3 = K_2K_4$$

$$\rho = \begin{pmatrix} 1 \\ K_1[A] \\ K_2[B] \\ K_1K_2\omega[A][B] \end{pmatrix}$$

$$\frac{K_4}{K_1} = \frac{K_3}{K_2} = \omega$$

cooperativity

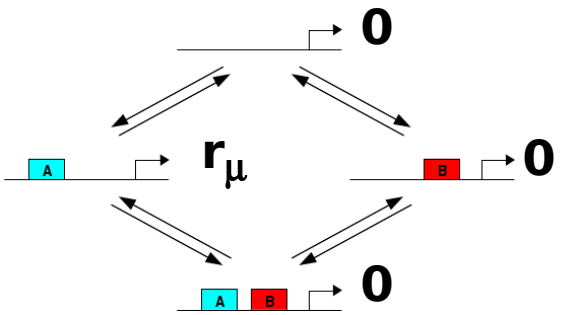
$\omega < 1$	negative cooperativity
$\omega = 1$	independence
$\omega > 1$	positive cooperativity

gene regulation function

$$\frac{d[\text{mRNA}]}{dt} = \frac{\sum_\mu r_\mu \rho_\mu}{\sum_\mu \rho_\mu} \qquad \rho \in \ker \mathcal{L}(G)$$

need to specify r_μ which determines transcription in microstate μ

eg: A is an activator and B is a perfect dominant repressor –



$$\frac{d[\text{mRNA}]}{dt} = \frac{r_{10} K_1[A]}{1 + K_1[A] + K_2[B] + K_1 K_2 \omega [A][B]}$$

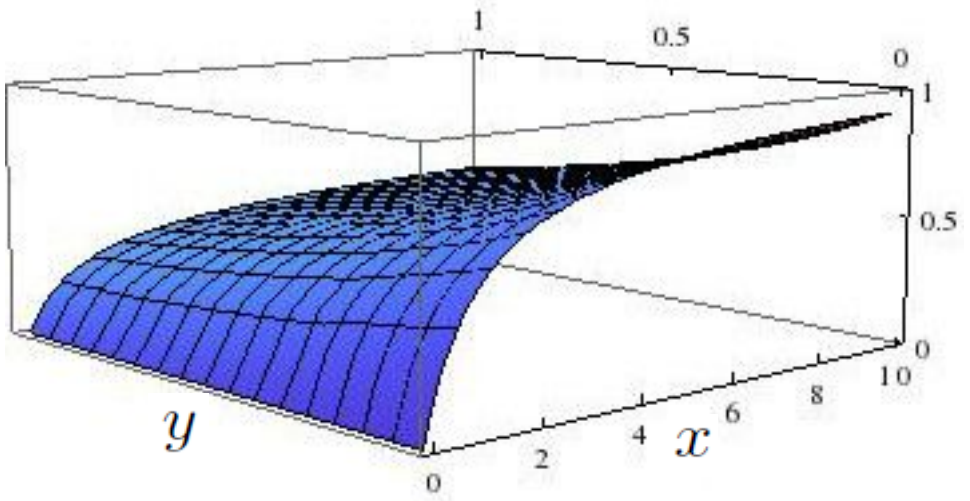
partition function

rational expression in the transcription factor concentrations

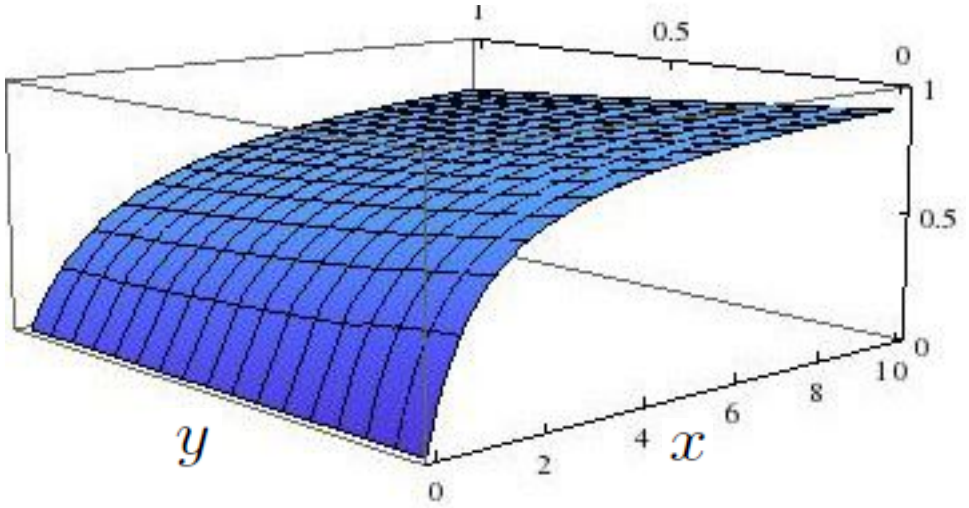
gene regulation function – impact of cooperativity

$r_{10} = 1 \quad x = K_1[A] \quad y = K_2[B]$

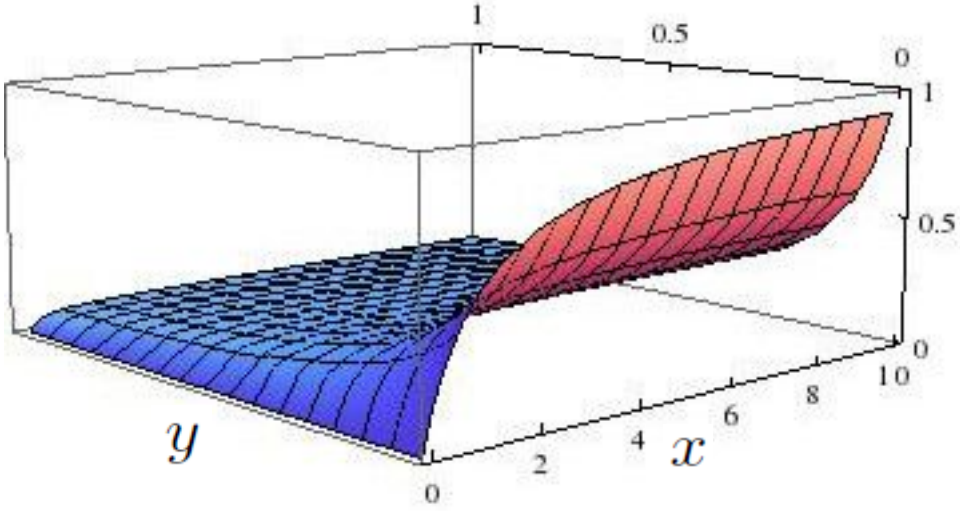
$\omega = 1$



$\omega = 0.1$

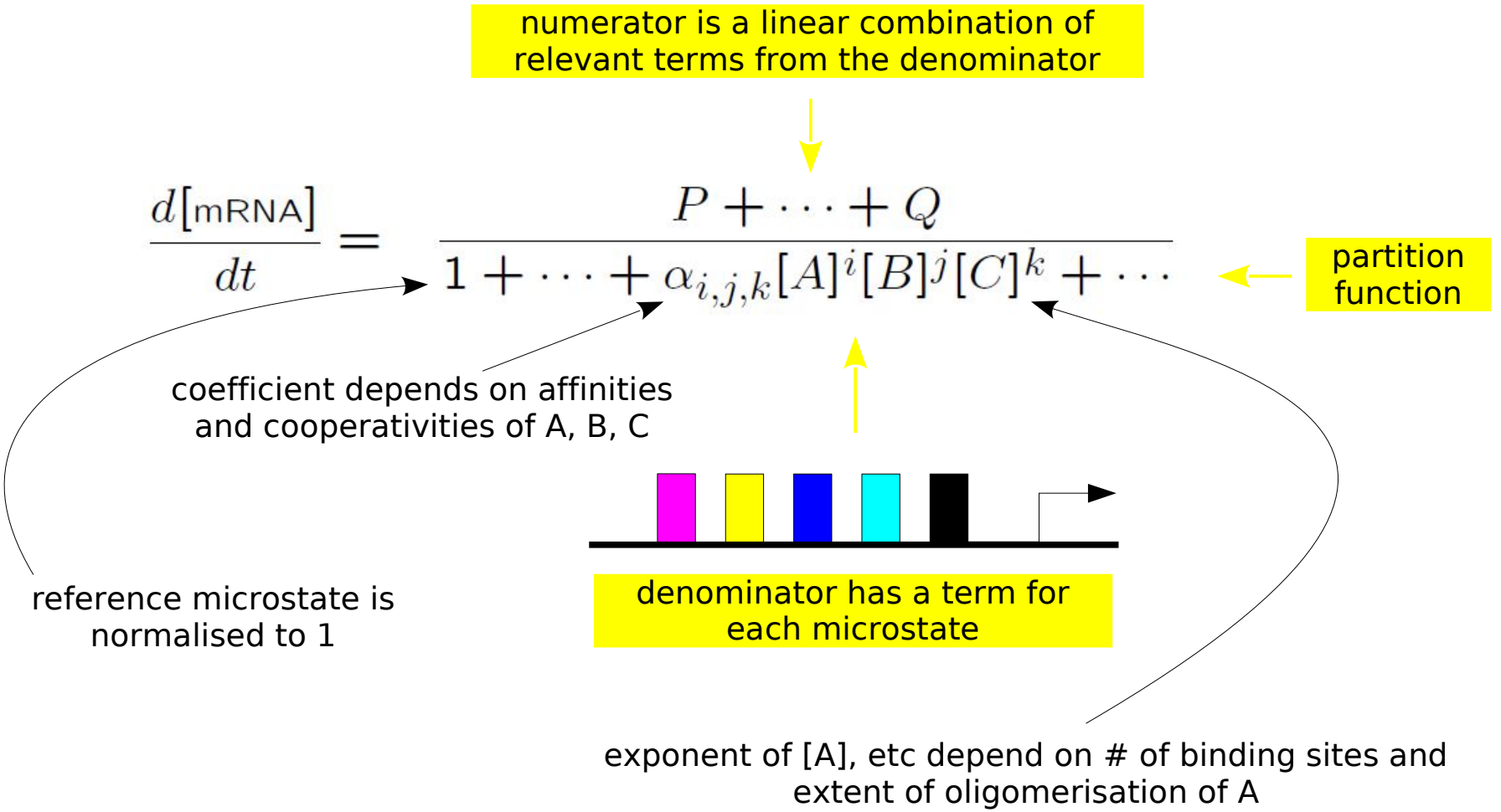


$\omega = 10$



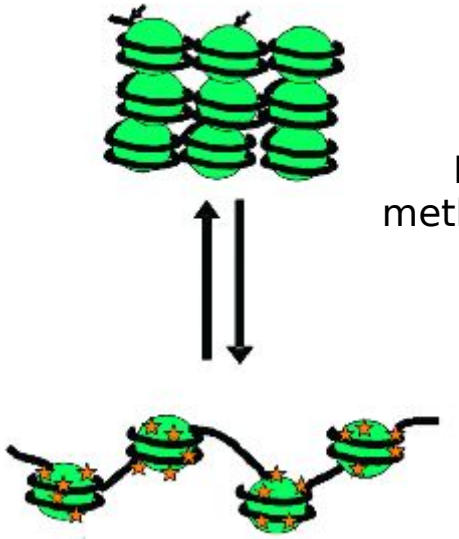
structure of equilibrium gene regulation functions

GRFs are rational expressions in the TF concentrations



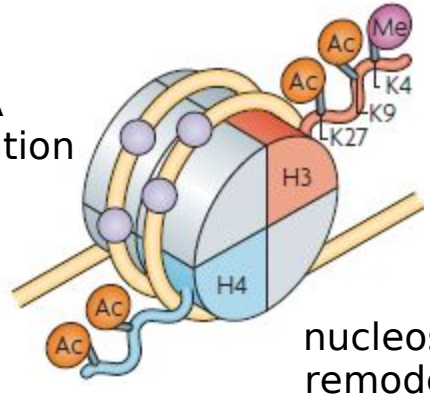
energy dissipation away from equilibrium

heterochromatin



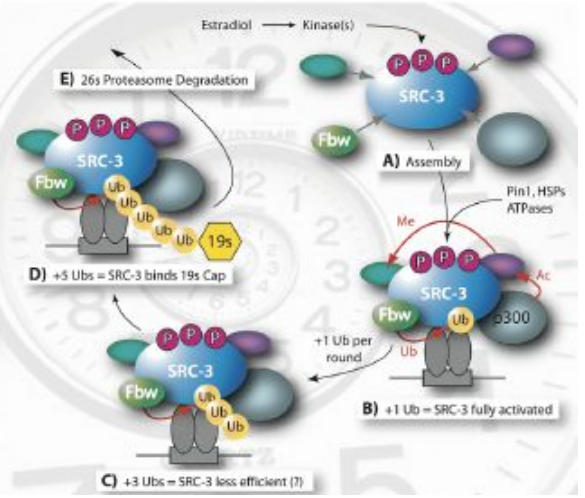
euchromatin

DNA methylation



histone PTM

nucleosome remodelling



PTM of TFs and co-regulators

Hopfield, *"Kinetic proofreading: a new mechanism for reducing errors in biosynthetic processes requiring high specificity "*, PNAS **71**:4135-9 1974

thermodynamic equilibrium sets an upper bound to how well information processing tasks can be undertaken by a biochemical system and the only way to exceed this bound is to dissipate energy and maintain the system away from equilibrium

calculating ρ away from equilibrium

Matrix-Tree Theorem: whenever G is strongly connected

$$\ker \mathcal{L}(G) = \langle \rho \rangle \quad \rho_i = \sum_{T \in \Theta_i(G)} \left(\prod_{j \xrightarrow{a} k \in T} a \right)$$

positive

$\Theta_i(G) =$ set of spanning trees rooted at *i*

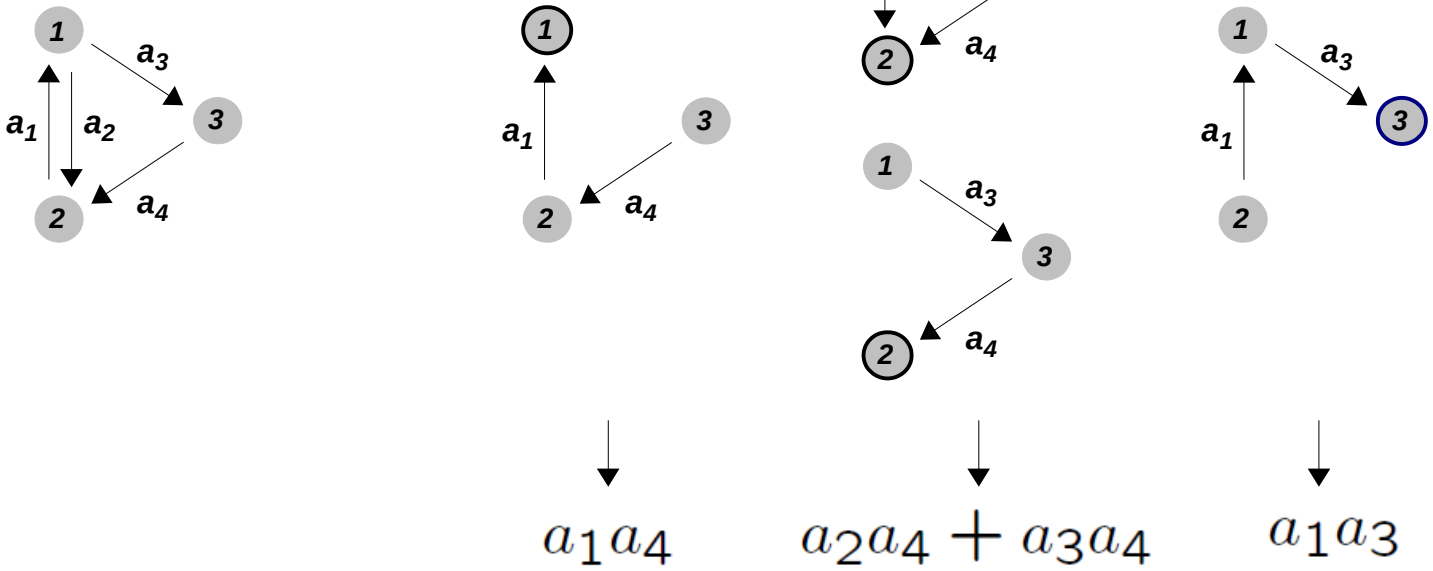
rooted spanning tree – a sub-graph T of G which

SPANS G	–	every node of G is also a node of T
is a **TREE**	–	T has no cycles, ignoring edge directions
is **ROOTED** at *i*	–	*i* is the only node of T with no outgoing edges

Bill Tutte, *"The dissection of equilateral triangles into equilateral triangles"*, Proc Camb Phil Soc **44**:463-82 1948

Mirzaev & Gunawardena, "*Laplacian dynamics on general graphs*", Bull Math Biol **75**:2118-49 2013 – Appendix

spanning tree formula



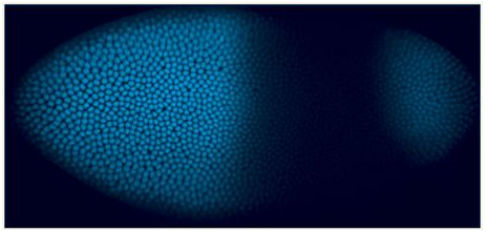
$$a_1 a_4 \qquad a_2 a_4 + a_3 a_4 \qquad a_1 a_3$$

$$\begin{pmatrix} -(a_2+a_3) & a_1 & 0 \\ a_2 & -a_1 & a_4 \\ a_3 & 0 & -a_4 \end{pmatrix} \begin{pmatrix} a_1 a_4 \\ (a_2+a_3)a_4 \\ a_1 a_3 \end{pmatrix} = 0$$

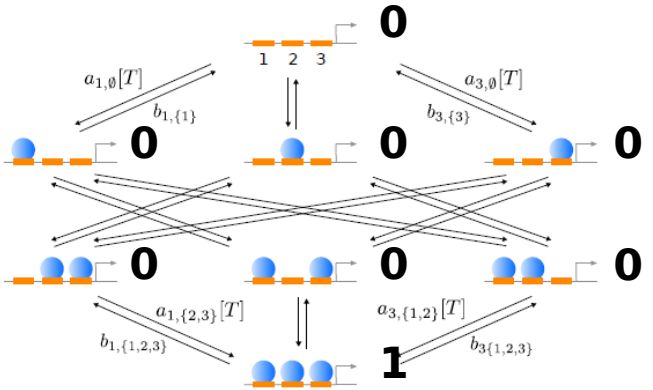
Laplacian matrix ρ

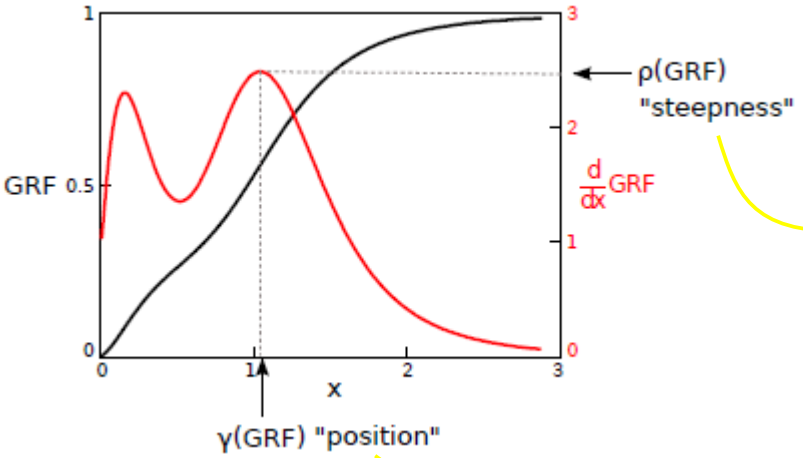
a Hopfield barrier for sharpness?

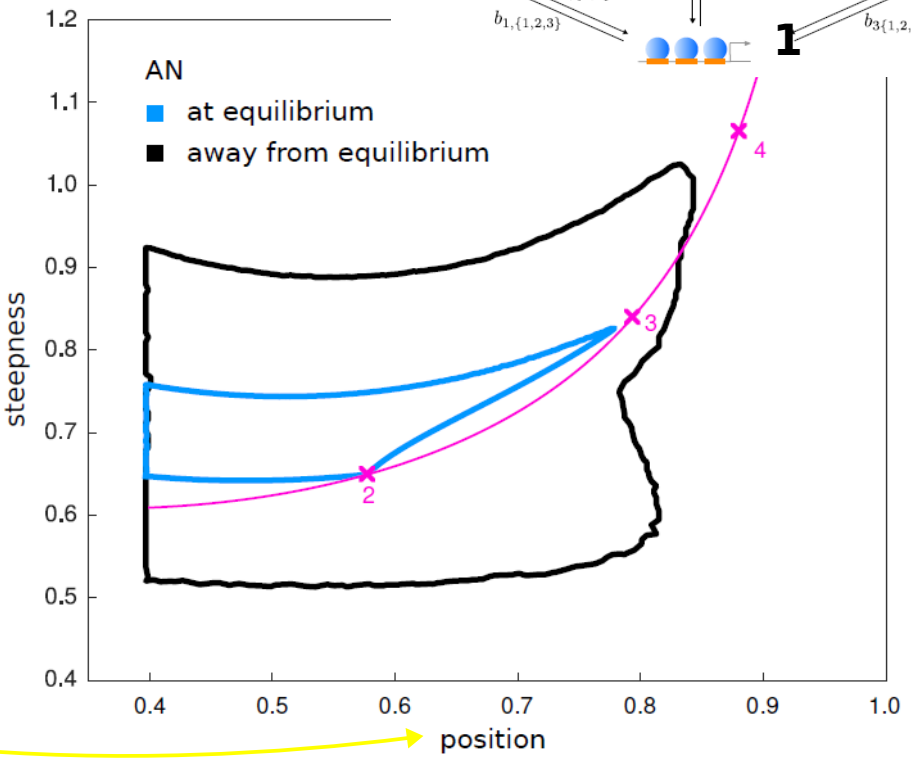
Hunchback expression in response to transcription factor Bicoid



TF binding at 3 sites with all-or-nothing expression

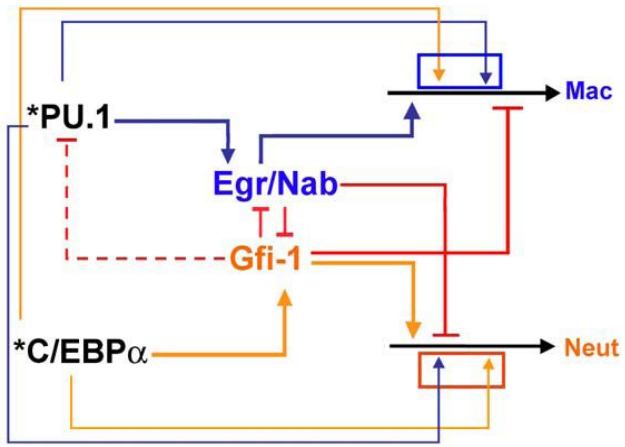


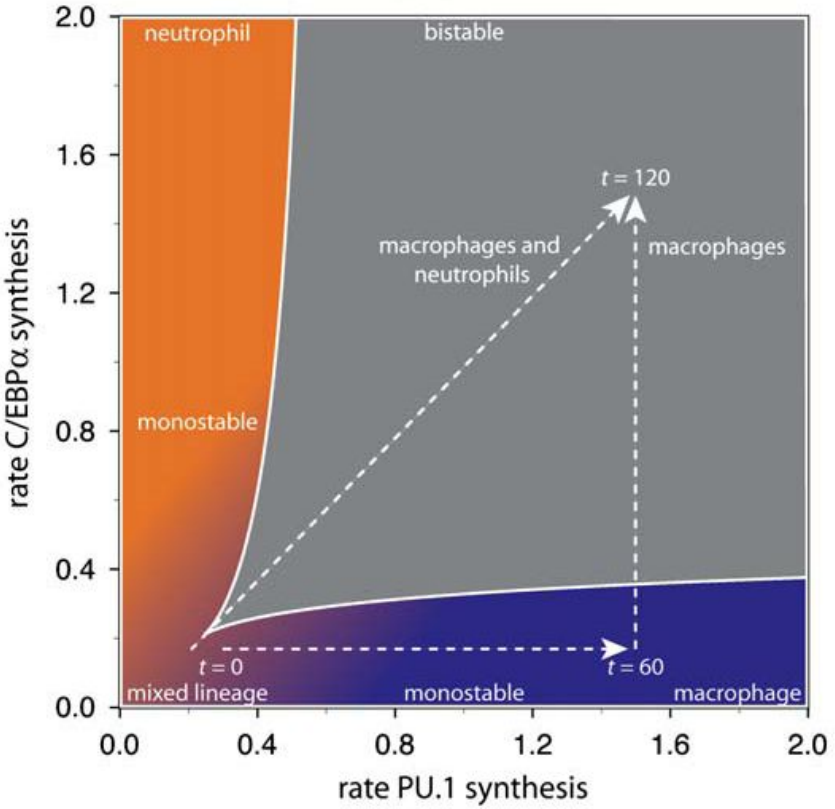




Estrada, Wong, DePace, Gunawardena, *"Higher-order cooperativity and energy dissipation can sharpen switching of eukaryotic genes"*, submitted, 2015

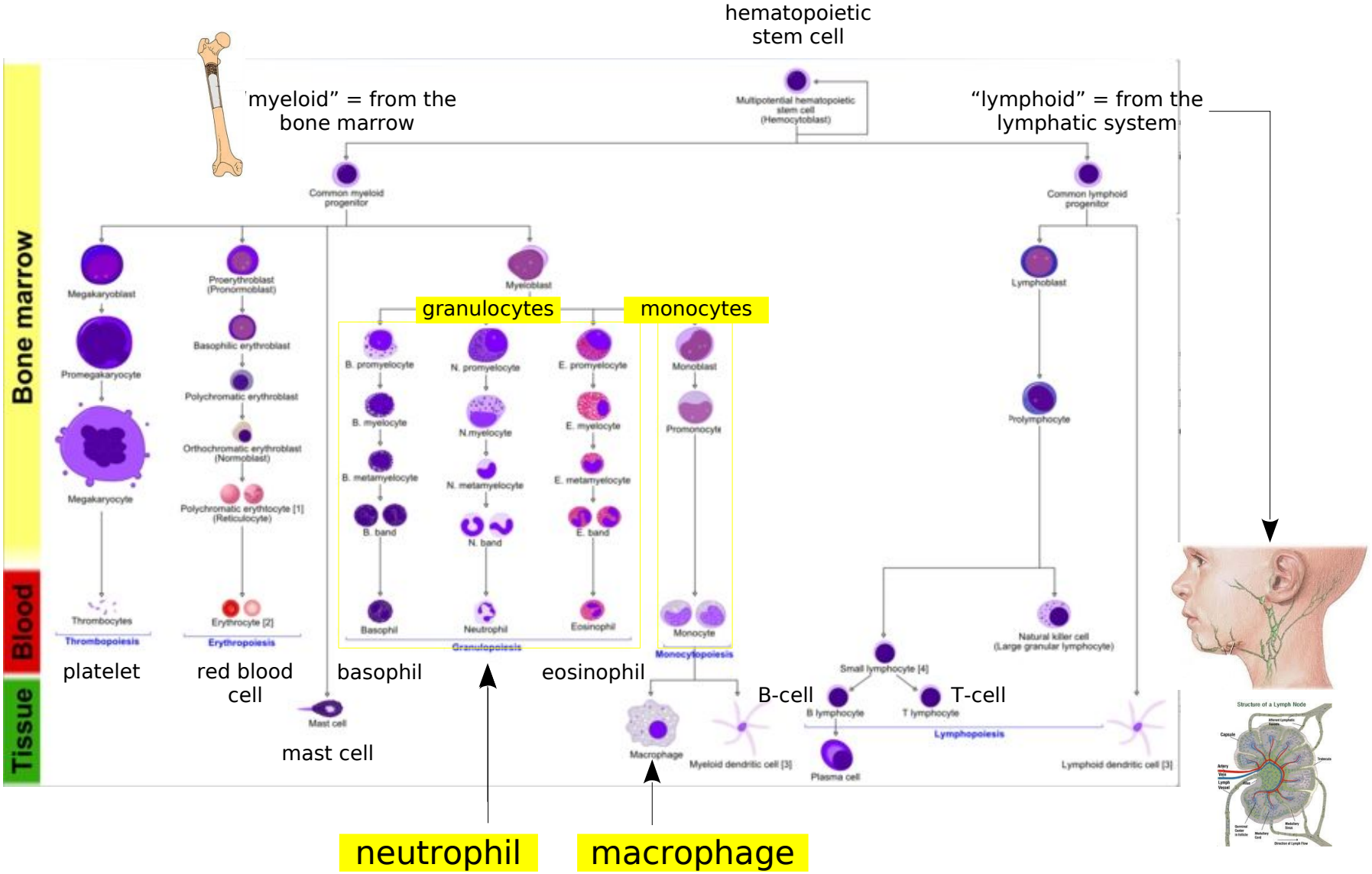
a gene regulation network (GRN) in hematopoiesis





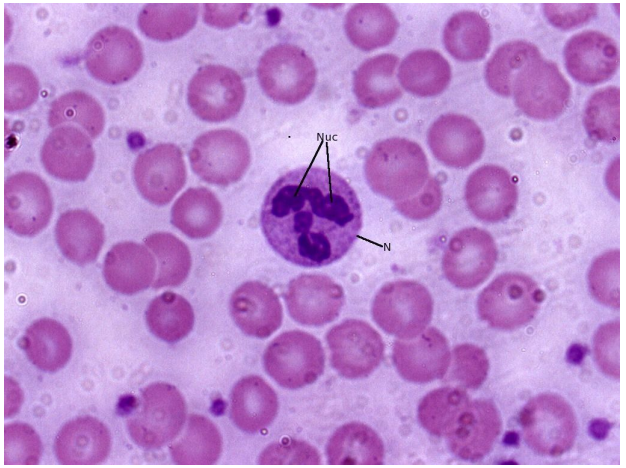
Laszlo, Spooner, Warmflash, Lancki, Lee, Sciammas, Gantner, Dinner, Singh, *"Multilineage transcriptional priming and determination of alternate hematopoietic cell fates"*, Cell **126**:755-66 2006

blood work


hematopoietic stem cell
"myeloid" = from the bone marrow
"lymphoid" = from the lymphatic system
Bone marrow
Blood
Tissue
granulocytes
monocytes
platelet
red blood cell
basophil
eosinophil
mast cell
B-cell
T-cell
neutrophil
macrophage

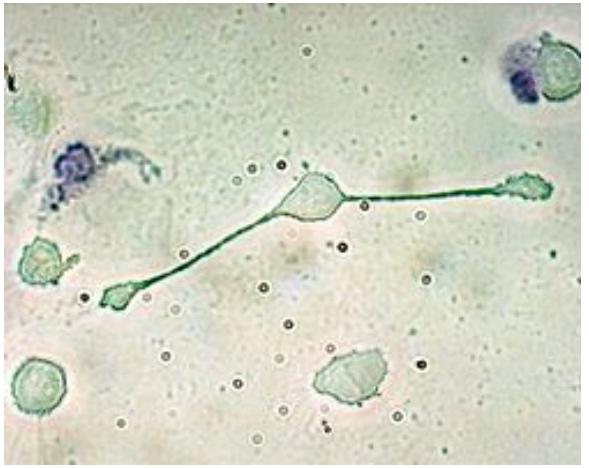
innate immunity – the first line of defense

neutrophil



- highly mobile light cavalry
- short lived (4-5 days in blood)
- phagocytosis of bacteria
- degranulation of antimicrobials
- extracellular “trapping”
- primary TF determinant **C/EBPα**

macrophage
(“big eater”)



- tissue resident heavy cavalry
- long lived
- phagocytosis of foreign or damaged cells
- coordinates with adaptive immune system
- wound healing
- primary TF determinant **PU.1**

“Paradoxically, both transcription factors (C/EBPα and PU.1) are highly expressed in macrophages and neutrophils and synergistically regulate transcription of genes that are active in one or the other cell type”