

# Theoretical biological physics

## 2. Principles of gene regulation

# Smoluchowski search picture

Search rate for a particle with diffusivity  $D_{3d}$  to find an immobile target of radius  $a$  (assuming immediate binding):

$$k_{\text{on}}^S = 4\pi D_{3d} a$$

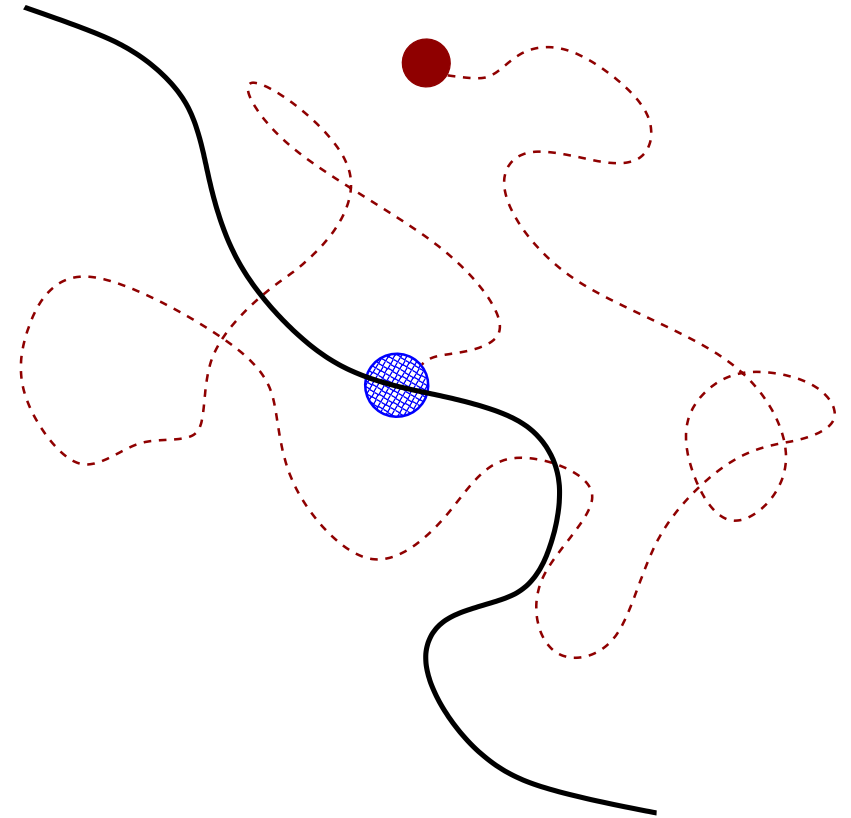
Protein-DNA interaction:  $a \approx \{\text{few bp}\} \approx 1\text{nm}$   
 $D_{3d} \approx 10\mu\text{m}^2/\text{sec}$  (typically  $\varnothing_{\text{TF}} \approx 5\text{nm}$ ):

$$k_{\text{on}}^S \approx \frac{10^8}{(\text{mol/l}) \cdot \text{sec}}$$

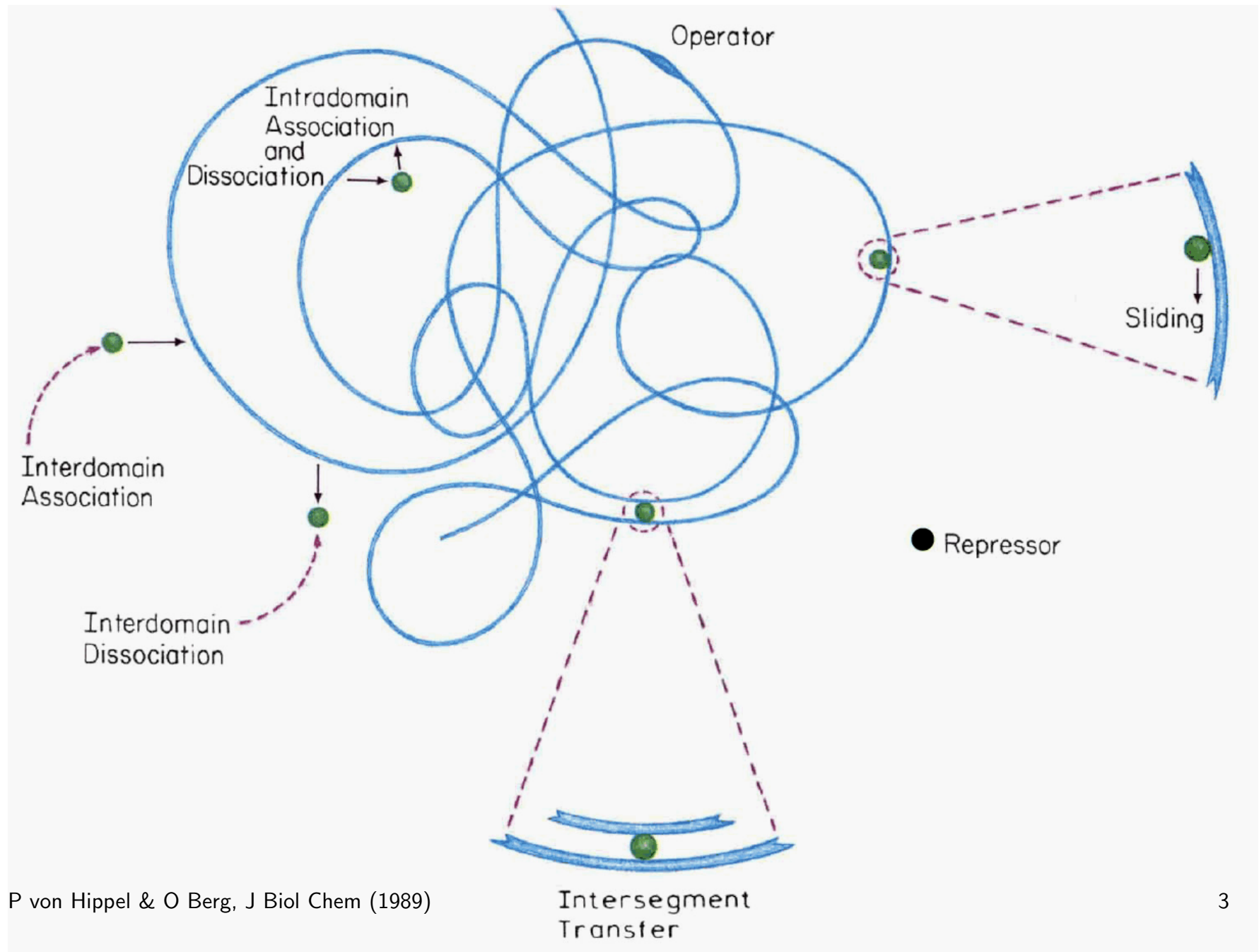
Lac repressor [AD Riggs, S Bourgeois, M Cohn, J Mol Biol 53, 401 (1970)]:

$$k_{\text{on}} \approx \frac{10^{10}}{(\text{mol/l}) \cdot \text{sec}}$$

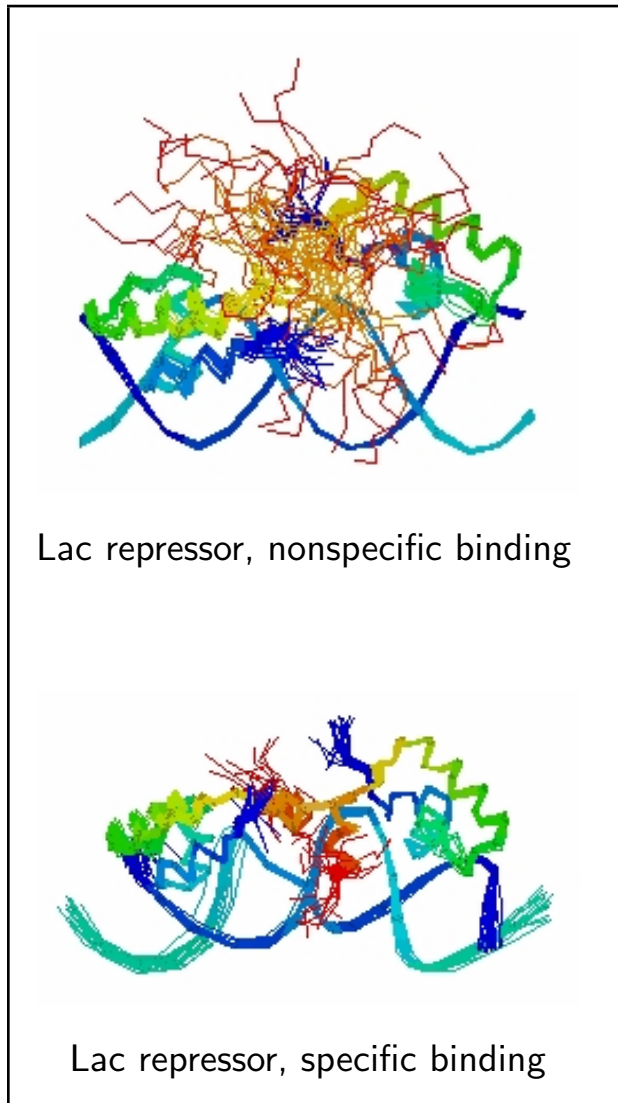
↪ Facilitated diffusion picture



# Facilitated diffusion: the Berg-von Hippel model



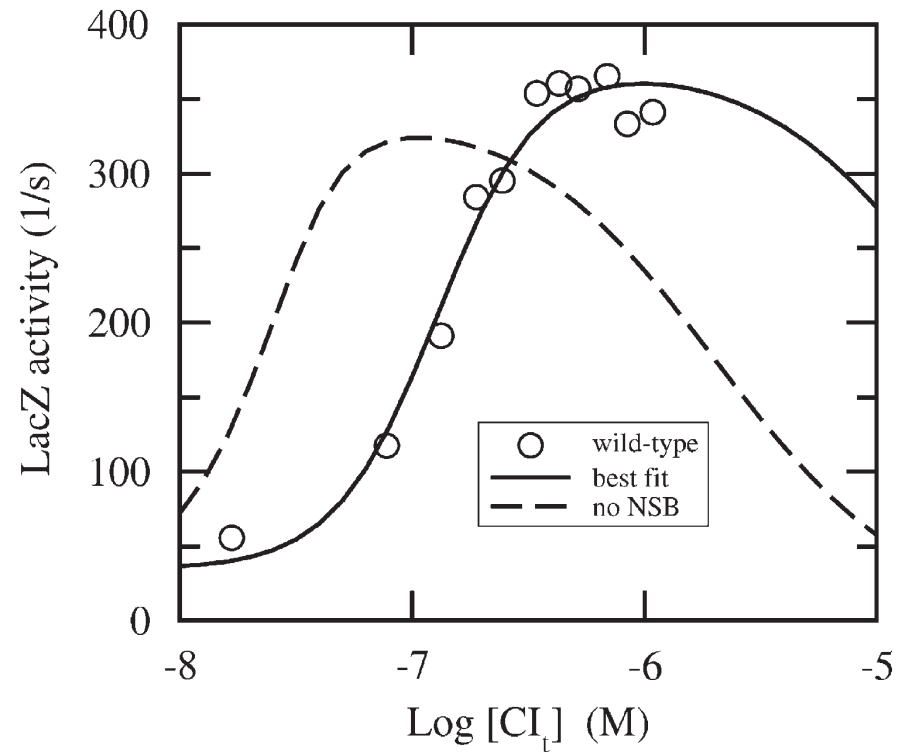
# Non-specific binding energy based on *in vivo* data



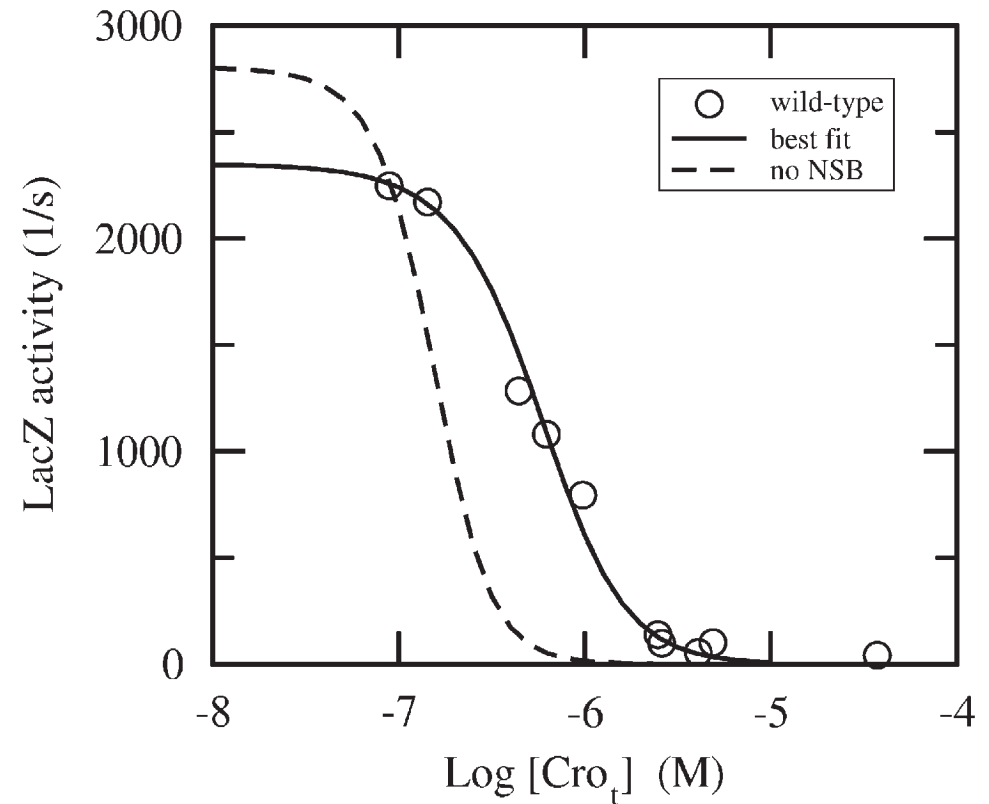
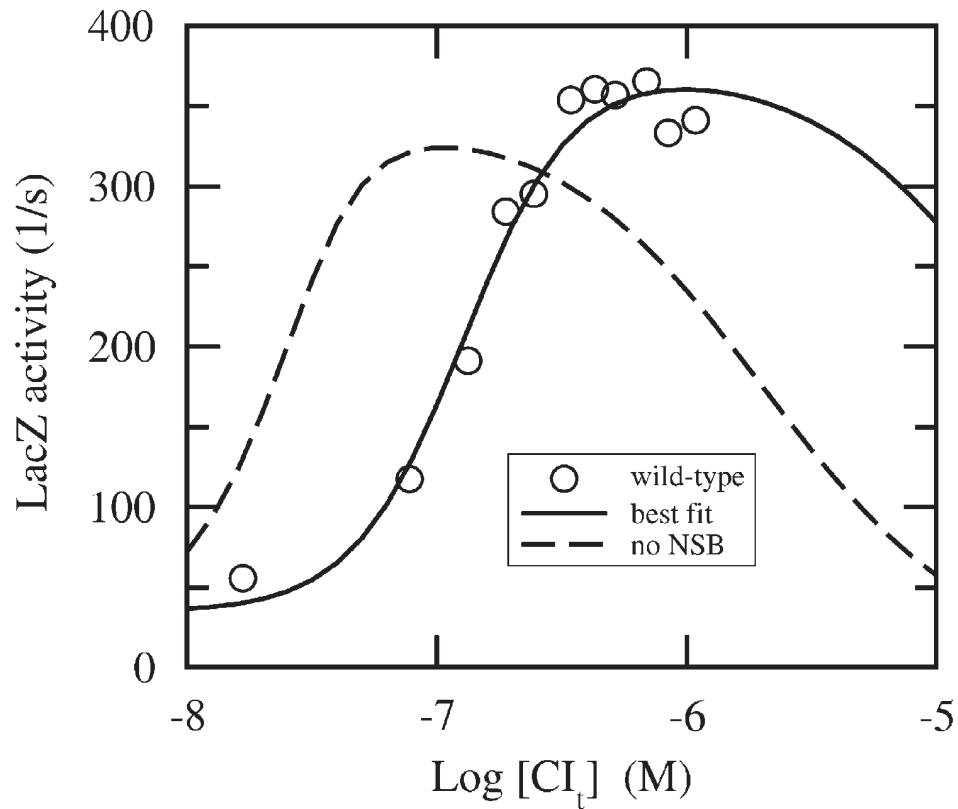
$$[X] = [X_{\text{free}}] + [X_{\text{@OP}}] + [X_{\text{NSB}}]$$

$$\Delta G_{\text{NSB}}(\text{CI}) = -4.1 \pm 0.9 \text{ kcal/mol,}$$

$$\Delta G_{\text{NSB}}(\text{Cro}) = -4.2 \pm 0.8 \text{ kcal/mol}$$

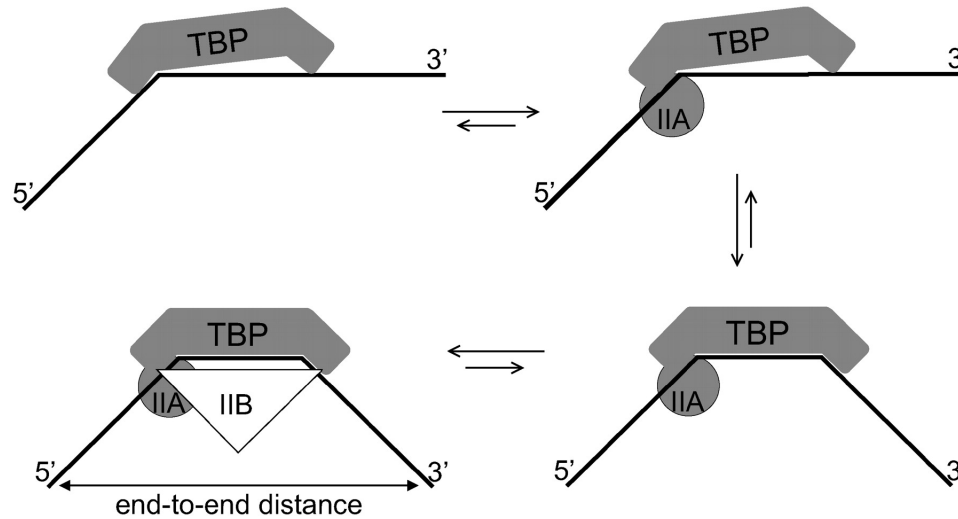
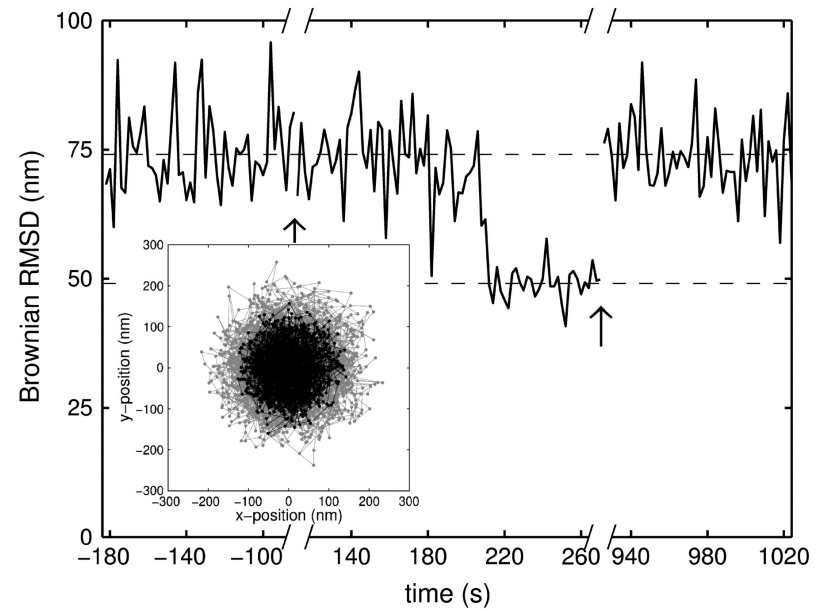
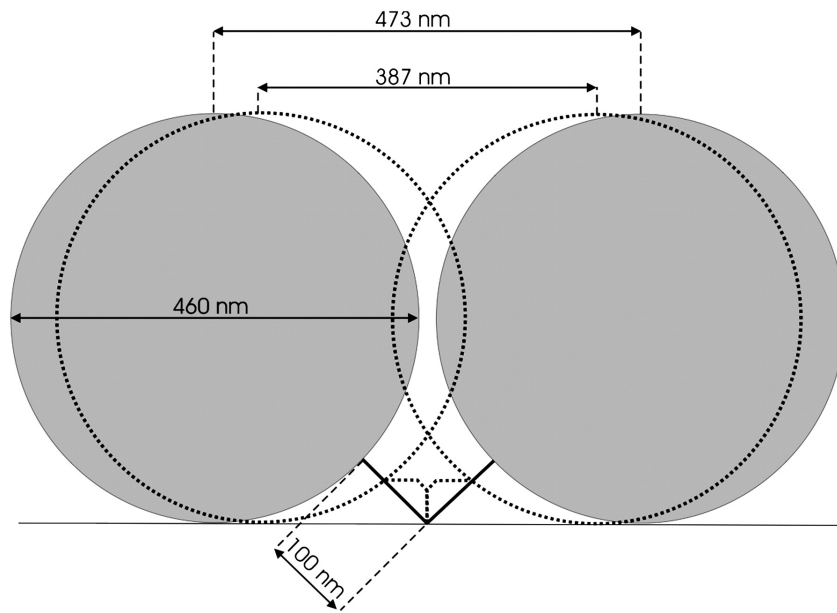


# Promoter activity as function of TF concentration



- ☕ Non-specific binding relevant
- ☕ LacZ activity optimum as function of [CI]
- ☕ Simple switch as function of [Cro]

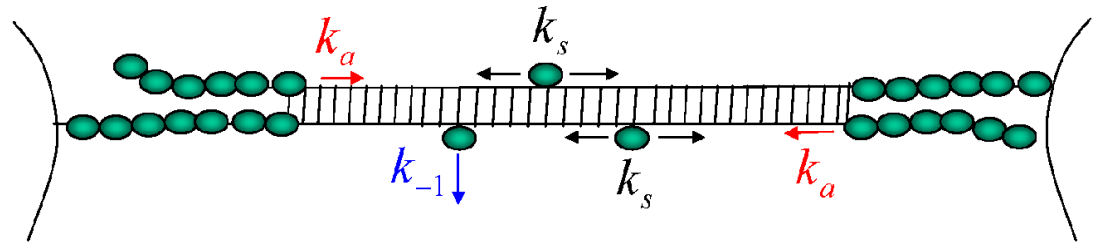
# Single molecule dynamics of TATA binding protein



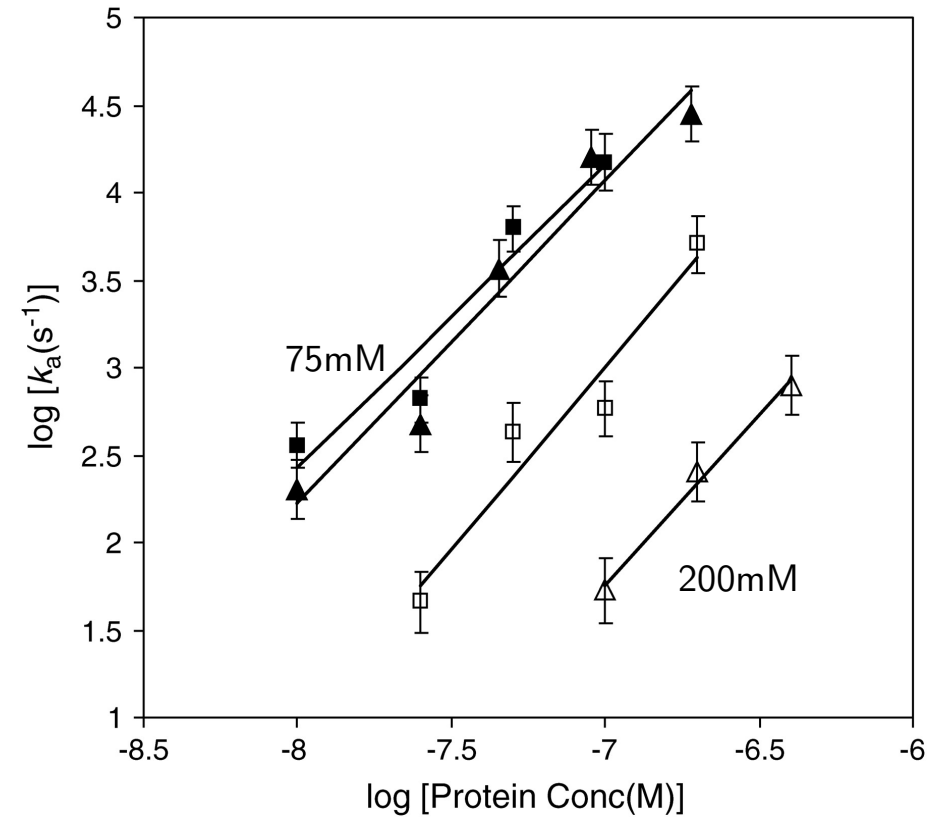
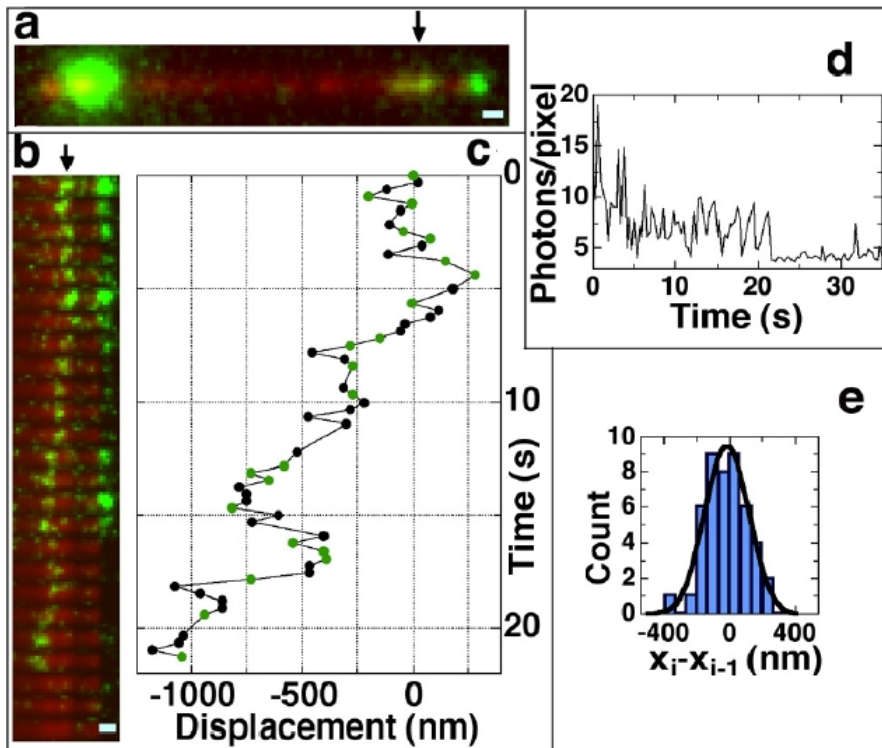
# Proof of 1D search mode

McGhee & von Hippel isotherm

$$f = \frac{N\lambda}{L} \simeq K_{ns}\lambda C, \quad f \ll 1$$

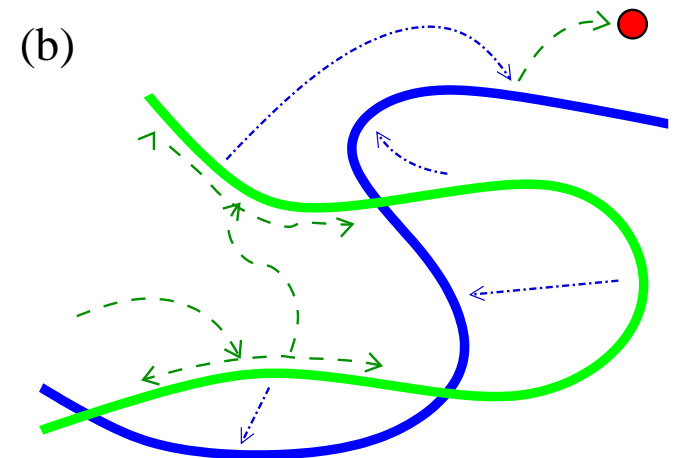
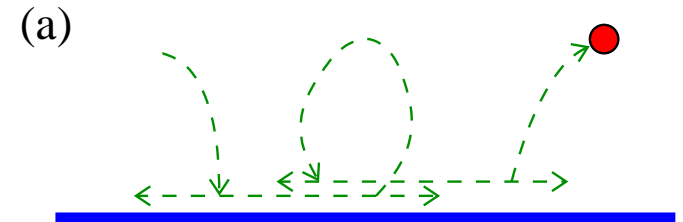
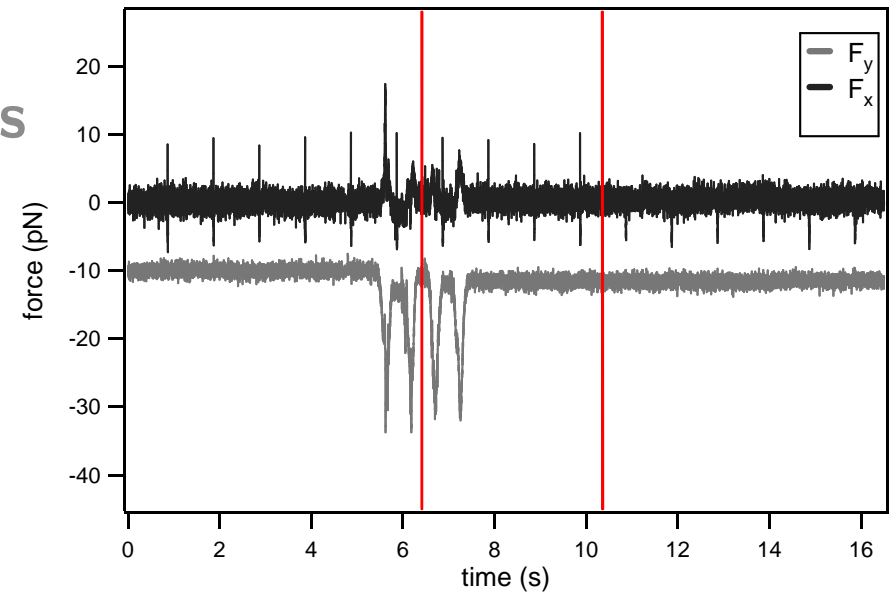
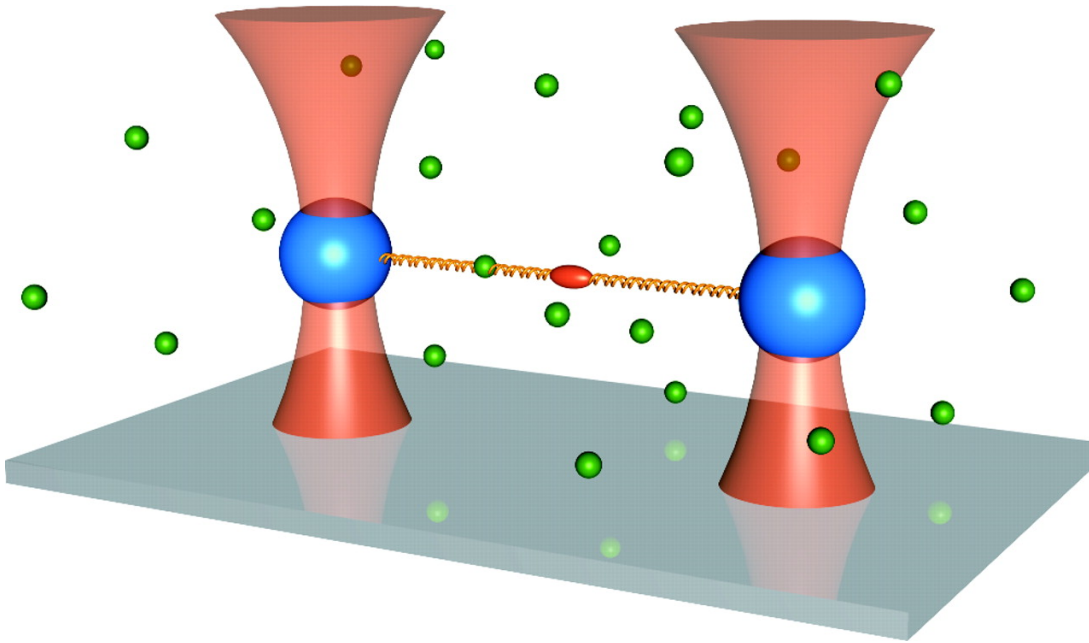


$$k_a \simeq \begin{cases} C, & \text{1D/3D Berg \& von Hippel} \\ C^2, & \text{Pure 1D search} \end{cases}$$



$$\Delta = 1.74 \pm 0.35, 1.85 \pm 0.24, 2.08 \pm 0.39, 1.95 \pm 0.17$$

# The rôle of DNA conformations



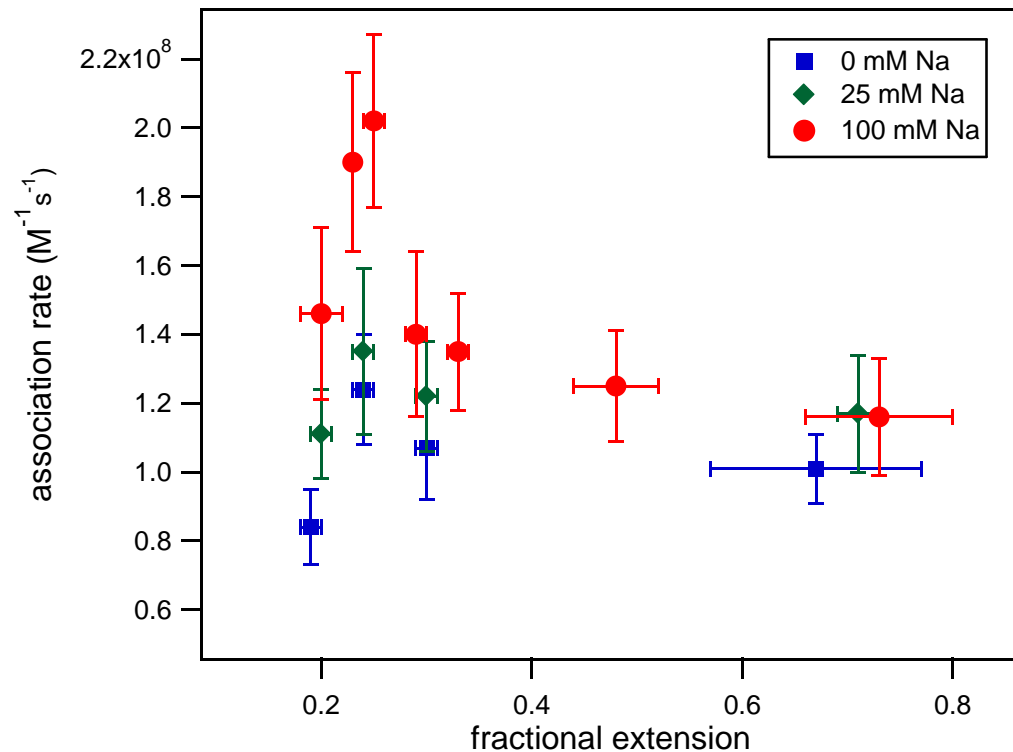
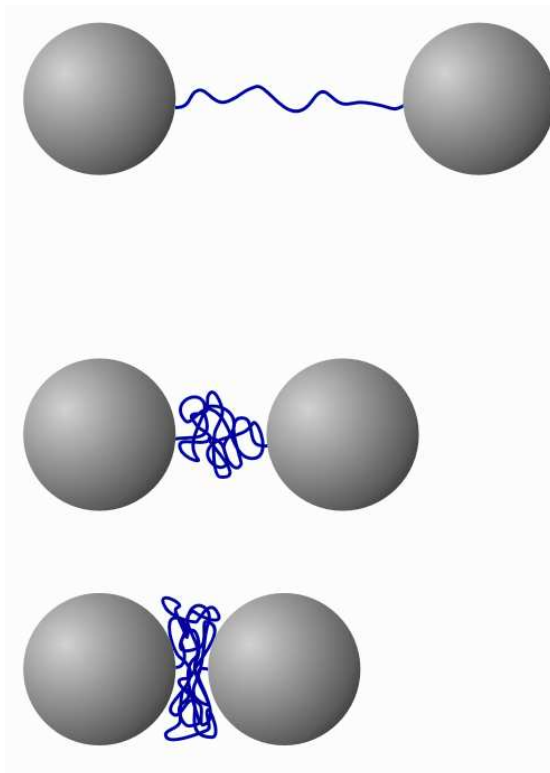
pCco5 plasmid DNA:  $6538\text{bp} \approx 2.2\mu\text{m} \approx 45l_p$   
 [comp  $\lambda$  DNA 48.5kbp]



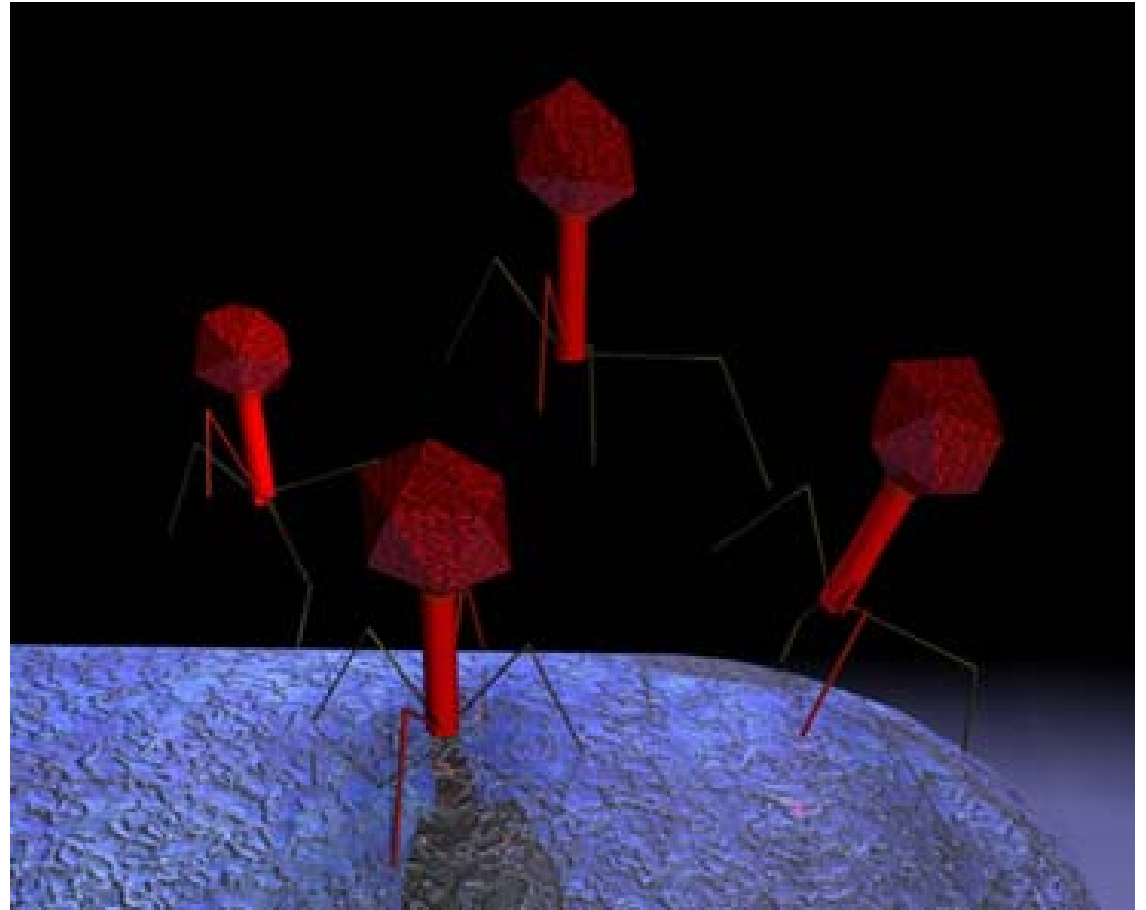
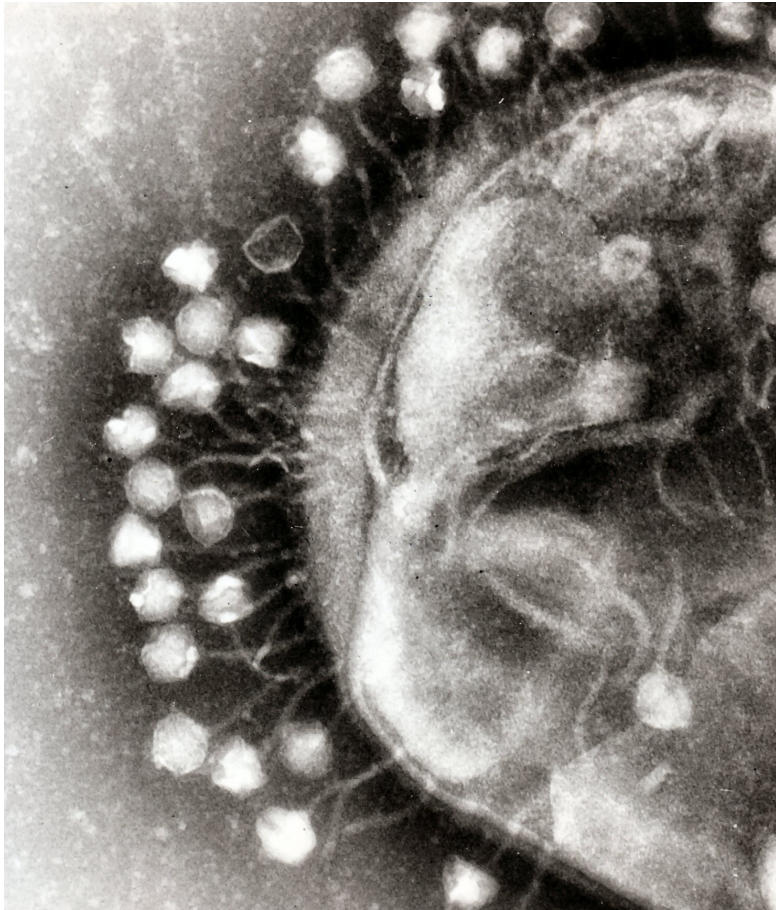
# More compact DNA conformations speed up the search

[NaCl]	$k_{\text{on}}^{\text{straight}}$ [Ms]	$l_{\text{sl}}^{\text{eff}}$ [bp]	$1/\sqrt{l_{\text{DNA}}}$ [bp]	$l_p$ [bp]	$R_{\text{theory}}$	$R_{\text{measured}}$
0 mM	$0.8 \times 10^8$	195	518	188	1.18	$1.3 \pm 0.2$
25 mM	$1.0 \times 10^8$	250	485	175	1.23	$1.1 \pm 0.2$
100 mM	$1.0 \times 10^8$	250	150	159	1.67	$1.7 \pm 0.3$
150 mM	$0.9 \times 10^9$	15.5	120	153	1.15	$1.3 \pm 0.4$

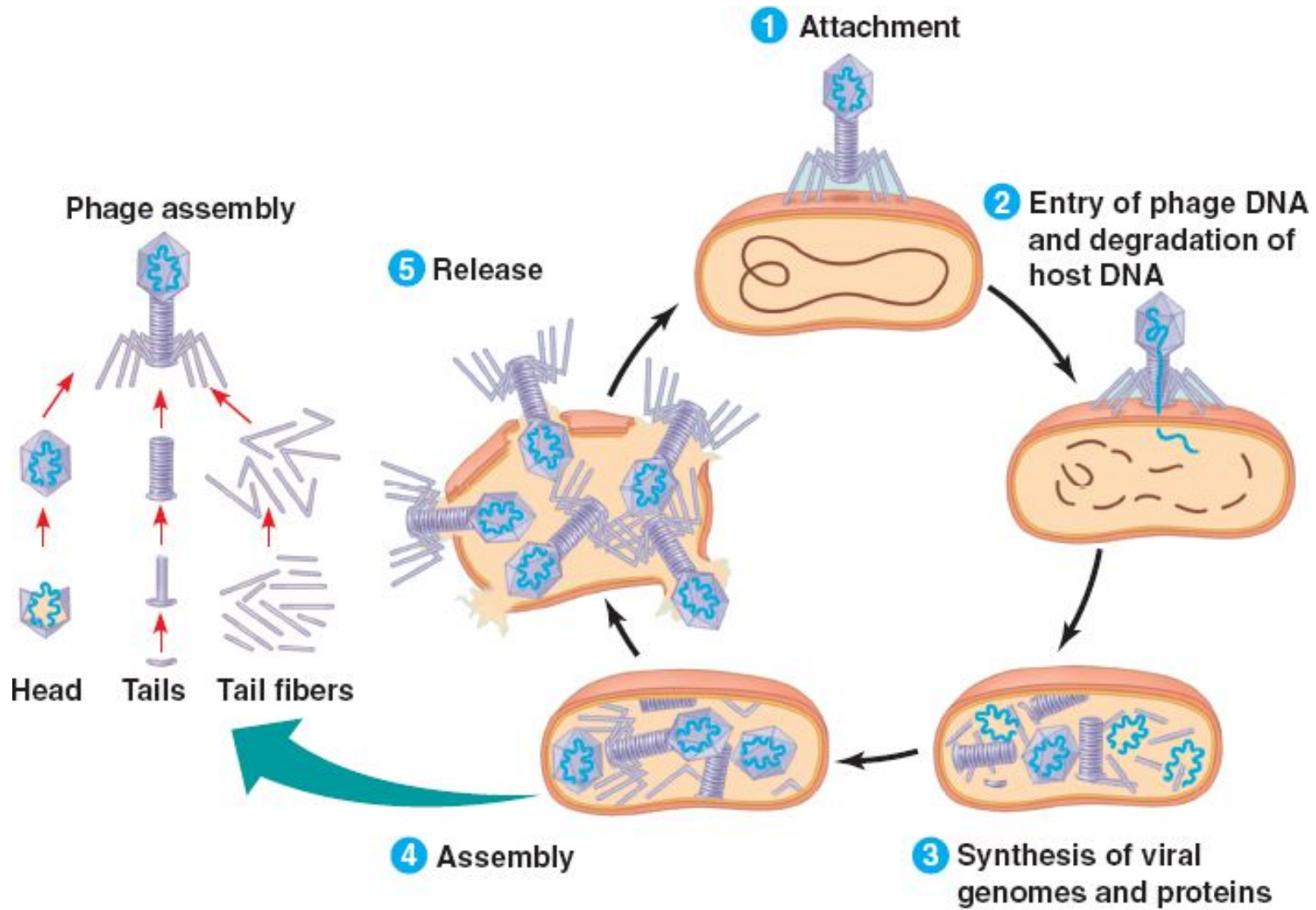
$R = k_{\text{on}}^{\text{max}} / k_{\text{on}}^{\text{straight}}$ : enhancement ratio of attachment rates @ max and straight configuration)



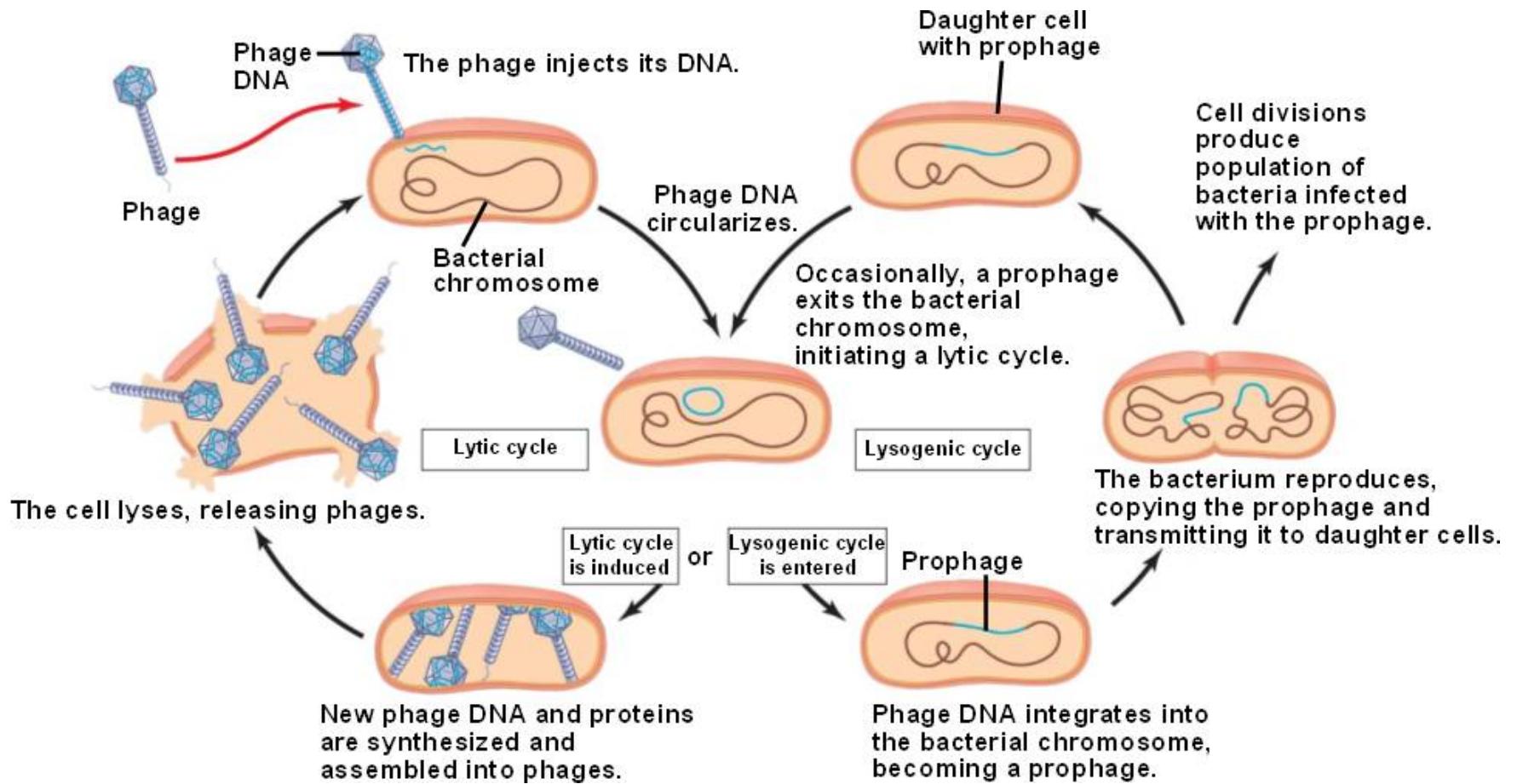
# Bacteria eaters (bacteriophages): bacteria get infected



# Purely lytic cycle



# Phage $\lambda$ pathways



# Geometrical considerations

Outer radius  $a_0 = 21\text{nm}$

Outer height  $b_0 = 54\text{nm}$

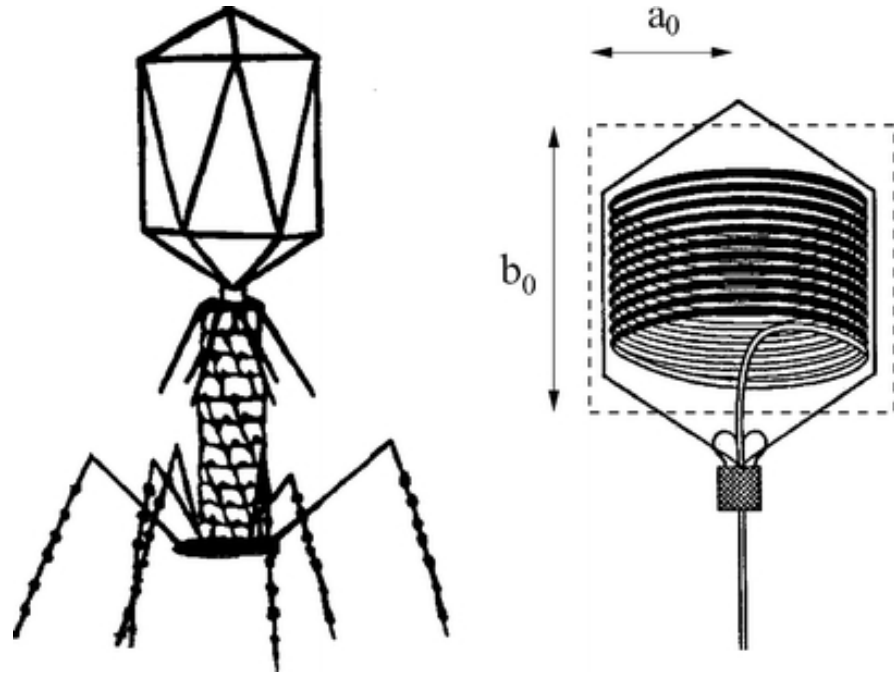
DNA length  $L = 6.6\mu\text{m}$

DNA persistence length  $\ell_p \approx 53\text{nm}$

Inner volume  $V \approx 4.6 \times 10^4 \text{nm}^3$

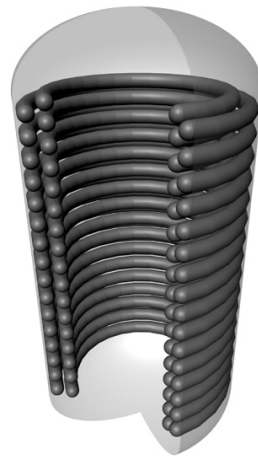
Cylinder /w  $L$  &  $d_{\text{eff}} \approx 2.5\text{nm}$ :

$V \approx 3.2 \times 10^4 \text{nm}^3$

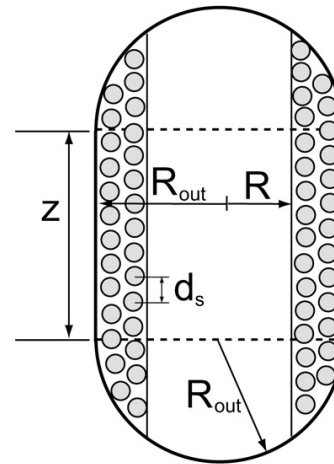




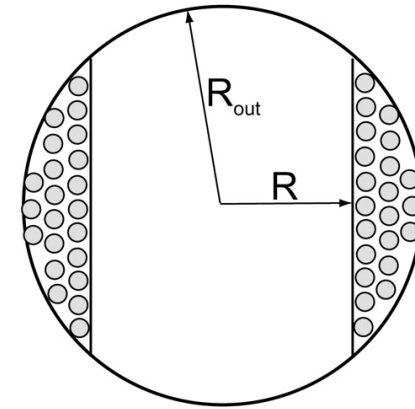
# DNA packaging



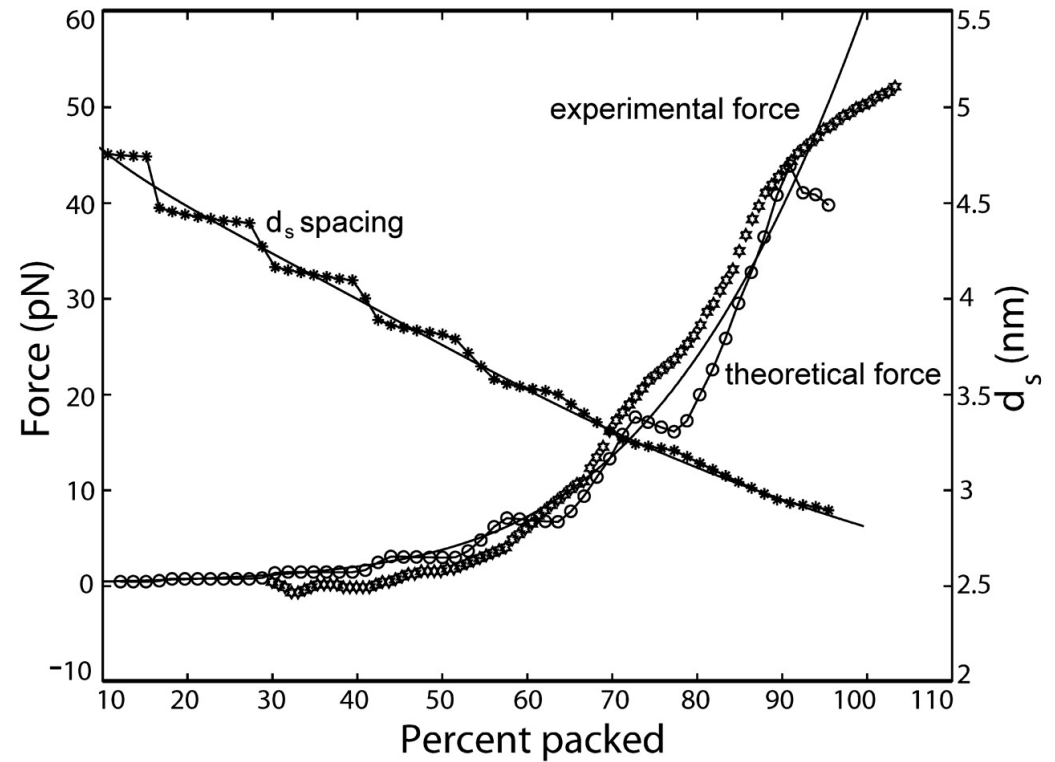
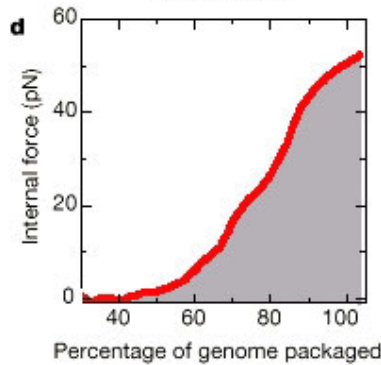
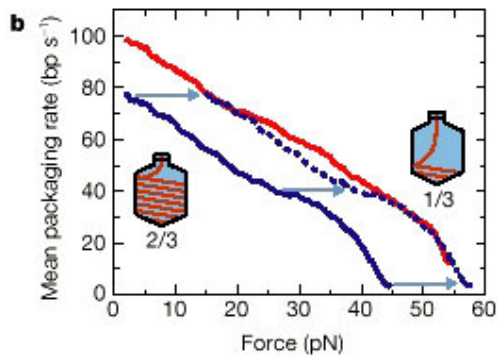
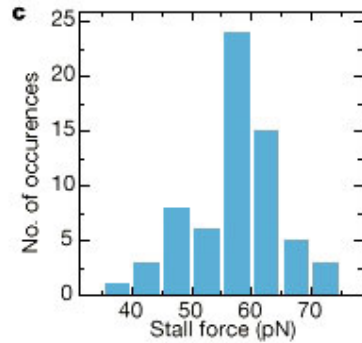
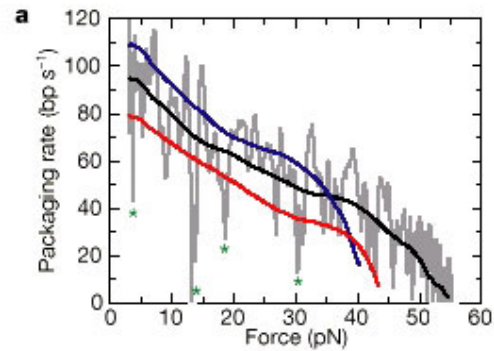
(a) Capsid in 3D



(b) Capped cylinder



(c) Sphere



Purohit et al, Biophys J (2005); comp Kindt et al, PNAS (2001)

# Stat Mech approach to gene stability

Probability to find system in one of the 40 states  $s$ :

$$f_s = \frac{\exp(-\Delta G(s)/RT)[\text{Cl}_2]^{i_s}[\text{Cro}_2]^{j_s}[\text{RNAP}]^{k_s}}{\sum_s \exp(-\Delta G(s)/RT)[\text{Cl}_2]^{i_s}[\text{Cro}_2]^{j_s}[\text{RNAP}]^{k_s}}$$

☕ Steady state description without dynamics

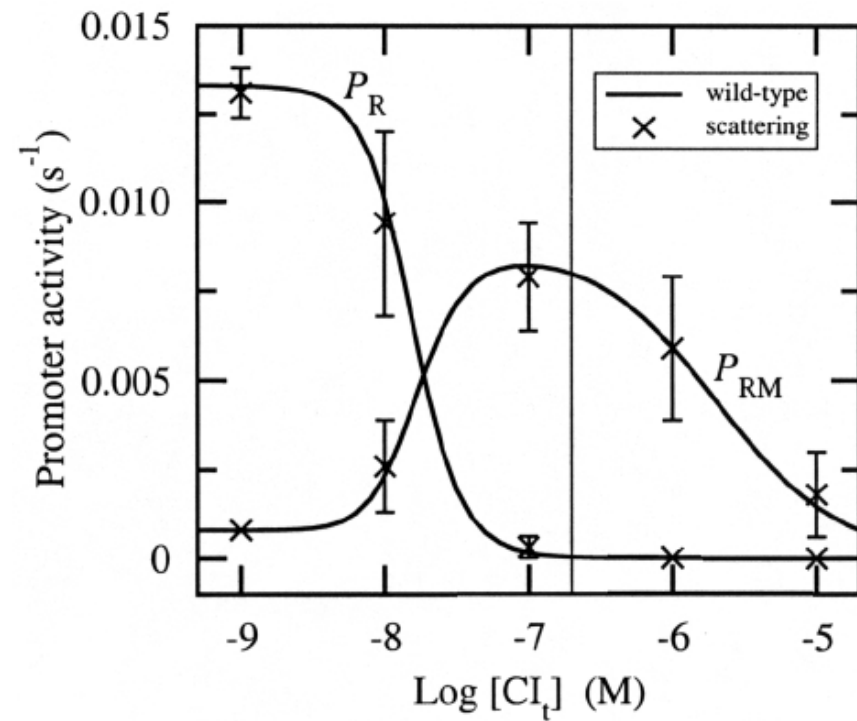
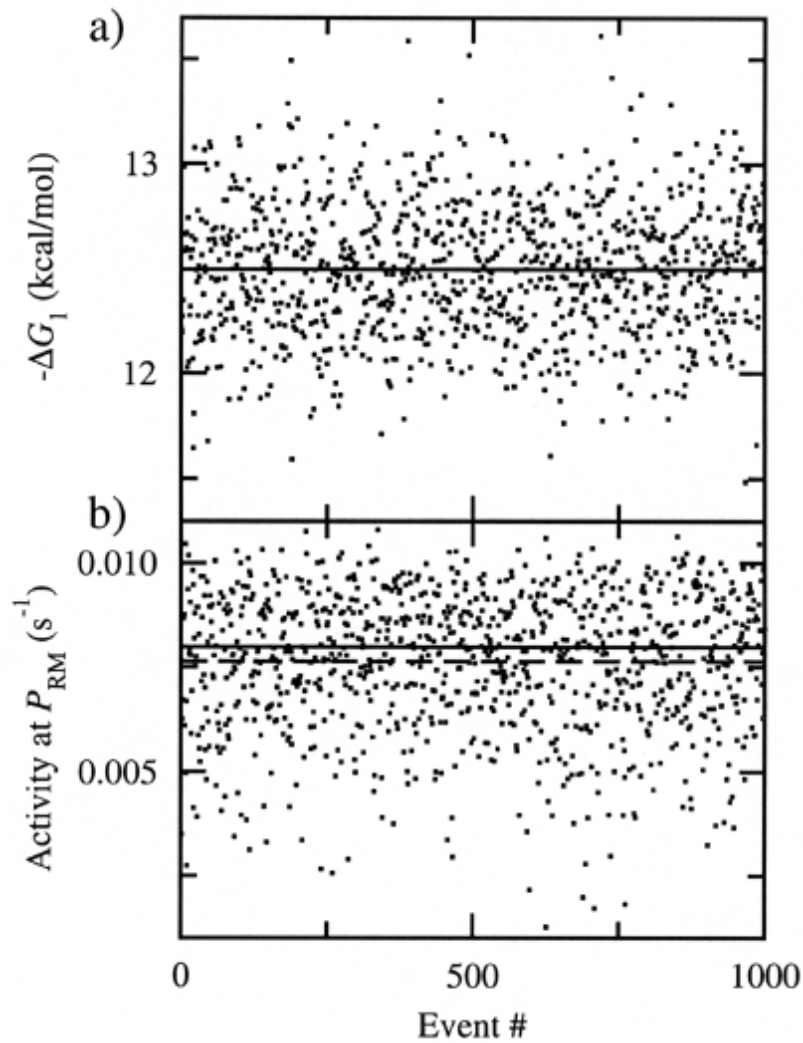
☕ Based on in vitro data

☕ Highly dimensional parameter space

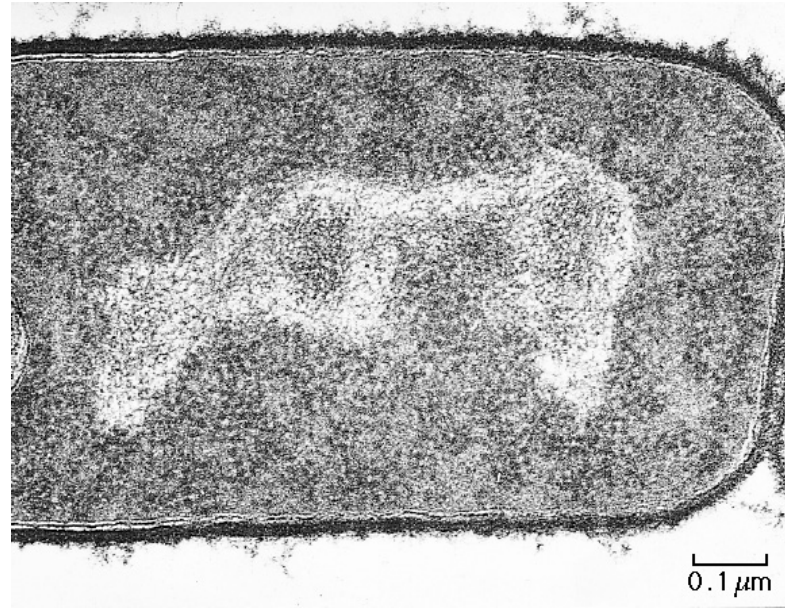
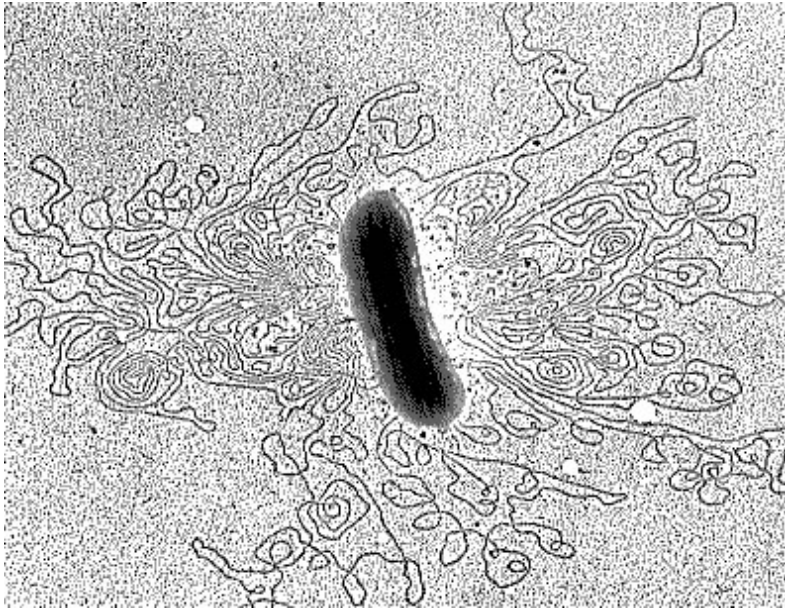
$s$	$O_{R3}$		$O_{R2}$		$O_{R1}$	terms	GFE
1	0		0		0	Reference state	0
2	0		0		R	$\Delta G_1$	-12.5
3	0		R		0	$\Delta G_2$	-10.5
4	R		0		0	$\Delta G_3$	-9.5
5	0		0		C	$\Delta G_{1'}$	-12.0
6	0		C		0	$\Delta G_{2'}$	-10.8
7	C		0		0	$\Delta G_{3'}$	-13.4
8	RNAP		0		0	$\Delta G_{RM}$	-11.5
9	0			RNAP		$\Delta G_R$	-12.5
10	0		R	$\leftrightarrow$	R	$\Delta G_1 + \Delta G_2 + \Delta G_{12}$	-25.7
11	R		0		R	$\Delta G_1 + \Delta G_3$	-22.0
12	R	$\leftrightarrow$	R		0	$\Delta G_2 + \Delta G_3 + \Delta G_{23}$	-22.9
13	0		C	$\leftrightarrow$	C	$\Delta G_{1'} + \Delta G_{2'} + \Delta G_{12'}$	-23.8
14	C		0		C	$\Delta G_{1'} + \Delta G_{3'}$	-25.4
15	C	$\leftrightarrow$	C		0	$\Delta G_{2'} + \Delta G_{3'} + \Delta G_{23'}$	-24.8
16	RNAP			RNAP		$\Delta G_{RM} + \Delta G_R$	-24.0
17	0		C		R	$\Delta G_1 + \Delta G_{2'}$	-23.3
18	0		R		C	$\Delta G_{1'} + \Delta G_2$	-22.5
19	R		0		C	$\Delta G_{1'} + \Delta G_3$	-21.5
20	C		0		R	$\Delta G_1 + \Delta G_{3'}$	-25.9
21	R		C		0	$\Delta G_{2'} + \Delta G_3$	-20.3
22	C		R		0	$\Delta G_2 + \Delta G_{3'}$	-23.9
23	R			RNAP		$\Delta G_R + \Delta G_3$	-22.0
24	RNAP		R		0	$\Delta G_2 + \Delta G_{RM}$	-22.0
25	RNAP		0		R	$\Delta G_1 + \Delta G_{RM}$	-24.0
26	C			RNAP		$\Delta G_R + \Delta G_{3'}$	-25.9
27	RNAP		C		0	$\Delta G_{2'} + \Delta G_{RM}$	-22.3
28	RNAP		0		C	$\Delta G_{1'} + \Delta G_{RM}$	-23.5
29	R		R	$\leftrightarrow$	R	$\Delta G_1 + \Delta G_2 + \Delta G_3 + \Delta G_{12}$	-35.2
30	C	$\leftrightarrow$	C	$\leftrightarrow$	C	$\Delta G_{1'} + \Delta G_{2'} + \Delta G_{3'} + \Delta G_{123'}$	-37.1
31	C		R	$\leftrightarrow$	R	$\Delta G_1 + \Delta G_2 + \Delta G_{3'} + \Delta G_{12}$	-39.1
32	R		C		R	$\Delta G_1 + \Delta G_{2'} + \Delta G_3$	-32.8
33	R	$\leftrightarrow$	R		C	$\Delta G_{1'} + \Delta G_2 + \Delta G_3 + \Delta G_{23}$	-34.9
34	R		C	$\leftrightarrow$	C	$\Delta G_{1'} + \Delta G_{2'} + \Delta G_3 + \Delta G_{12'}$	-33.3
35	C		R		C	$\Delta G_{1'} + \Delta G_2 + \Delta G_{3'}$	-35.9
36	C	$\leftrightarrow$	C		R	$\Delta G_1 + \Delta G_{2'} + \Delta G_{3'} + \Delta G_{23'}$	-37.3
37	RNAP		R	$\leftrightarrow$	R	$\Delta G_1 + \Delta G_2 + \Delta G_{RM} + \Delta G_{12}$	-37.2
38	RNAP		C	$\leftrightarrow$	C	$\Delta G_{1'} + \Delta G_{2'} + \Delta G_{RM} + \Delta G_{12'}$	-35.3
39	RNAP		C		R	$\Delta G_1 + \Delta G_{2'} + \Delta G_{RM}$	-34.8
40	RNAP		R		C	$\Delta G_{1'} + \Delta G_2 + \Delta G_{RM}$	-34.0



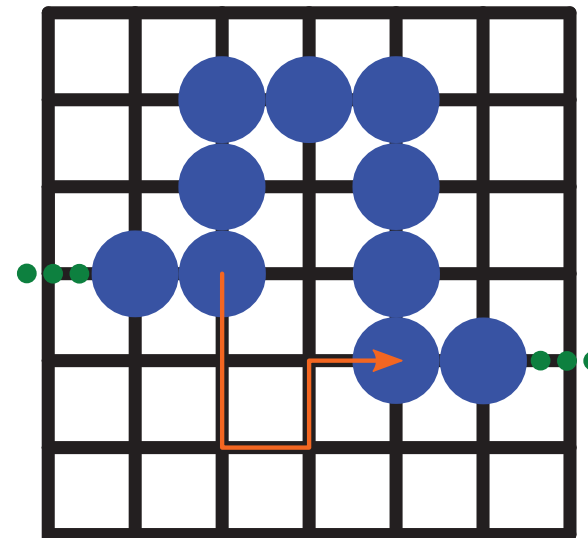
# Stability analysis of phage $\lambda$ switch



# In vivo gene regulation



Chromosome is approx a SAW  
[M Buenemann & P Lenz, PLoS ONE (2010)]



M Bauer & RM, PLoS ONE (2013)

# Intra/intercellular signalling is diffusion controlled

