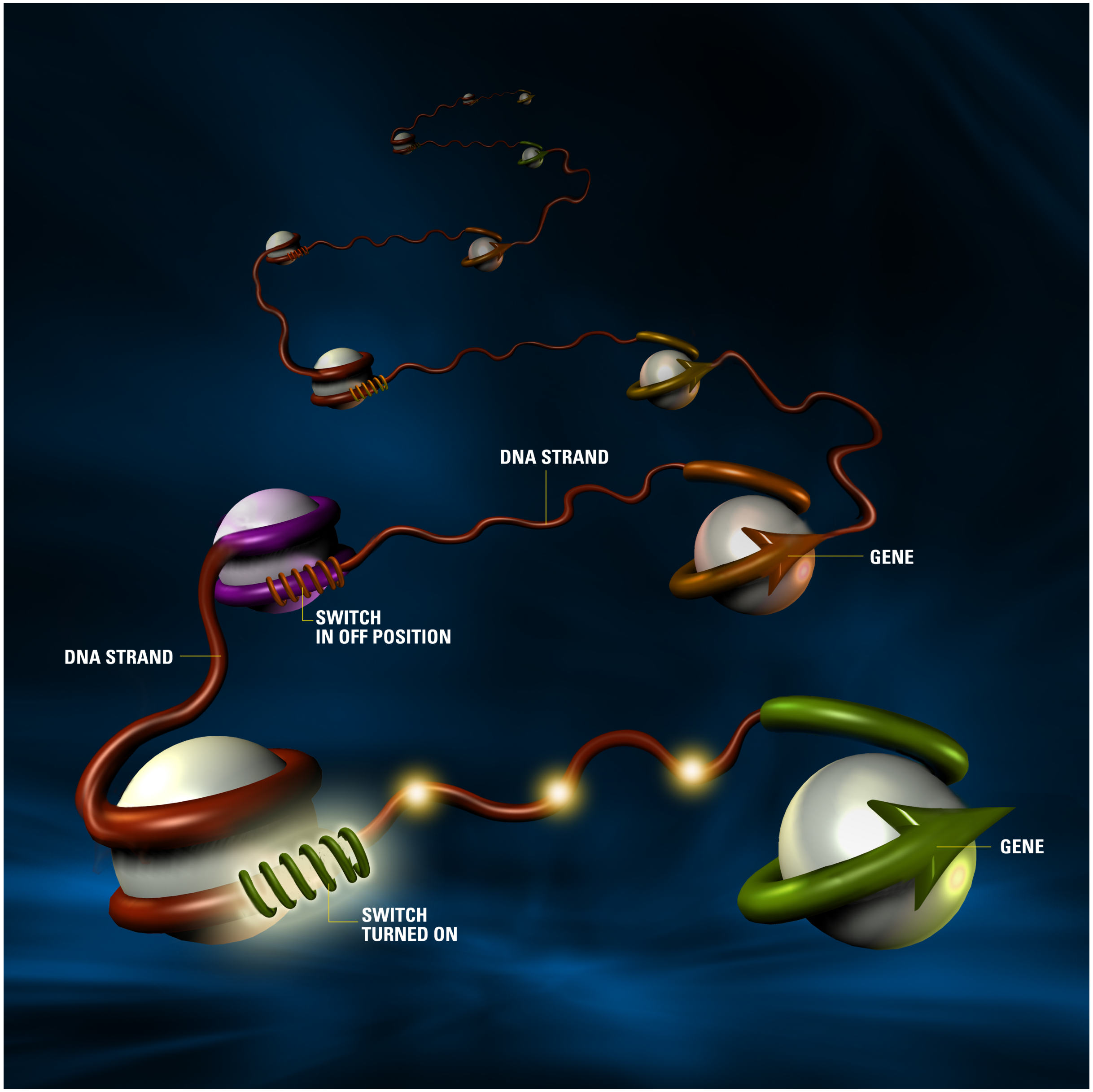
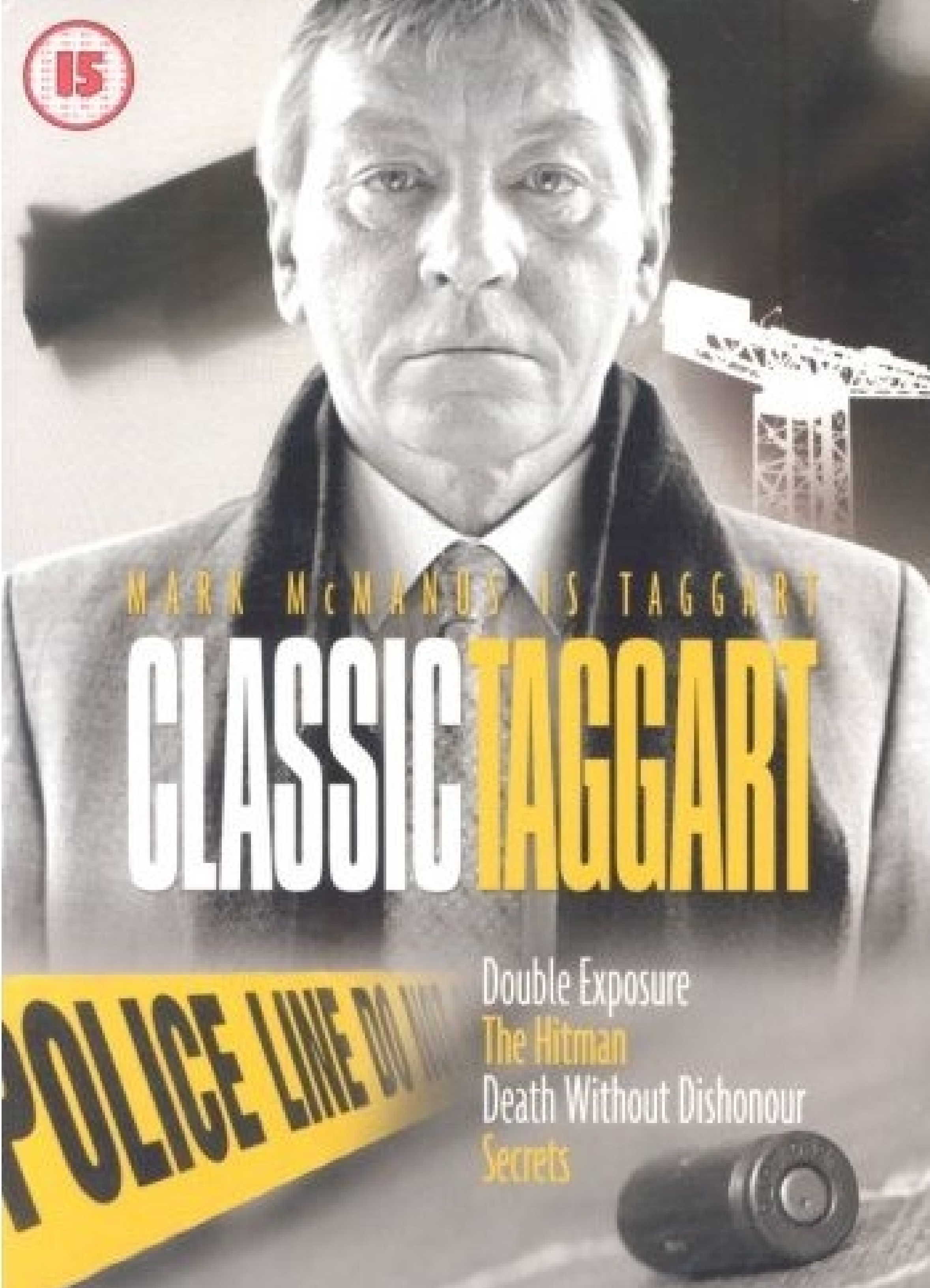
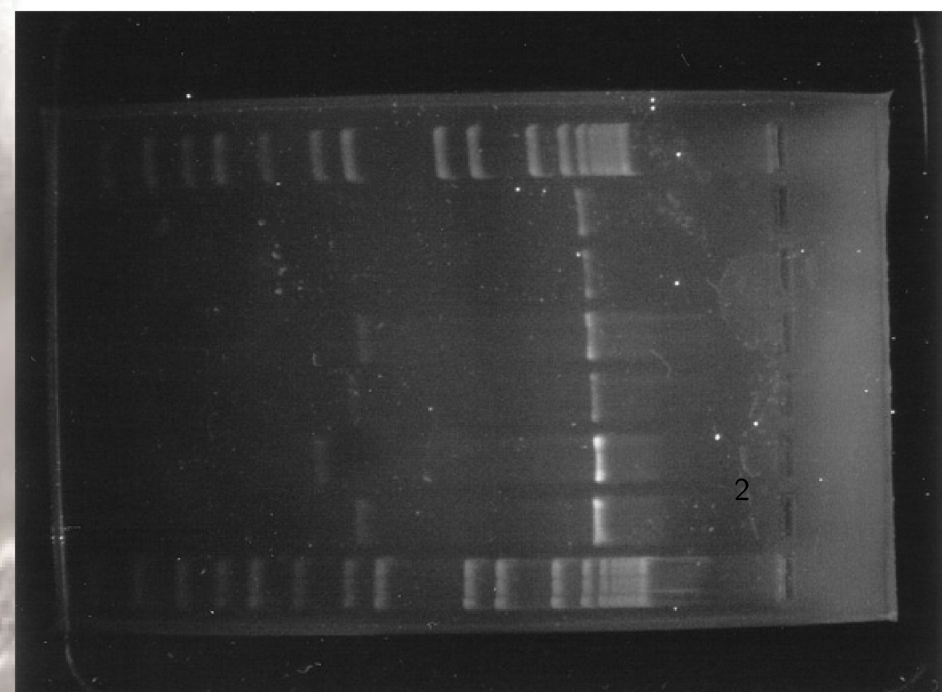
Biological Physics of DNA



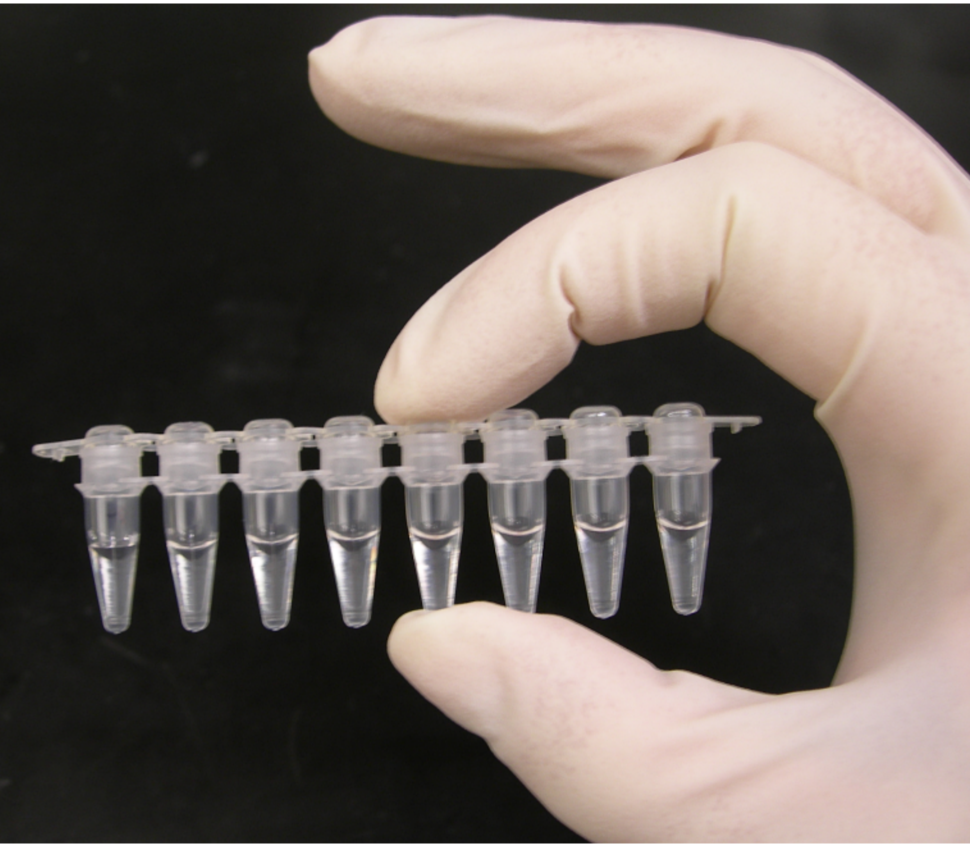
Whodunnit...


15
MARK McMANUS IS TAGGART
CLASSIC TAGGART
Double Exposure
The Hitman
Death Without Dishonour
Secrets
POLICE LINE DO

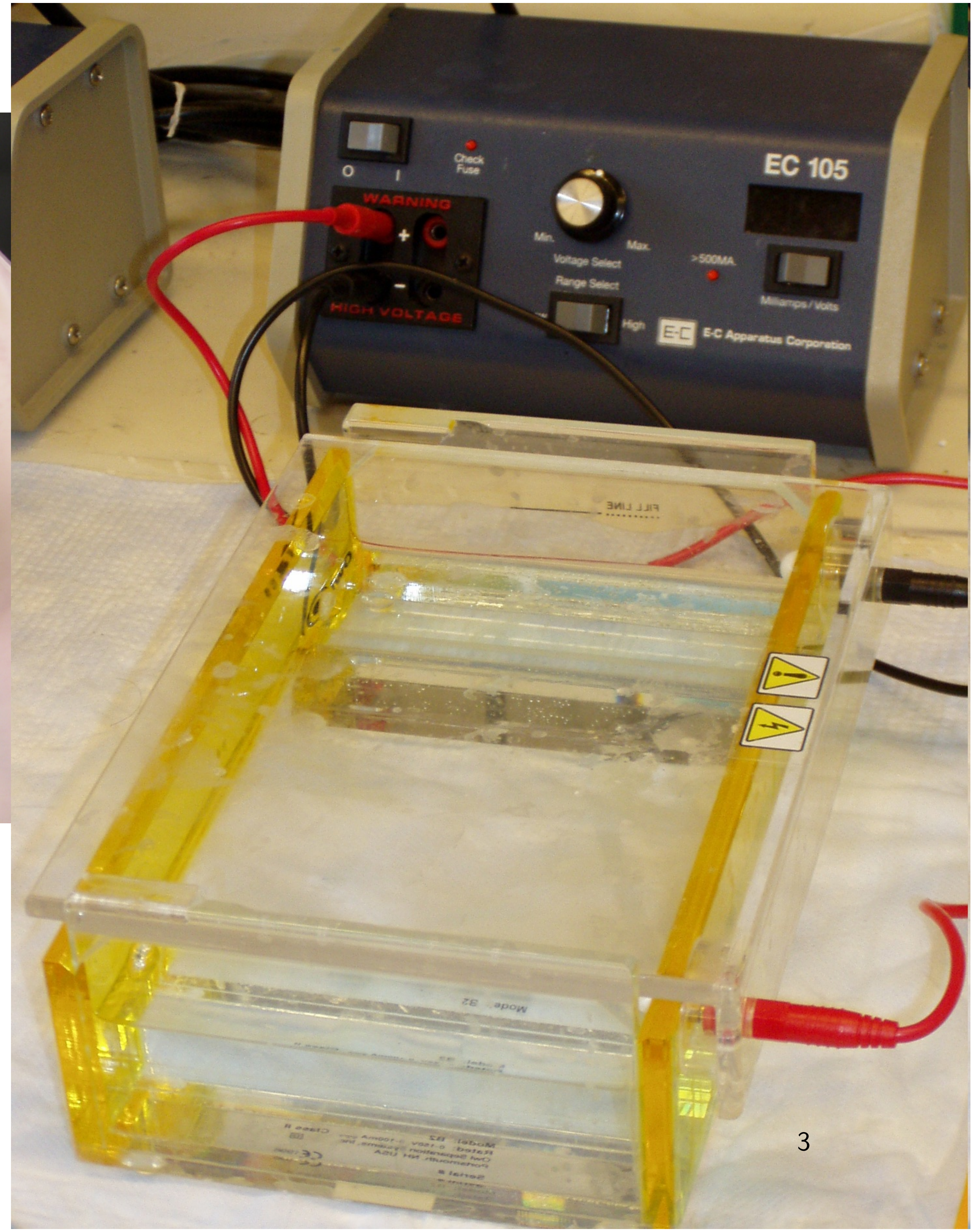

CSI: DNA LABORATORY
CRIME SCENE INVESTIGATION
CSI:

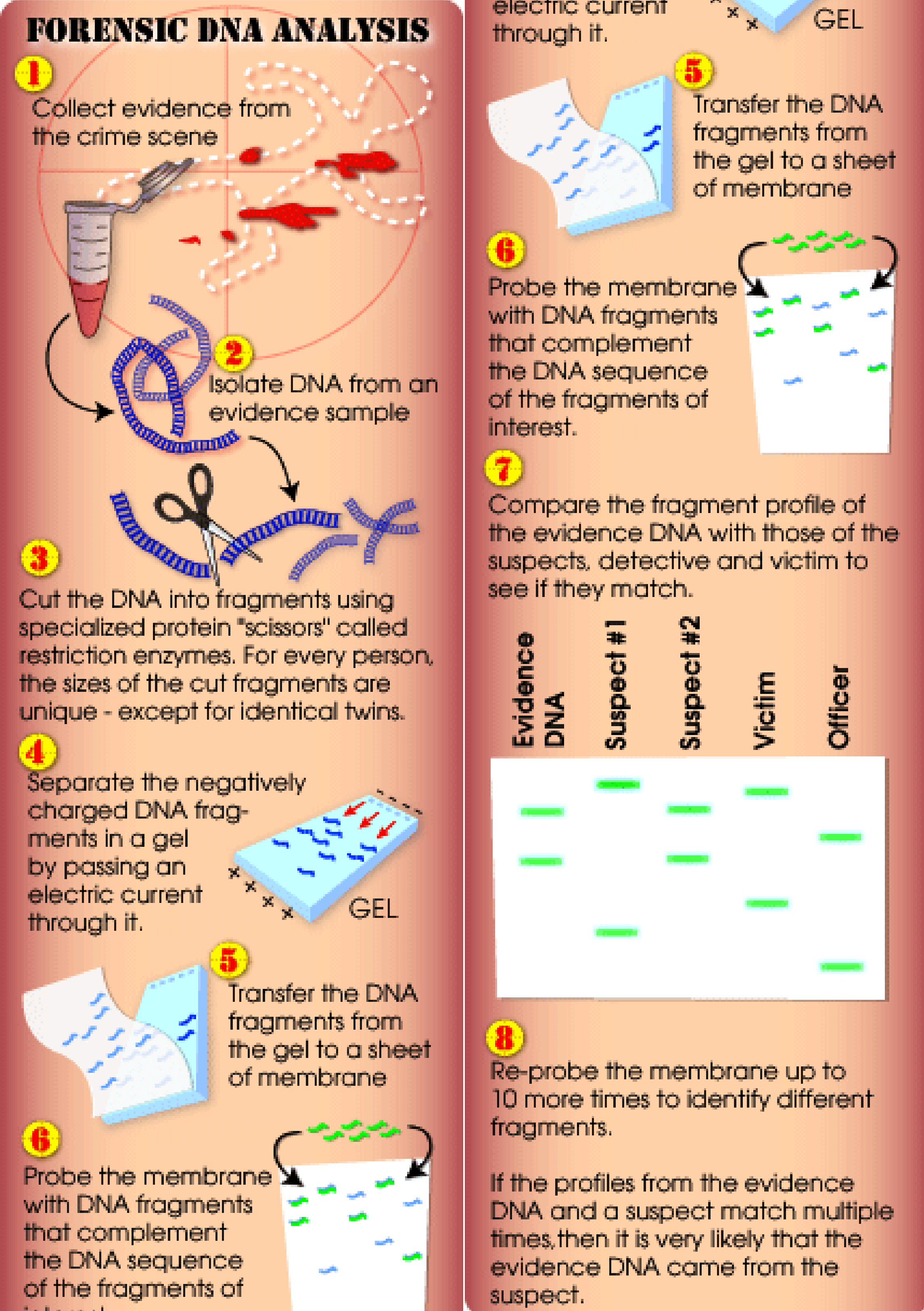


☟ DNA multiplication/PCR



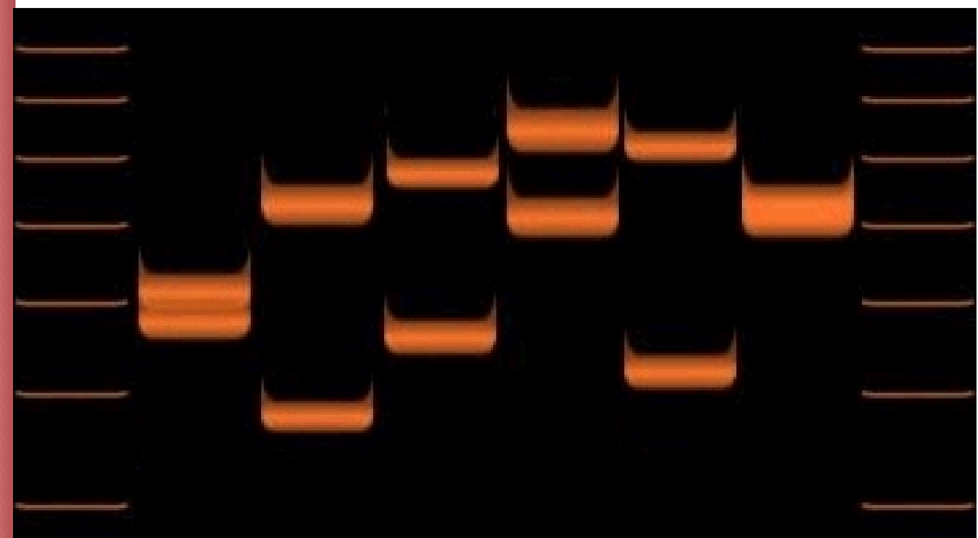
Gel electrophoresis ☞





NB: betw 4 & 5 denature fragments to ssDNA

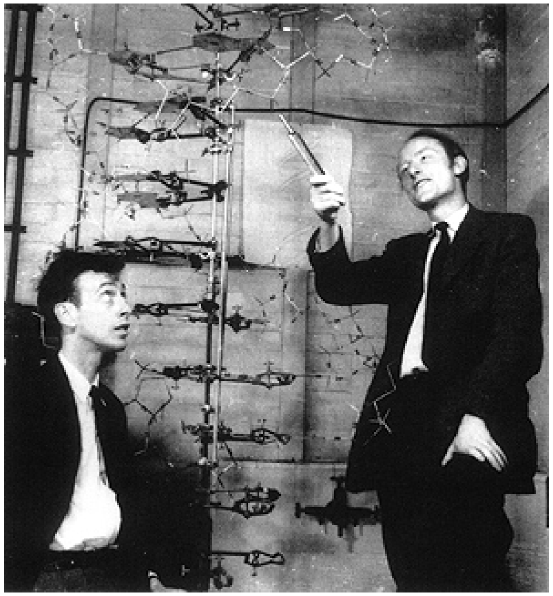
Labelling: eg radioact probe fragm & Xray film

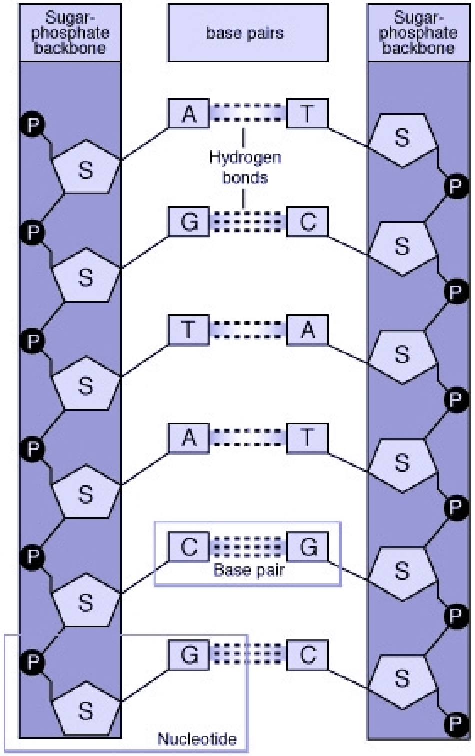


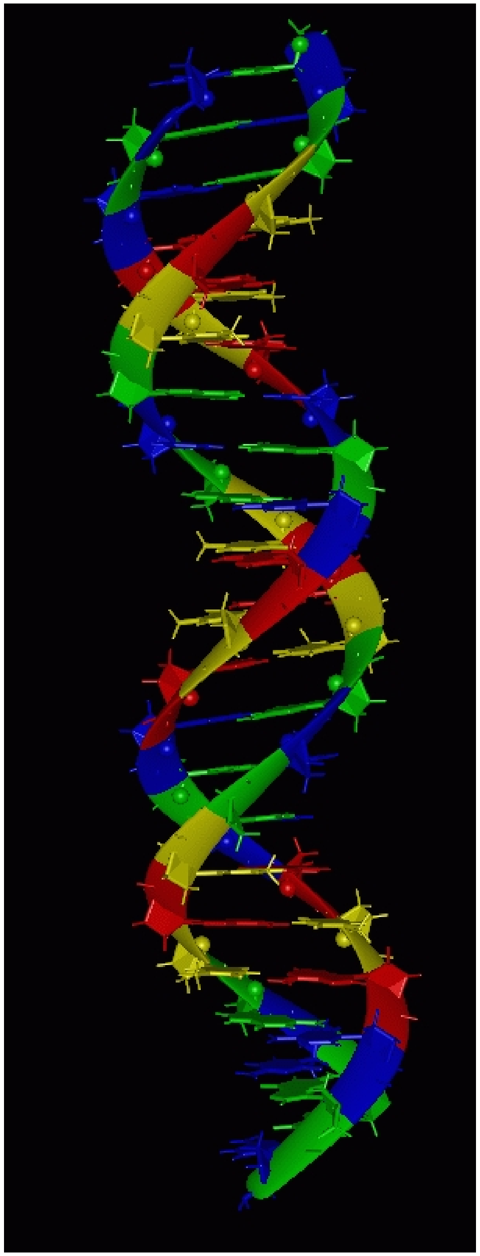
Variations of VNTR allele lengths in 6 individuals

http://learn.genetics.utah.edu/features/forensics/

Chief character: DeoxyriboNucleic Acid







$d \simeq 2\text{nm}$

$\Delta_{\text{bp}-\text{bp}} \simeq 3.5\text{Å}$

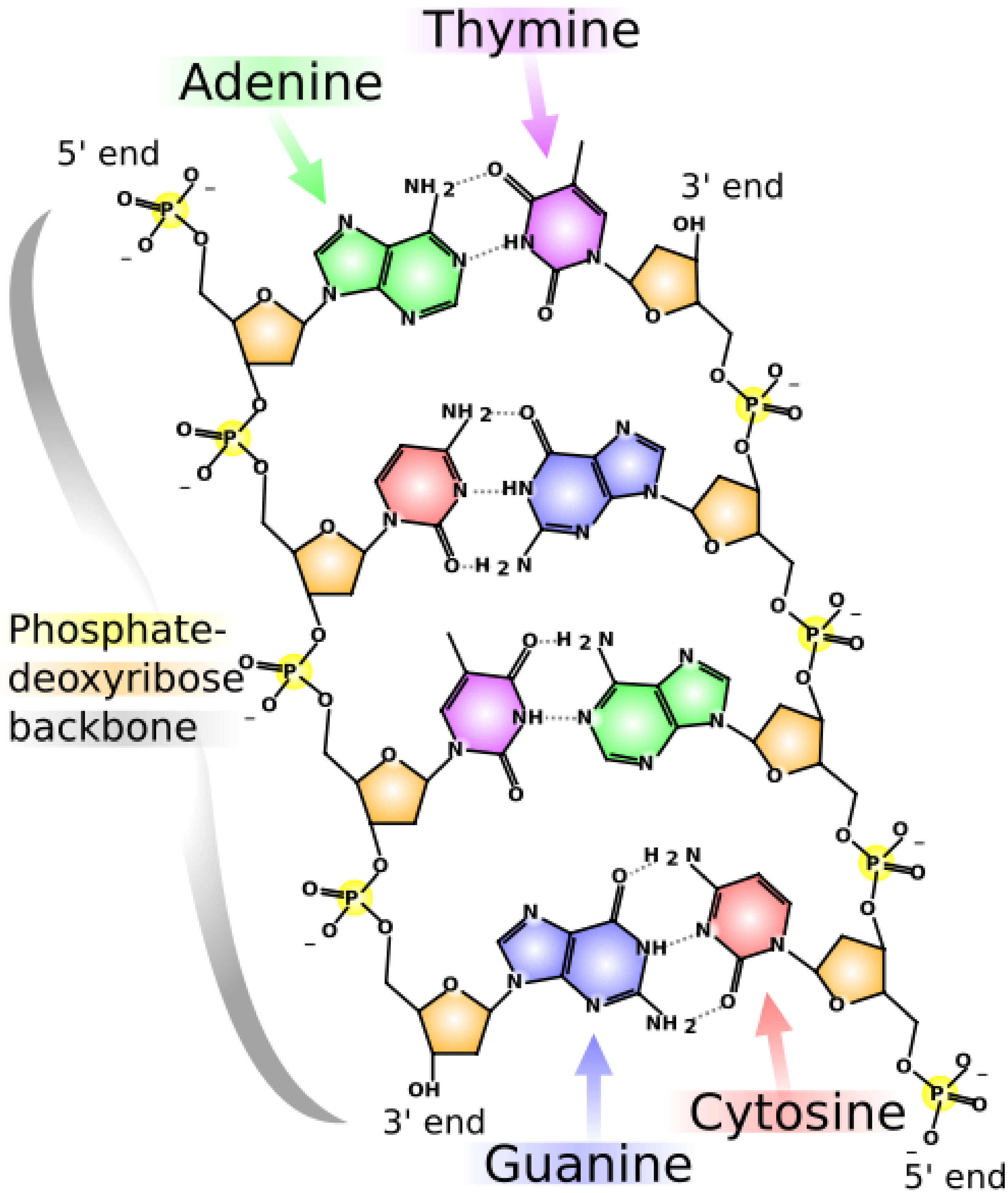
$\ell_p(\text{dsDNA}) \simeq 50\text{nm}$

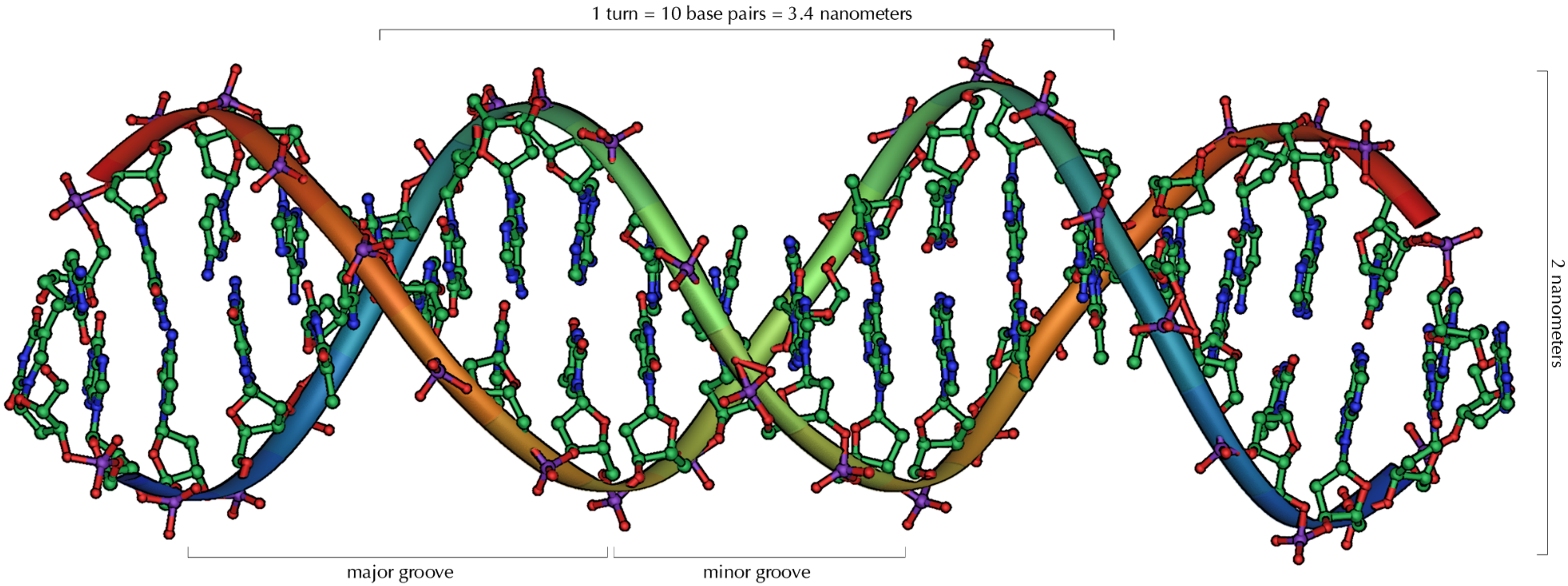
$\ell_p(\text{ssDNA}) \simeq 1\text{nm}$

ϕ29-phage	6μm
E.coli	3mm
Human cell	2m
Sth Amer lungfish	35m

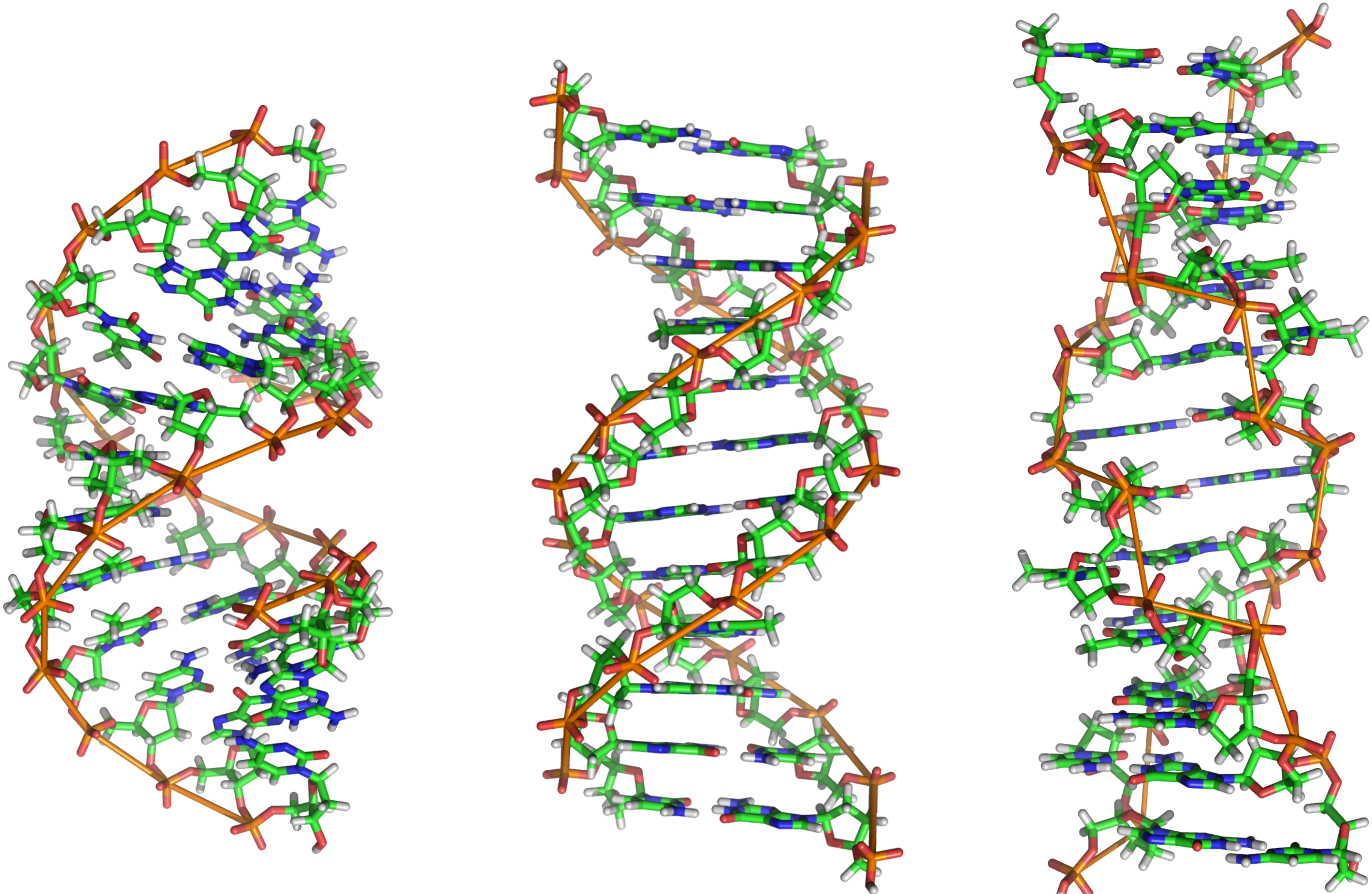
Frank-Kamenetskii, Unravelling DNA

www.imb-jena.de Image Library of Biological Macromolecules

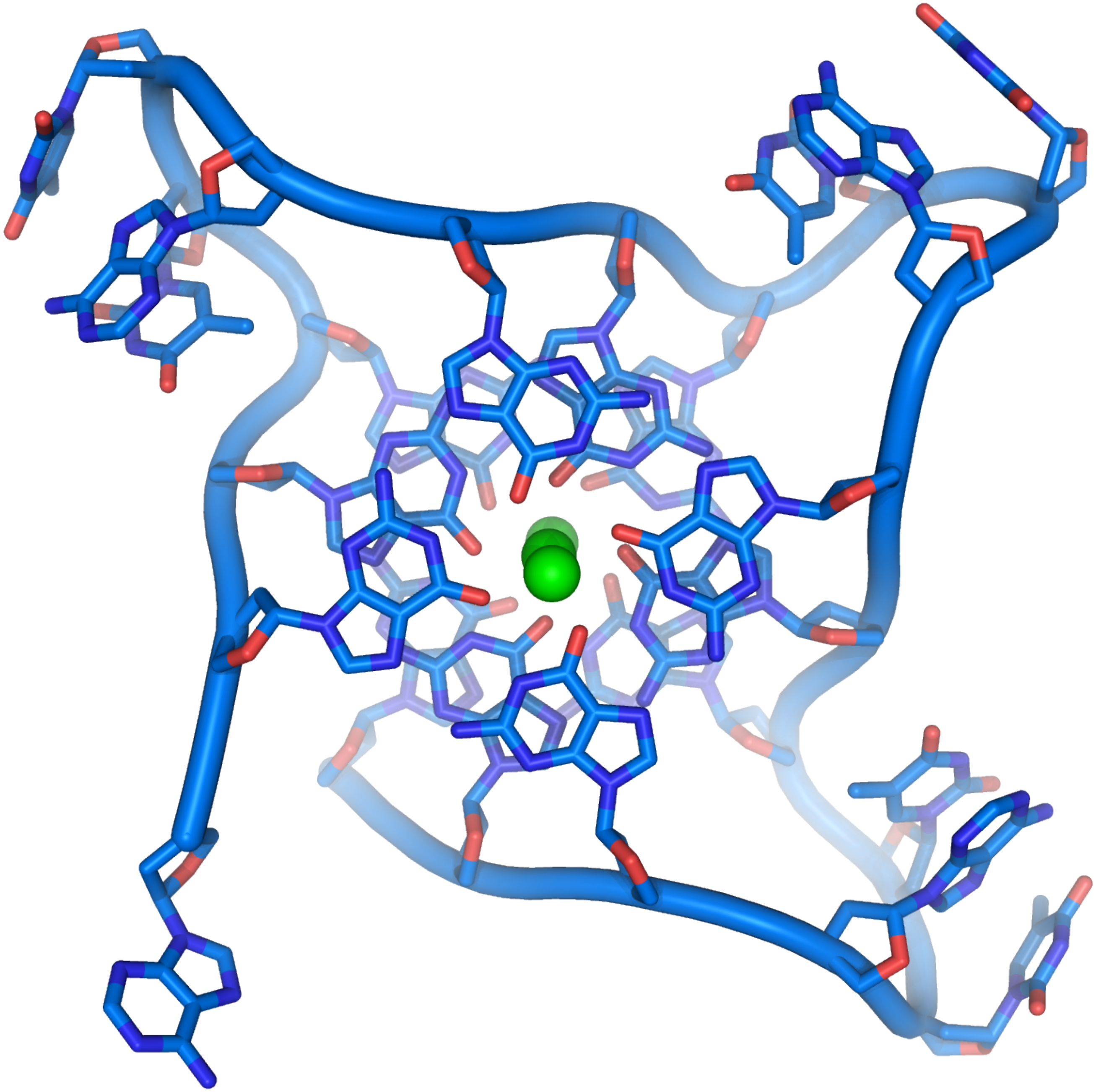

Thymine
Adenine
5' end
3' end
Phosphate-
deoxyribose
backbone
3' end
Guanine
Cytosine
5' end


1 turn = 10 base pairs = 3.4 nanometers
2 nanometers
major groove
minor groove

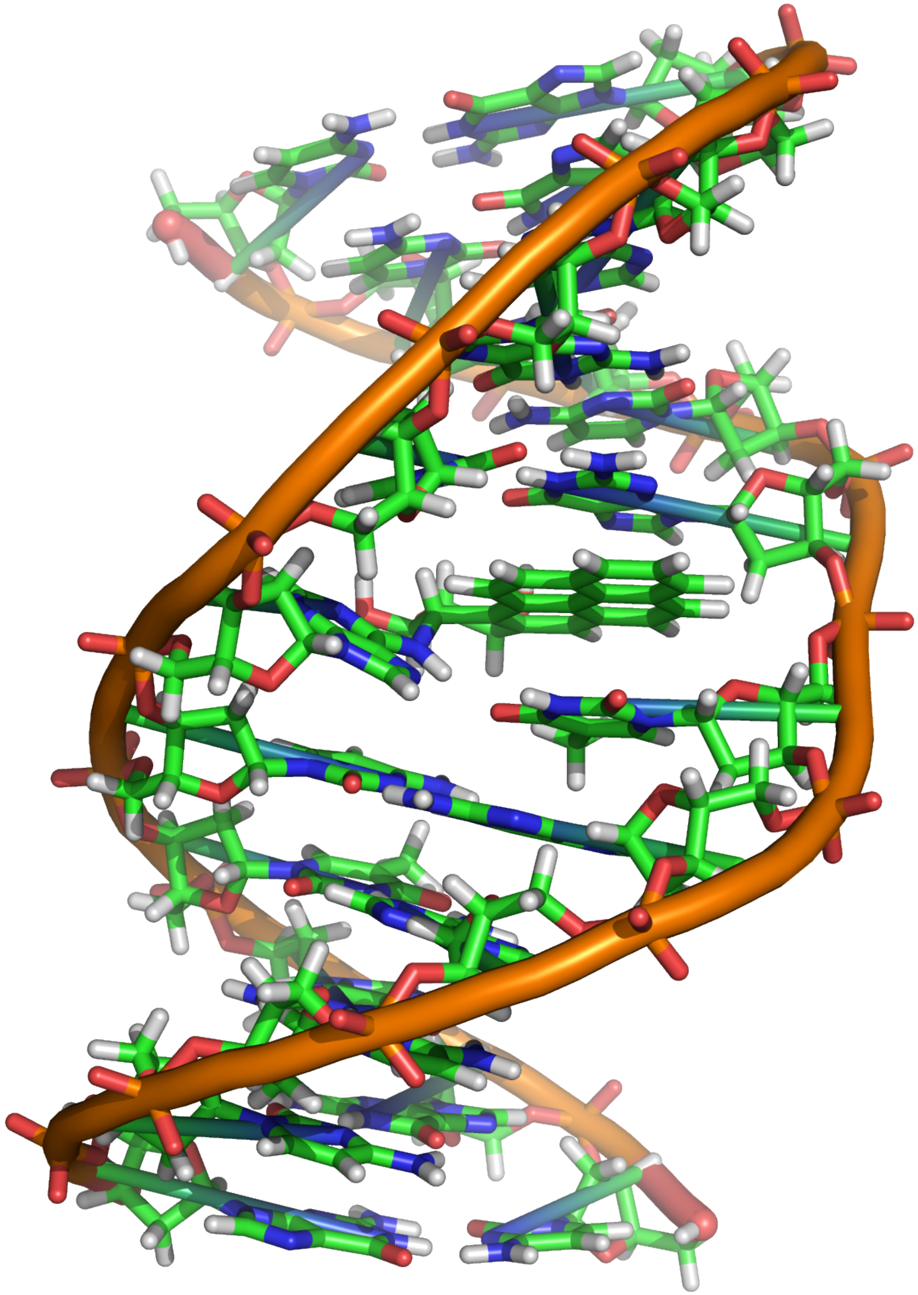
A-DNA, B-DNA, Z-DNA



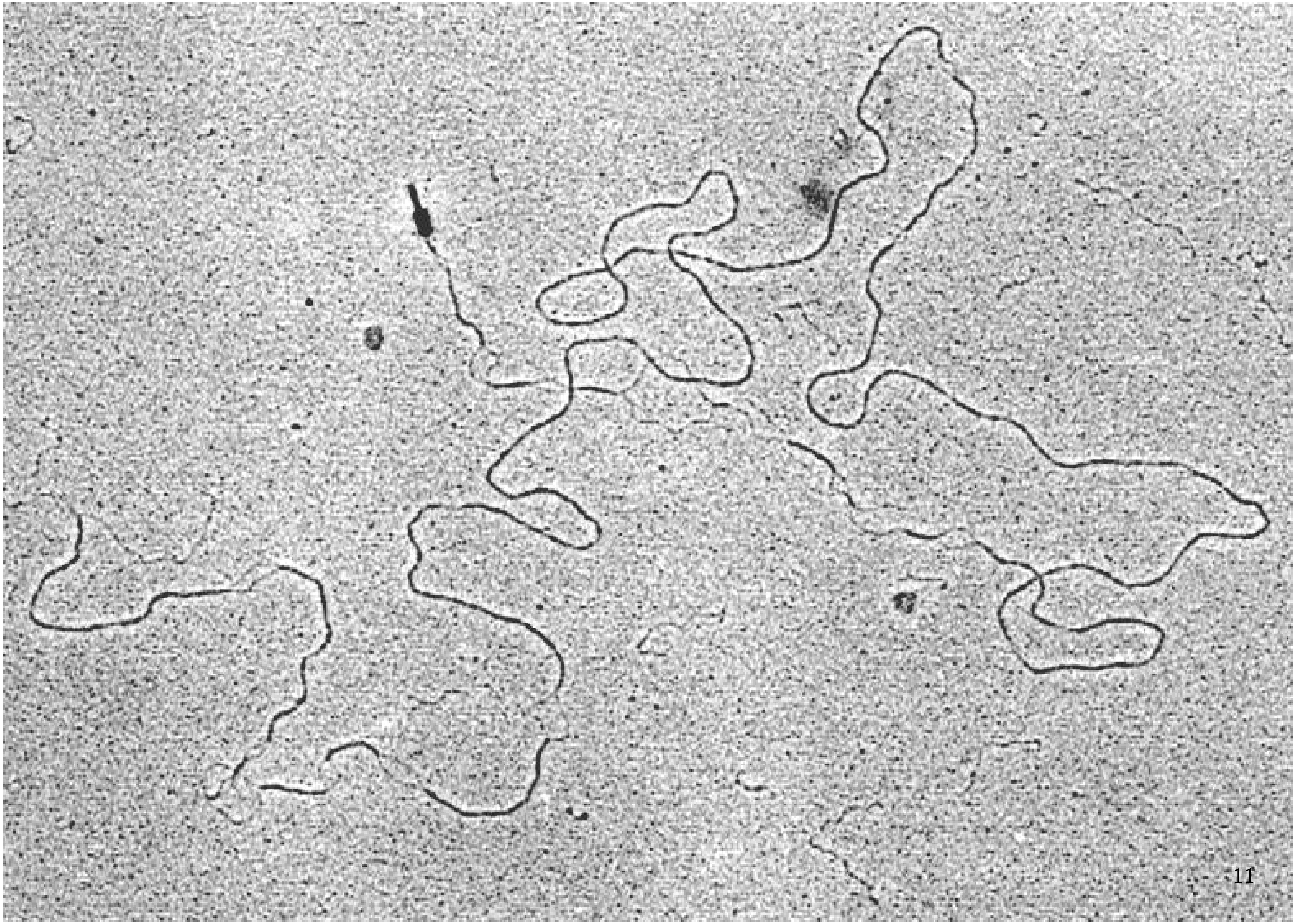
Telomeric quadruplex DNA GC-rich, alternative to telomere loop



DNA mutation induced by tobacco benzopyrene

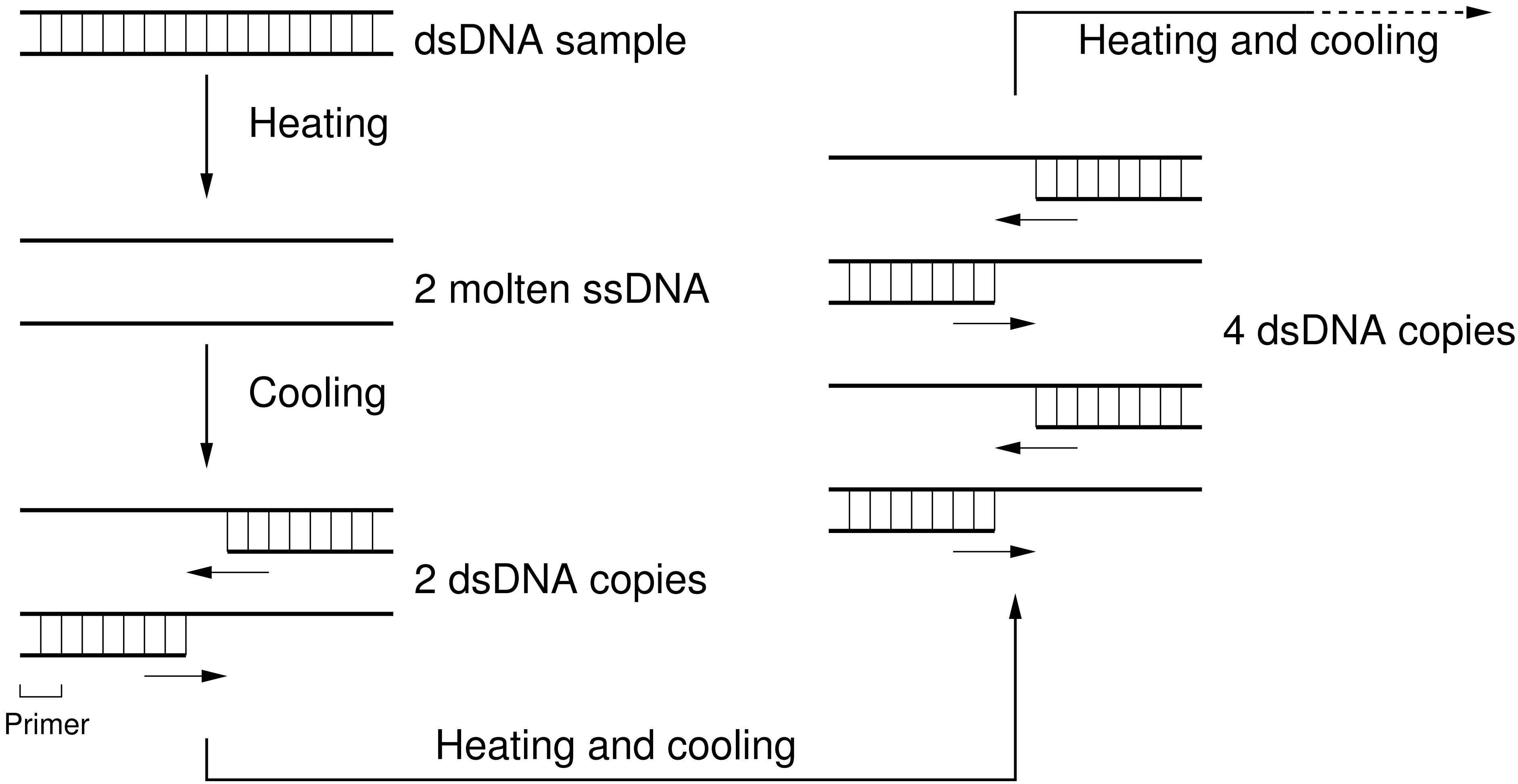


Packaging of DNA in bacteria



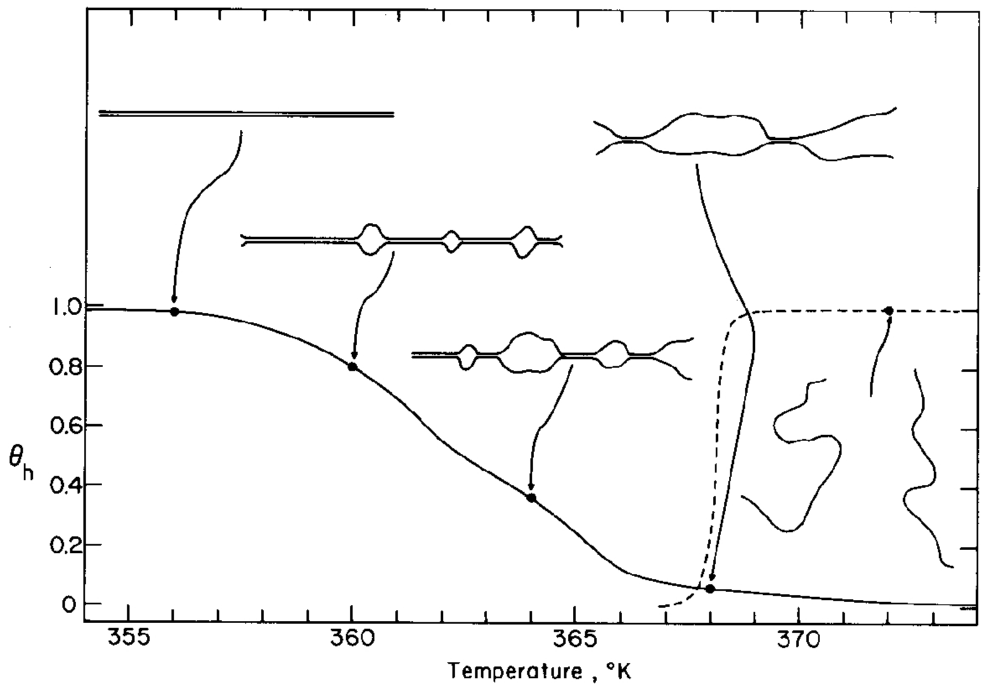
DNA melting

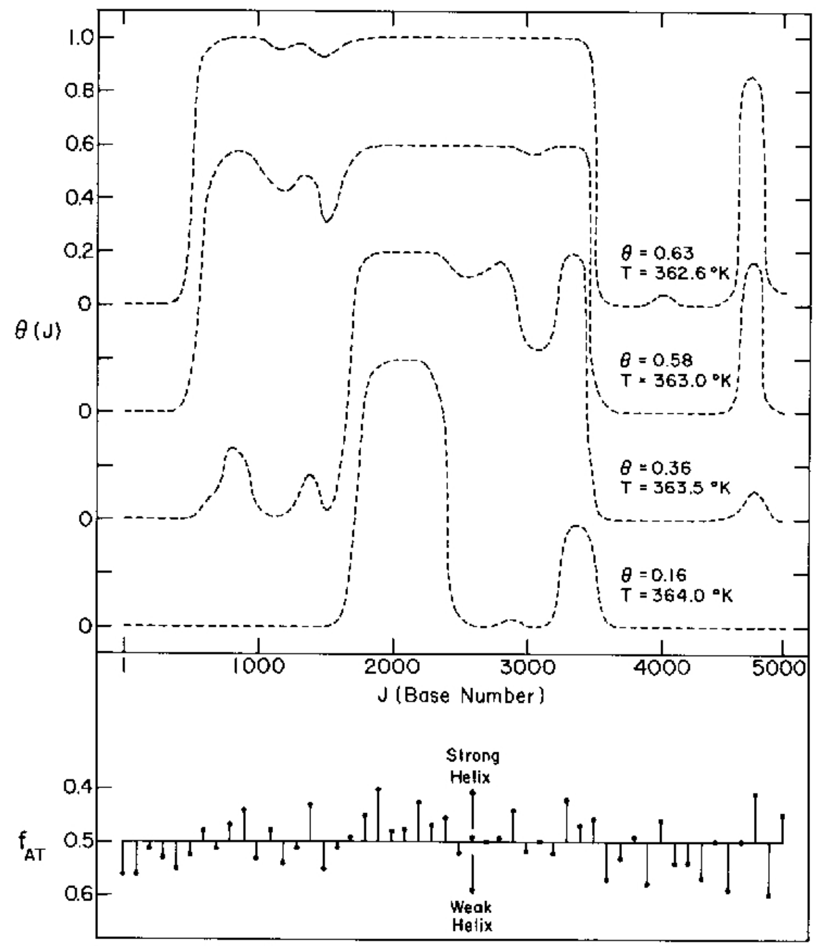
Polymerase chain reaction



DNA melting in bulk solution (UV absorption)

Thermal melting profile:

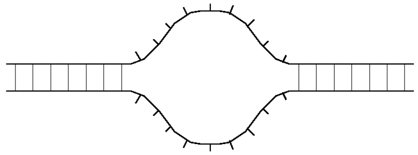




DNA stability and function

Partition factor for bubble of m broken bps:

$$\mathscr{Z}(m) = \sigma_0 u^m (1+m)^{-c}$$



$$u = \exp\Big(\beta[\Delta G_{ij} + \mathfrak{T}\theta_0]\Big) \therefore \theta_0 = \tfrac{2\pi}{10.35}$$

$$\substack{\cdot\mathrm{AA}\cdot \\ \cdot\mathrm{TT}\cdot} \quad \Delta G = -8.45\frac{\mathrm{kcal}}{\mathrm{mol}} + \underbrace{24.86\frac{\mathrm{cal}}{\mathrm{mol}\cdot\mathrm{K}}T}_{7.72\mathrm{kcal/mol}@37^\circ\mathrm{C}}$$

$$\substack{\cdot\mathrm{AA}\cdot \\ \cdot\mathrm{TT}\cdot} \quad T_m \approx 68^\circ\mathrm{C} \;\wedge\; \substack{\cdot\mathrm{GG}\cdot \\ \cdot\mathrm{CC}\cdot} \quad T_m \approx 102^\circ\mathrm{C}$$

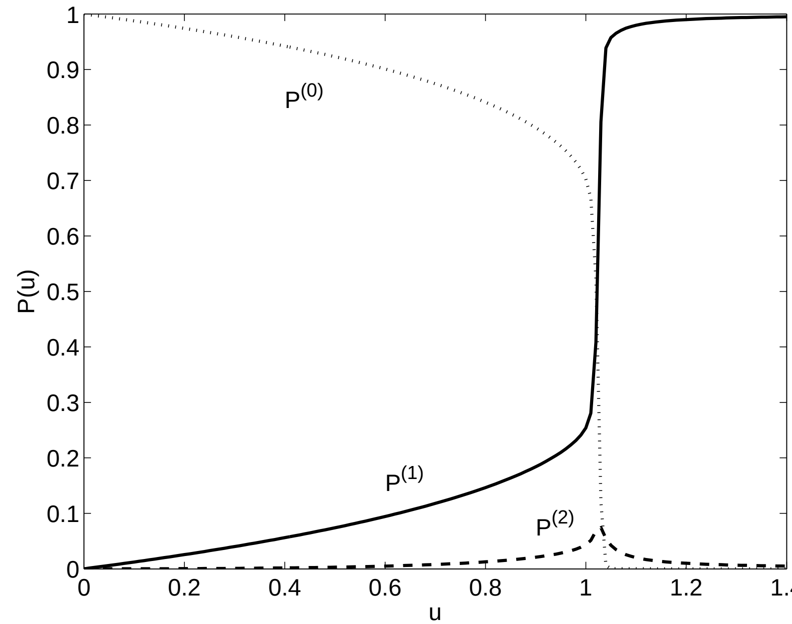
$$\substack{\cdot\mathrm{TA}\cdot \\ \cdot\mathrm{AT}\cdot} \; \Delta G = 0.1 k_B T \wedge \substack{\cdot\mathrm{GC}\cdot \\ \cdot\mathrm{CG}\cdot} \; \Delta G = -3.9 k_B T$$

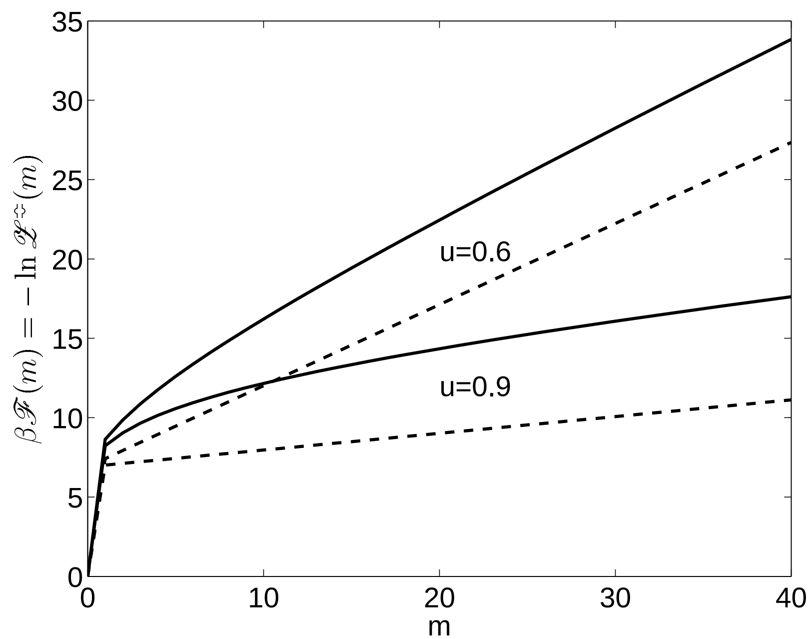
Loop initiation: $\sigma_0 \simeq 10^{-3\ldots-5} \triangleq 7\ldots12 k_B T$

Loop closure exponent: $c \approx 1.76$ (1)

Zipping rate: $k^{-1} \simeq 20\ldots100\,\mu\mathrm{sec}$

Bubble lifetime: $\tau_{\mathrm{bubble}} \simeq 1\,\mathrm{msec}$





Poland & Scheraga, *Theory of helix-coil transitions*; Krueger et al, Biophys J (2006)

Cooperativity factor σ_0 or ring factor ϵ

Classically,

$$\sigma_0 = \exp\left(-\frac{F_s}{RT}\right) \approx 10^{-5} \ldots 10^{-4}, \quad F_s \approx 8\text{kcal/mol}$$

Claim: $F_s = T\Delta S$:

$$\exp\left(-\frac{\Delta S_{\mathrm{TA/AT}}}{R}\right) = 1.19 \times 10^{-5},\ \text{Heteropolymer, best value: } \sigma_0 = 1.26 \times 10^{-5}$$

$$\exp\left(-\frac{\Delta S_{\mathrm{AT/GC}}}{R}\right) = 3 \times 10^{-6},\ \text{GC-rich: } \sigma_0 = 3.5 \times 10^{-6}$$

$$\exp\left(-\frac{\Delta S_{\mathrm{GC/GC}}}{R}\right) = 9.7 \times 10^{-7},\ \text{GC homopolymer}$$

Ring factor (nicked DNA!):

$$\sigma_0 = \xi \exp\left(-\frac{\Delta G^{\mathrm{ST}}}{RT}\right)$$

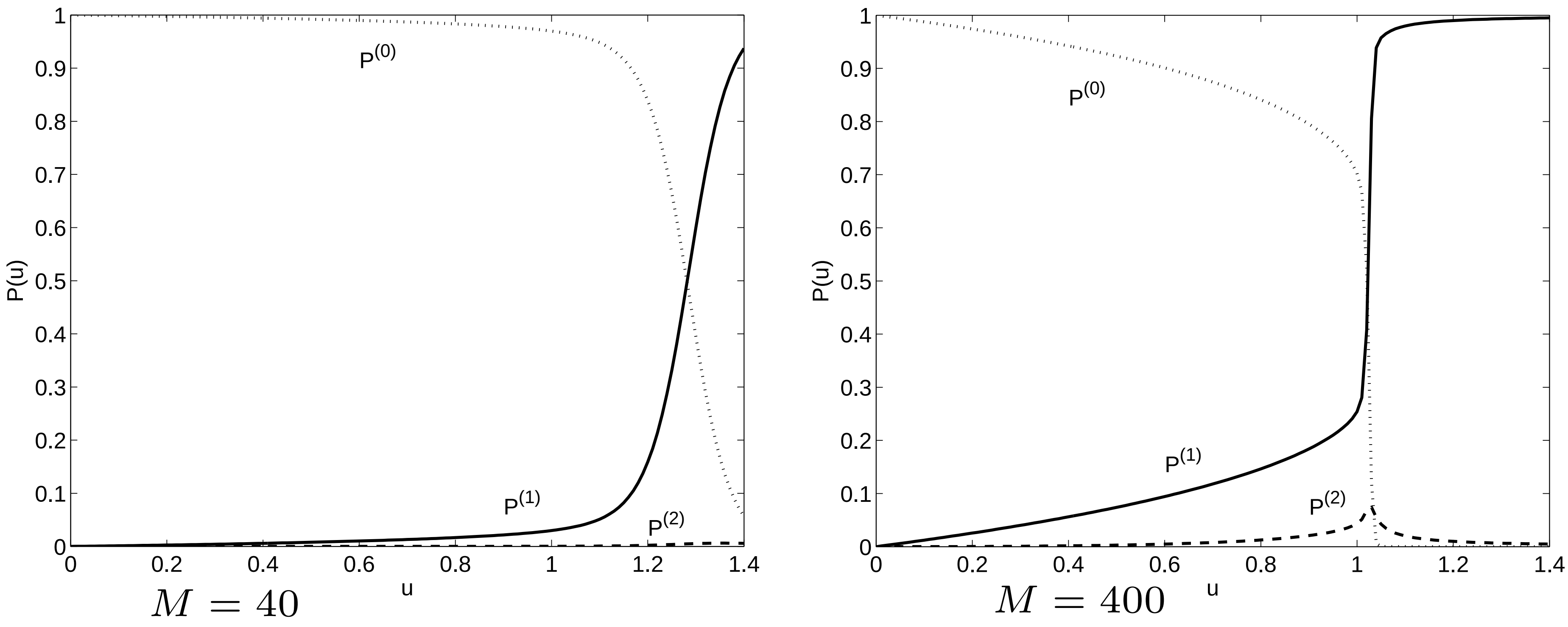
such that for the lowest stacking free energy, $\sigma_0 \approx 5.6 \times 10^{-4}$

Poland-Scheraga Free Energy

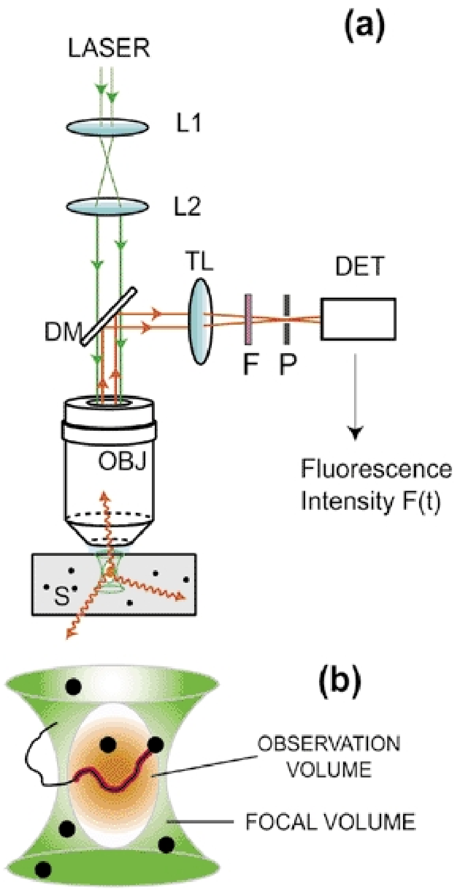
$$\mathscr{Z}(M) = 1 + \sigma_0 \mathscr{Z}^{(1)}(M) + \sigma_0^2 \mathscr{Z}^{(2)}(M) + \dots$$

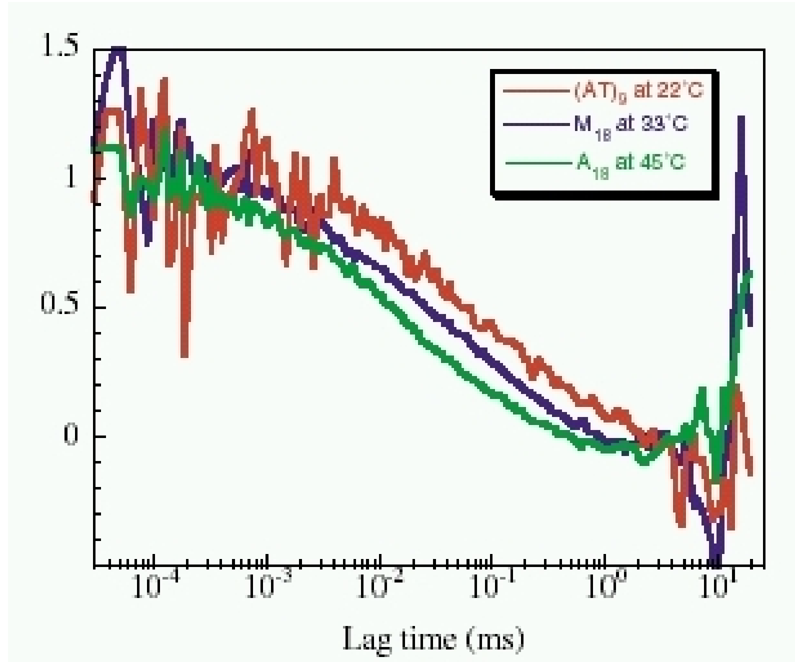
$$\mathscr{Z}^{(1)}(M) = \sum_{m=1}^{M} (M - m + 1) g(m) \; \therefore \; g(m) = u^m (1+m)^{-c}$$

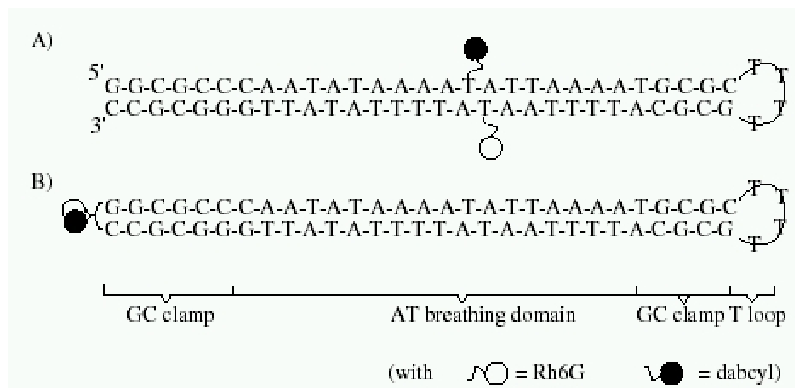
$$\mathscr{Z}^{(2)}(M) = \frac{1}{2} \sum_{m=1}^{M-1} \sum_{m'=1}^{M-m-1} (M - m - m' + 1)(M - m - m') g(m) g(m')$$



Watching a single DNA breathe







Altan-Bonnet, Libchaber, Krichevsky, Phys Rev Lett (2003)

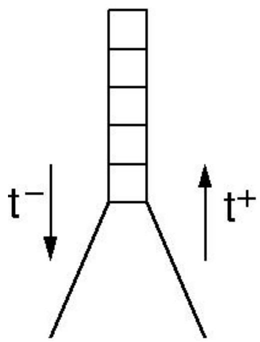
Master equation for bubble breathing

$$\frac{\partial}{\partial t}P(m,t) = \mathrm{t}^+(m-1)P(m-1,t) + \mathrm{t}^-(m+1)P(m+1,t) - \Big(\mathrm{t}^+(m) + \mathrm{t}^-(m)\Big)P(m,t)$$

Unzipping rate:

$$\mathrm{t}^+(m) = ku\left(\frac{1+m}{2+m}\right)^c$$

$$\therefore\ \mathrm{t}^+(0) = 2^{-c}k\sigma_0 u$$



Zipping rate:

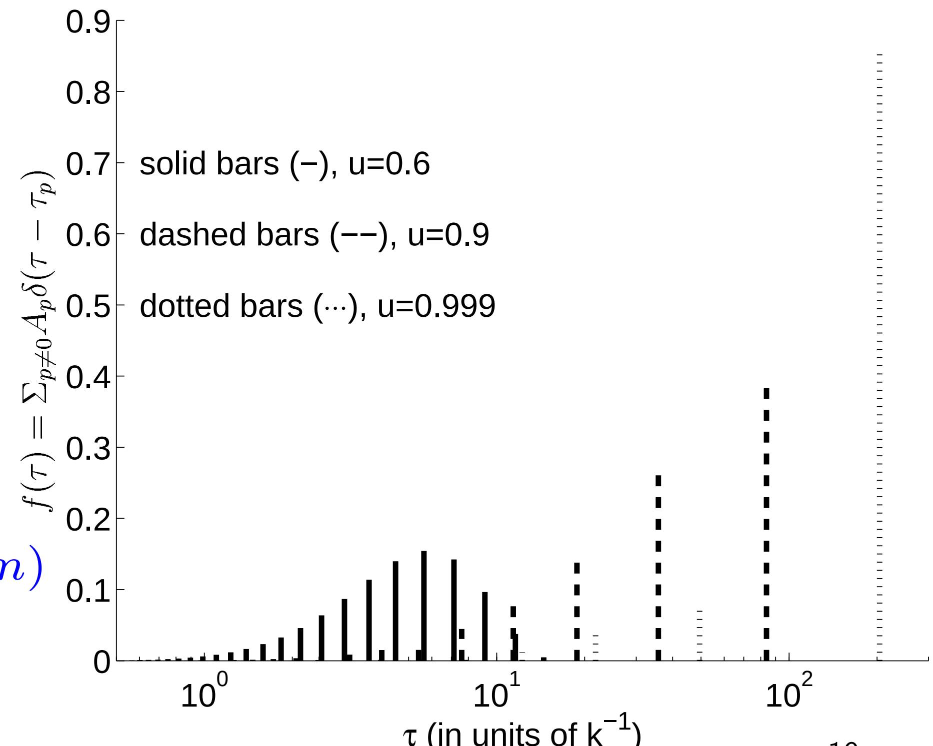
$$\mathrm{t}^-(m,n) = k$$

Detailed balance:

$$\mathscr{Z}^{\diamond}(m) = \sigma_0 u^m (1+m)^{-c}$$

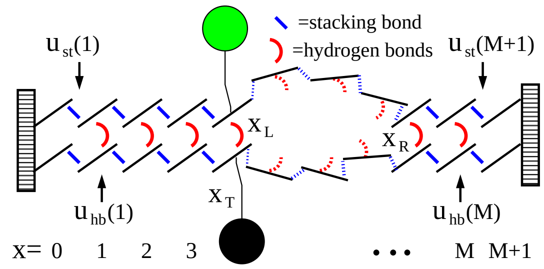
$$\mathrm{t}^+(m-1)\mathscr{Z}^{\diamond}(m-1) = \mathrm{t}^-(m)\mathscr{Z}^{\diamond}(m)$$

$$P_{\mathrm{eq}}(m)/P_{\mathrm{eq}}(m') = \mathscr{Z}^{\diamond}(m)/\mathscr{Z}^{\diamond}(m')$$



T. Ambjörnsson & RM, J Phys Cond Mat (2005)

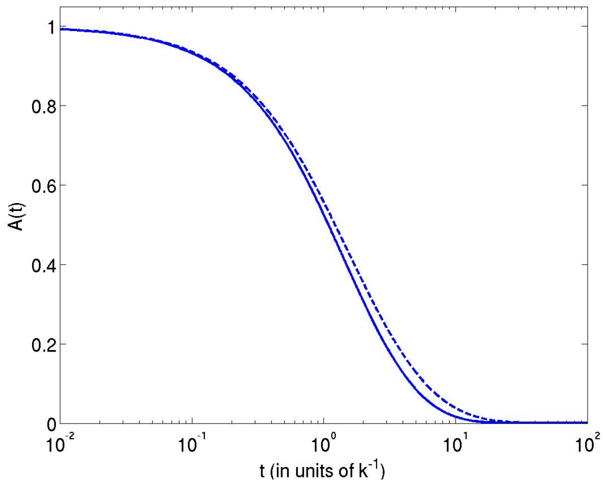
Experiment measures a tagged base-pair:



Random variable $I(t) \in \{0, 1\}$

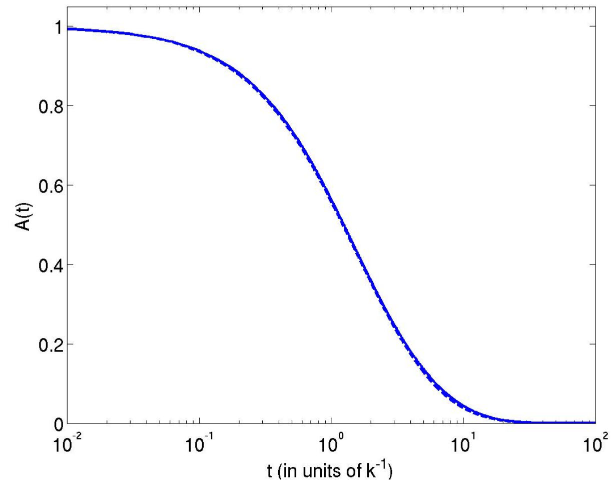
(Similar if $x' \pm \Delta$ broken bps necessary for signal)

Size $\langle m(t)m(0)\rangle - \langle m\rangle^2$ (- - -) vs blinking autocorrelation $\langle I(t)I(0)\rangle - \langle I\rangle^2$ (—):



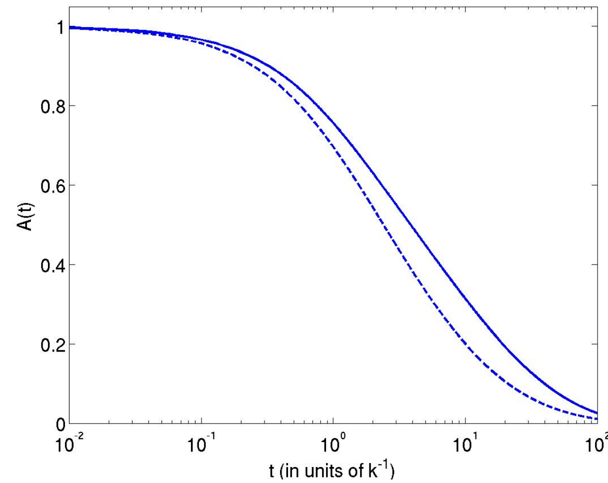
Tag close to clamp

$u = 0.6$



Centre-tag

$u = 0.6$

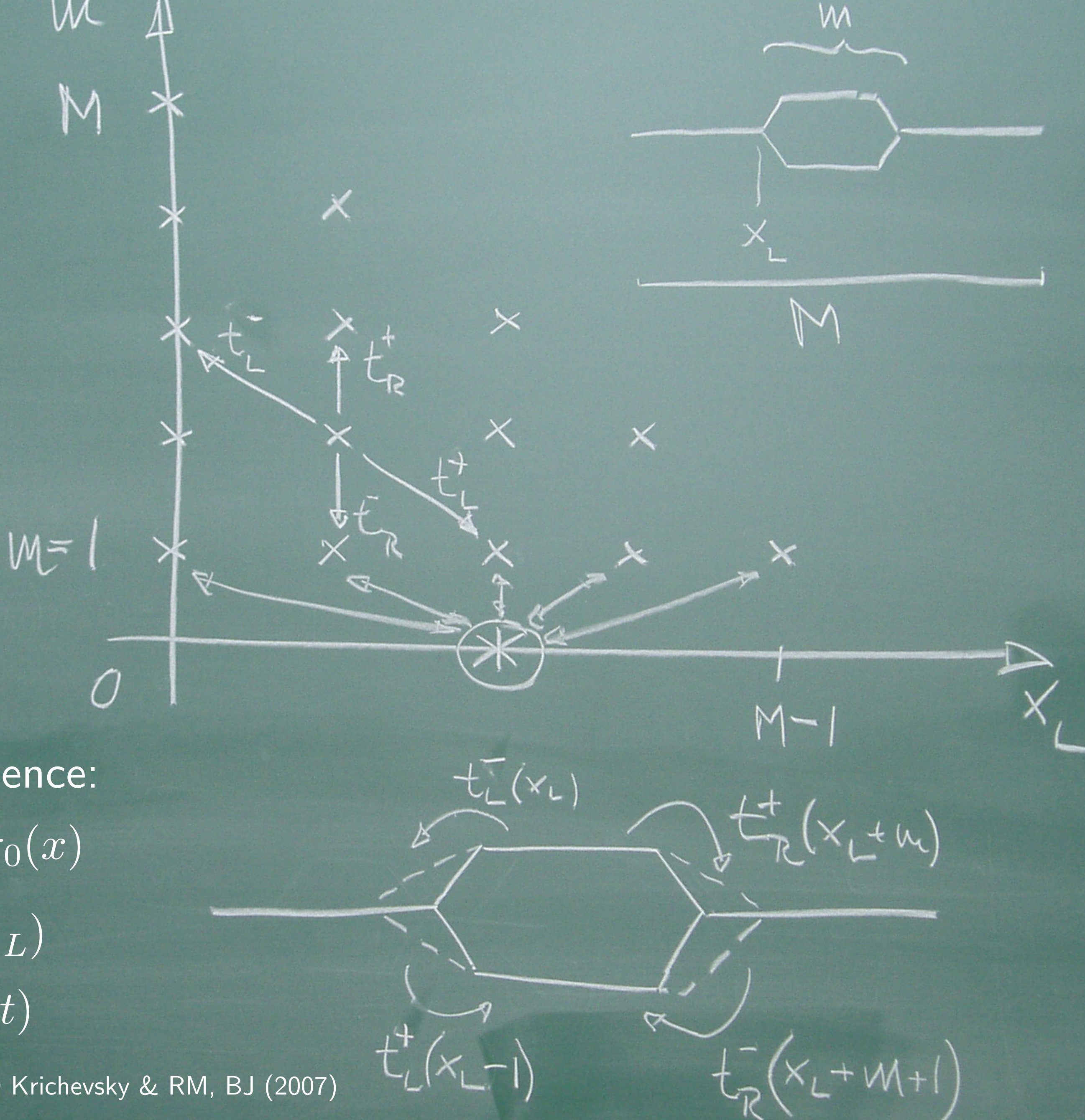


Centre-tag

$u = 0.9$

T Ambjörnsson, SK Banik, O Krichevsky & RM, in prep

Heteropolymer case:



Sequence dependence:

$u, \sigma_0 \rightarrow u(x), \sigma_0(x)$

$\mathsf{t}^{\pm} \rightarrow \mathsf{t}^{\pm}_{R/L}(x_{R/L})$

$P \rightarrow P(m, x_L, t)$

T Ambjörnsson, SK Banik, O Krichevsky & RM, BJ (2007)

Partition sum for heteropolymeric DNA

$$\mathscr{Z}(x_L, m) = \frac{2^c \xi}{(1+m)^c} \prod_{x=x_L+1}^{x_L+m} u_{\text{hb}}(x) \prod_{x=x_L+1}^{x_L+m+1} u_{\text{st}}(x)$$

Where the weights correspond to the free energy of base pairing (2x) and stacking (10x):

$$u_{\text{hb}}(x) = \exp\{-\epsilon_{\text{hb}}(x)/[k_B T]\}, \; u_{\text{st}}(x) = \exp\{-\epsilon_{\text{hb}}(x)/[k_B T]\}$$

Values @ 37 °C and 100 mM NaCl:

$$\epsilon_{\text{hb}}(\text{AT}) = 1.0 k_B T, \; \epsilon_{\text{hb}}(\text{GC}) = 0.2 k_B T$$

$$\epsilon_{\text{st}}(\text{TA/AT}) = -0.9 k_B T, \; \epsilon_{\text{st}}(\text{GC/CG}) = -4.1 k_B T$$

Note that @ $T = 37$°C, $k_B T = 0.62$kcal/mol

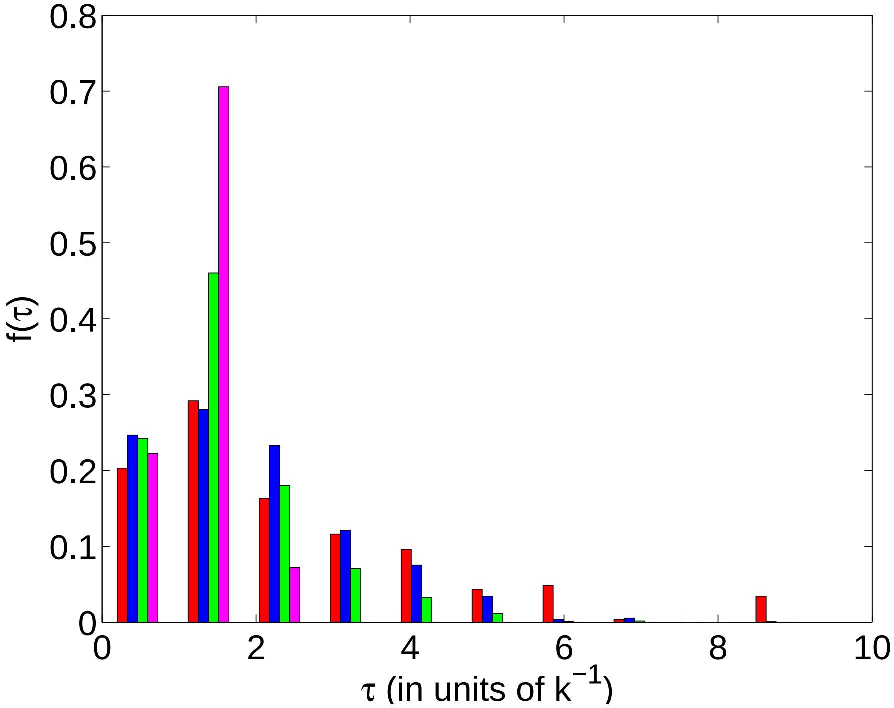
Krueger et al, Biophys J (2006)

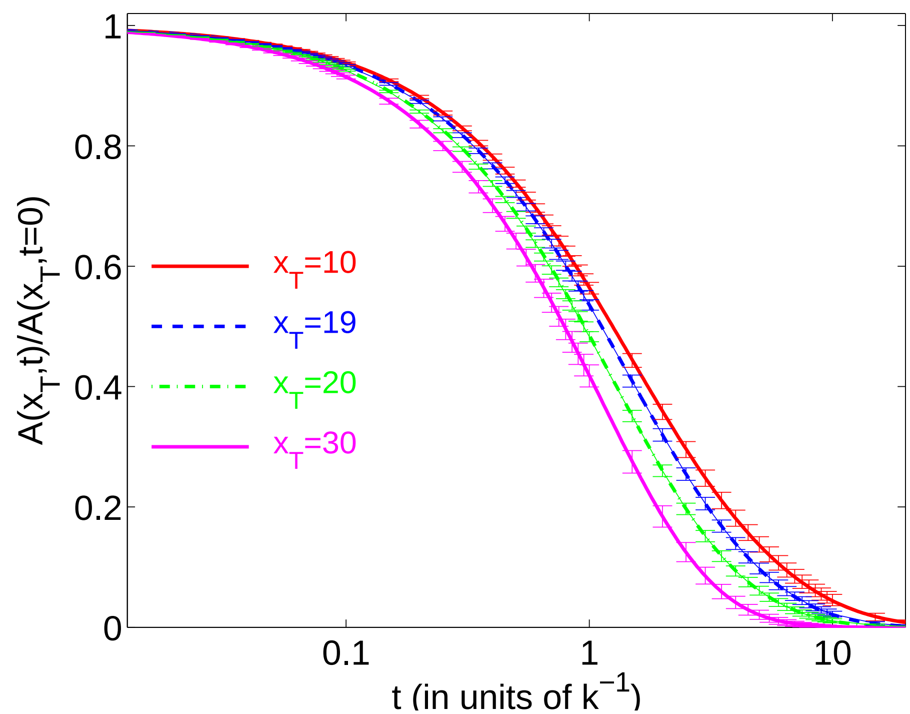
Table 1. Stacking ΔG_{KL}^{ST} and base-pairing ΔG^{BP} parameters used in our calculations*

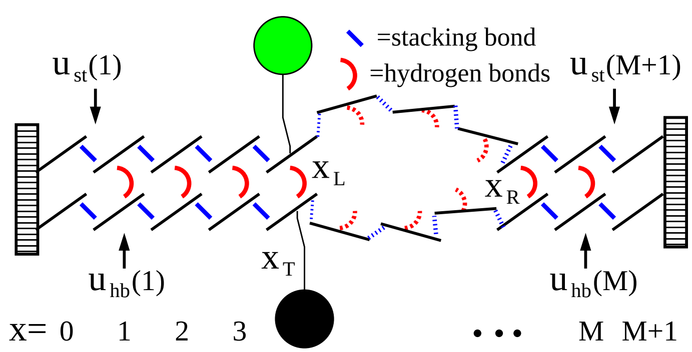
ΔG_{KL}^{ST}	$^{5'}$KL	A	T	G	C
5'	A	–1.50	–1.73	–1.45	–2.20
	T	–0.58	–1.50	–0.94	–1.82
	G	–1.82	–2.20	–1.83	–2.56
	C	–0.94	–1.45	–1.30	–1.83
ΔG^{BP}	A·T		0.65		
	G·C		0.13		

* Stacking and base-paring parameters in kcal/mol correspond to 37°C, 0.1 *M* NaCl.

Breathing dynamics is sensitive to sequence







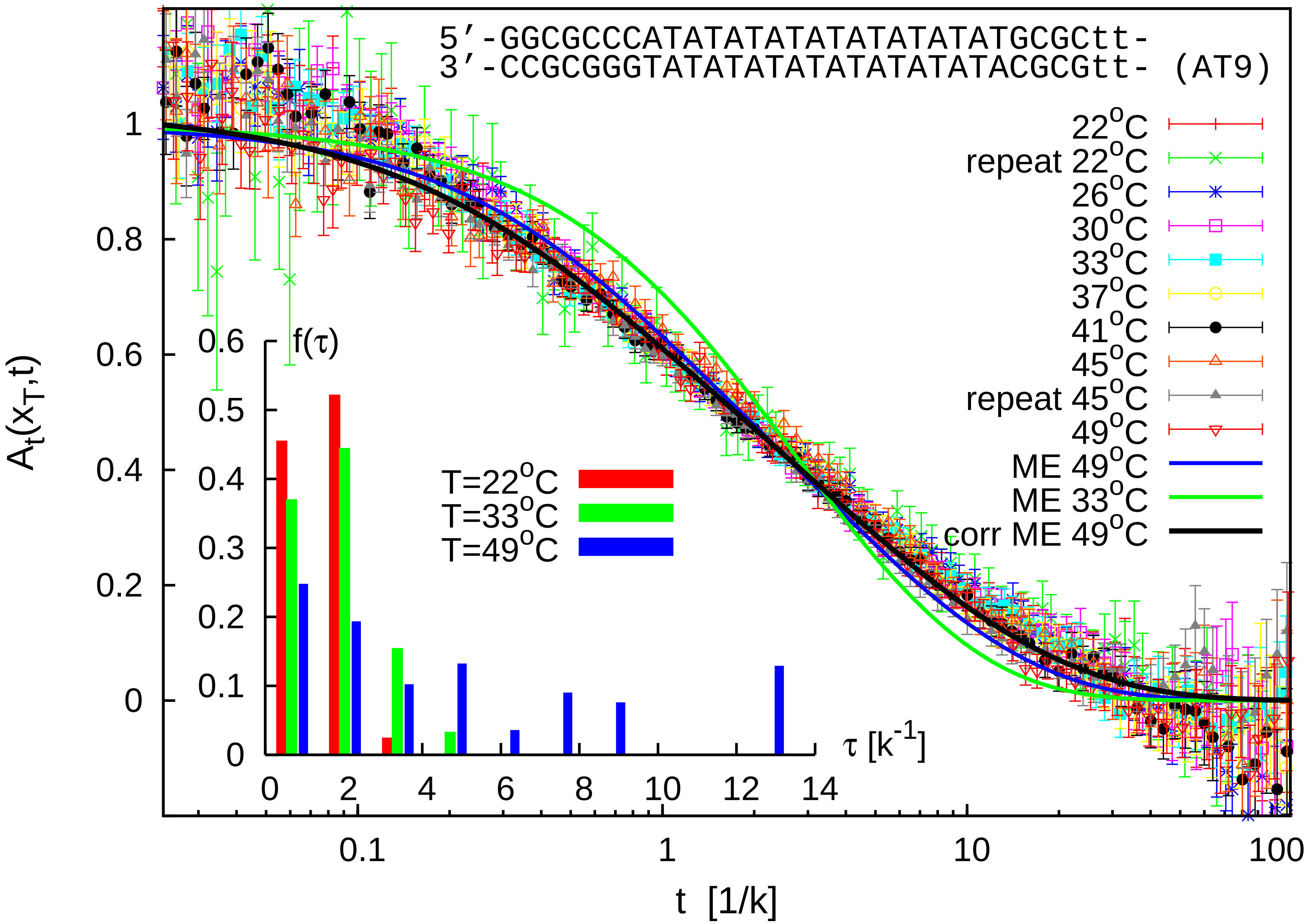
AAAAAAAAAAAAAAAAAAAAGGGGGGGGGGGGGGGGGGG

TTTTTTTTTTTTTTTTTTTTCCCCCCCCCCCCCCCCCCC

012345678901234567890123456789012345678 9

T Ambjörnsson, SK Banik, O Krichevsky & RM, Biophys J (2007)

Fluorescence autocorrelation function



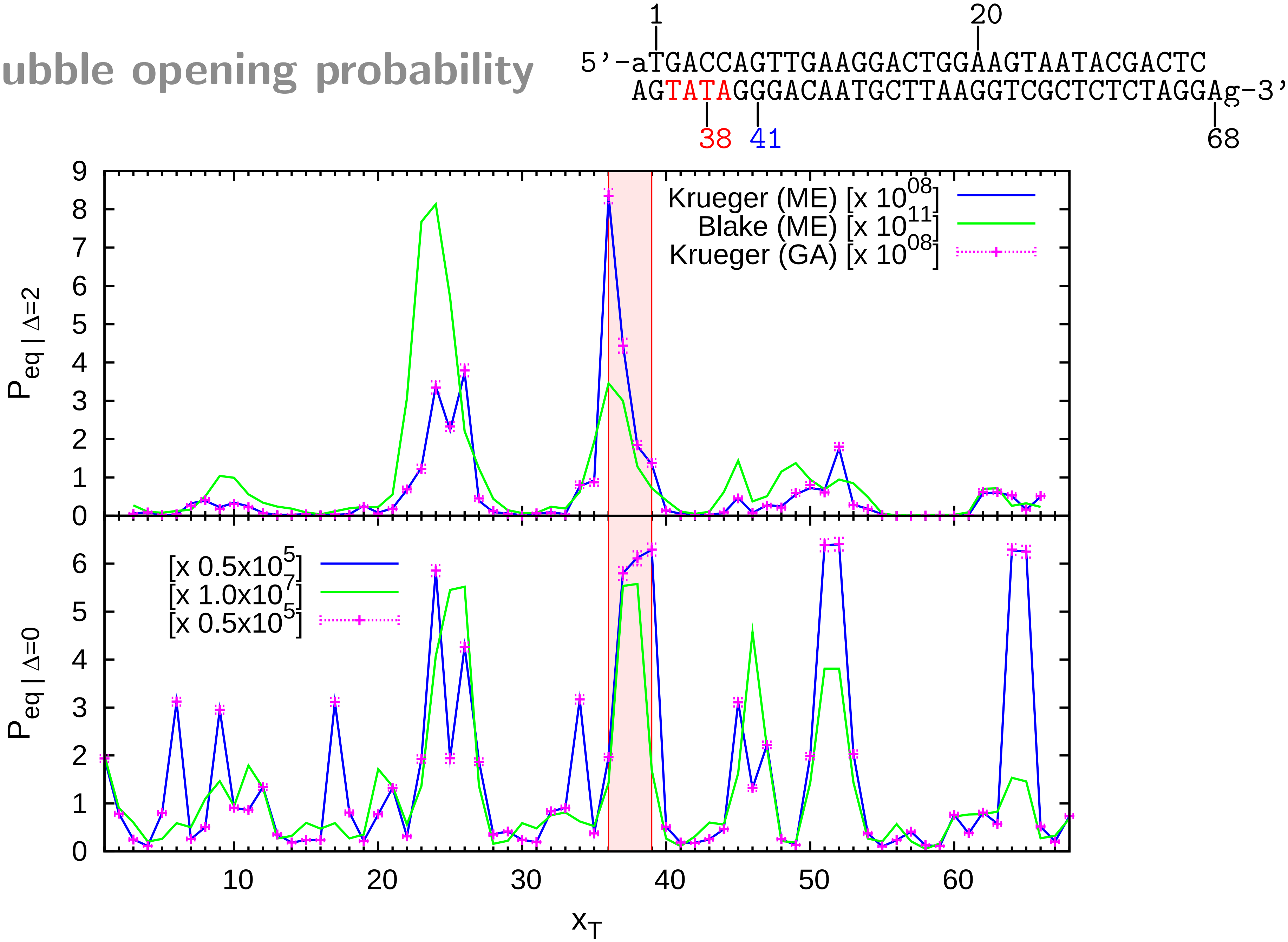
T Ambjörnsson, SK Banik, O Krichevsky & RM, Phys Rev Lett (2006)

Phage T7 promoter sequence

```
    1                  20
    |                  |
5’-aTGACCAGTTGAAGGACTGGAAGTAATACGACTC
   AGTATAGGGACAATGCTTAAGGTCGCTCTCTAGGAg-3’
       |  |                          |
       38 41                         68
```

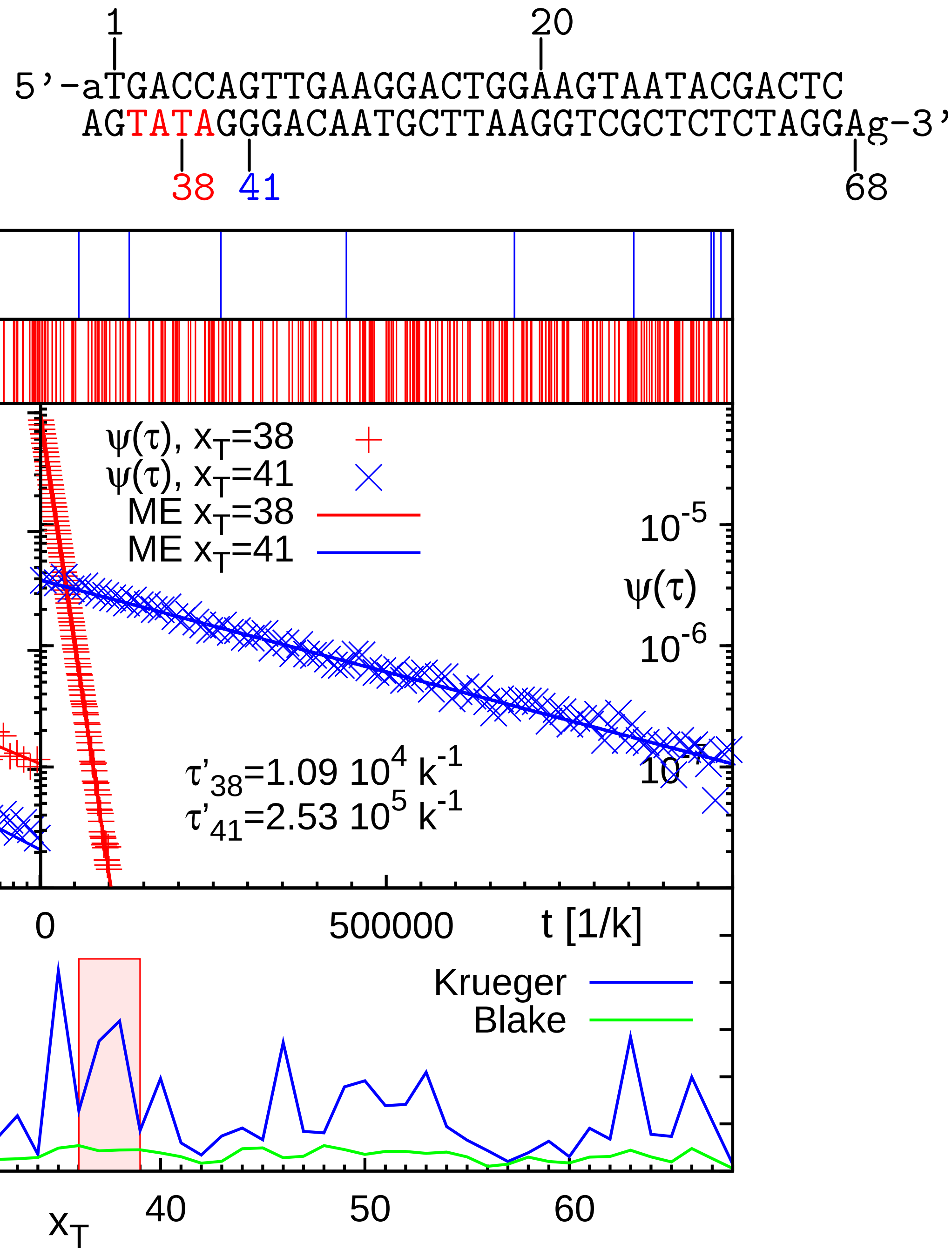
T Ambjörnsson, SK Banik, O Krichevsky & RM, Phys Rev Lett (2006)

Bubble opening probability



T Ambjörnsson, SK Banik, O Krichevsky & RM, Phys Rev Lett (2006)

Fluorescence repeat & time

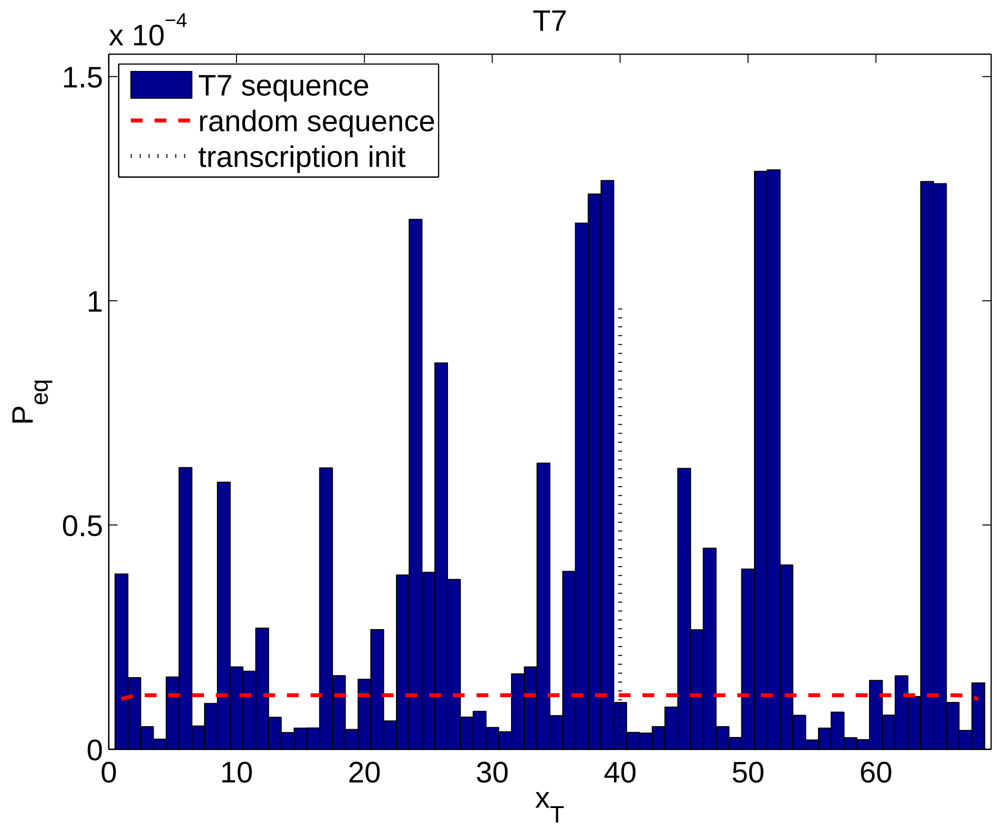


T Ambjörnsson, SK Banik, O Krichevsky & RM, Phys Rev Lett (2006); SK Banik, T Ambjörnsson & RM, Europhys Lett (2005)

Biological relevance of DNA breathing

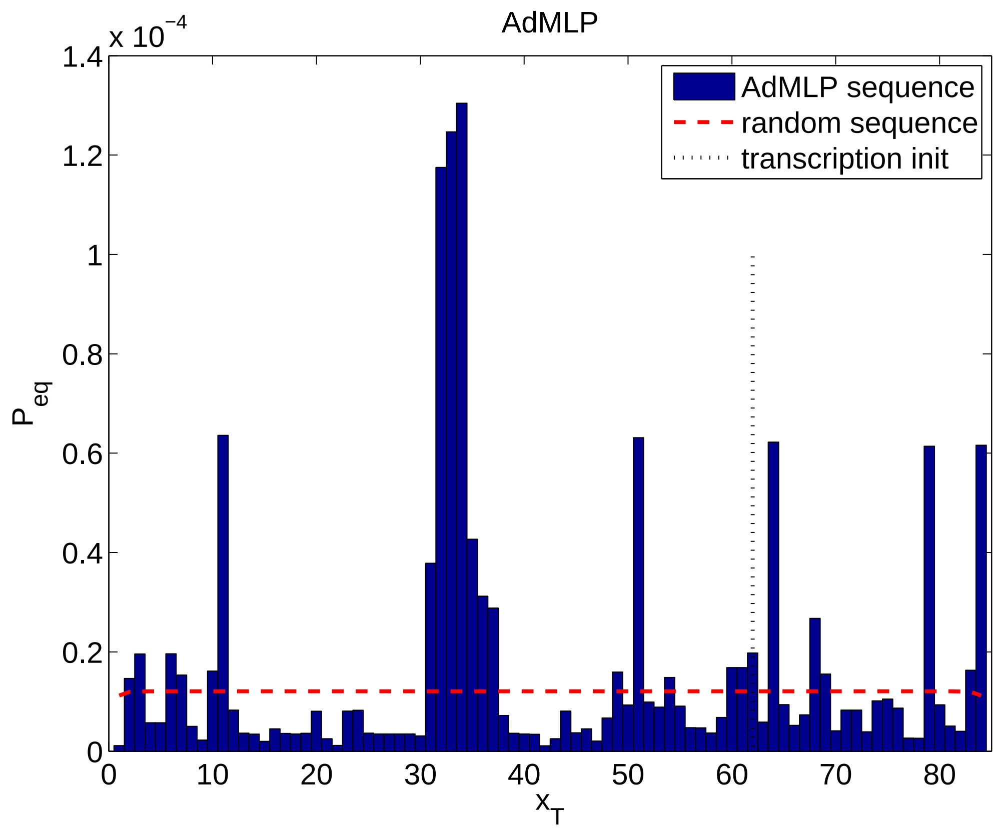
Bacteriophage T7 core promoter:

```
   1                  20
   |                  |
5'-aTGACCAGTTGAAGGACTGGAAGTAATACGACTC
   AGTATAGGGACAATGCTTAAGGTCGCTCTCTAGGAg-3'
      |  |                           |
     38 41                          68
```



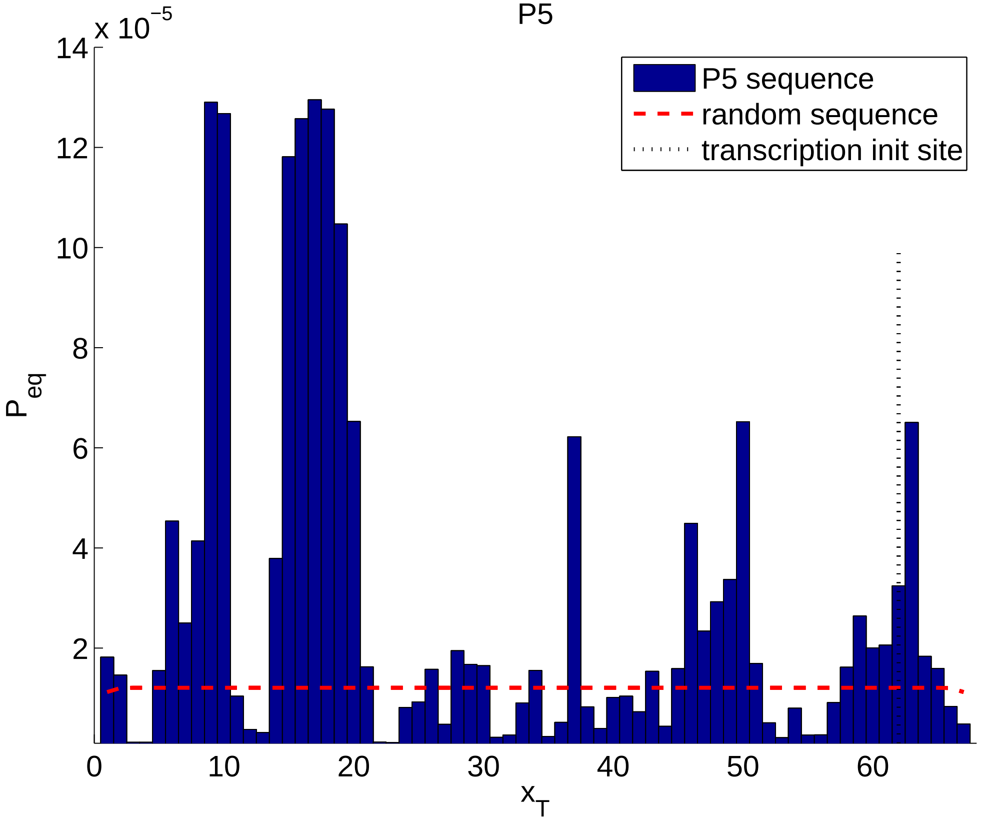
Adenovirus Major Late Promoter:

```
    1                            31
    |                             |
5'- GCCACGTGACCAGGGGTCCCCGCCGGGGGGT
    ATAAAAGGGGGCGGACCTCTGTTCGTCCTC
    ACTGTCTTCCGGATCGCTGTCCAG -3'
   |                      |
  TSS                    84
```

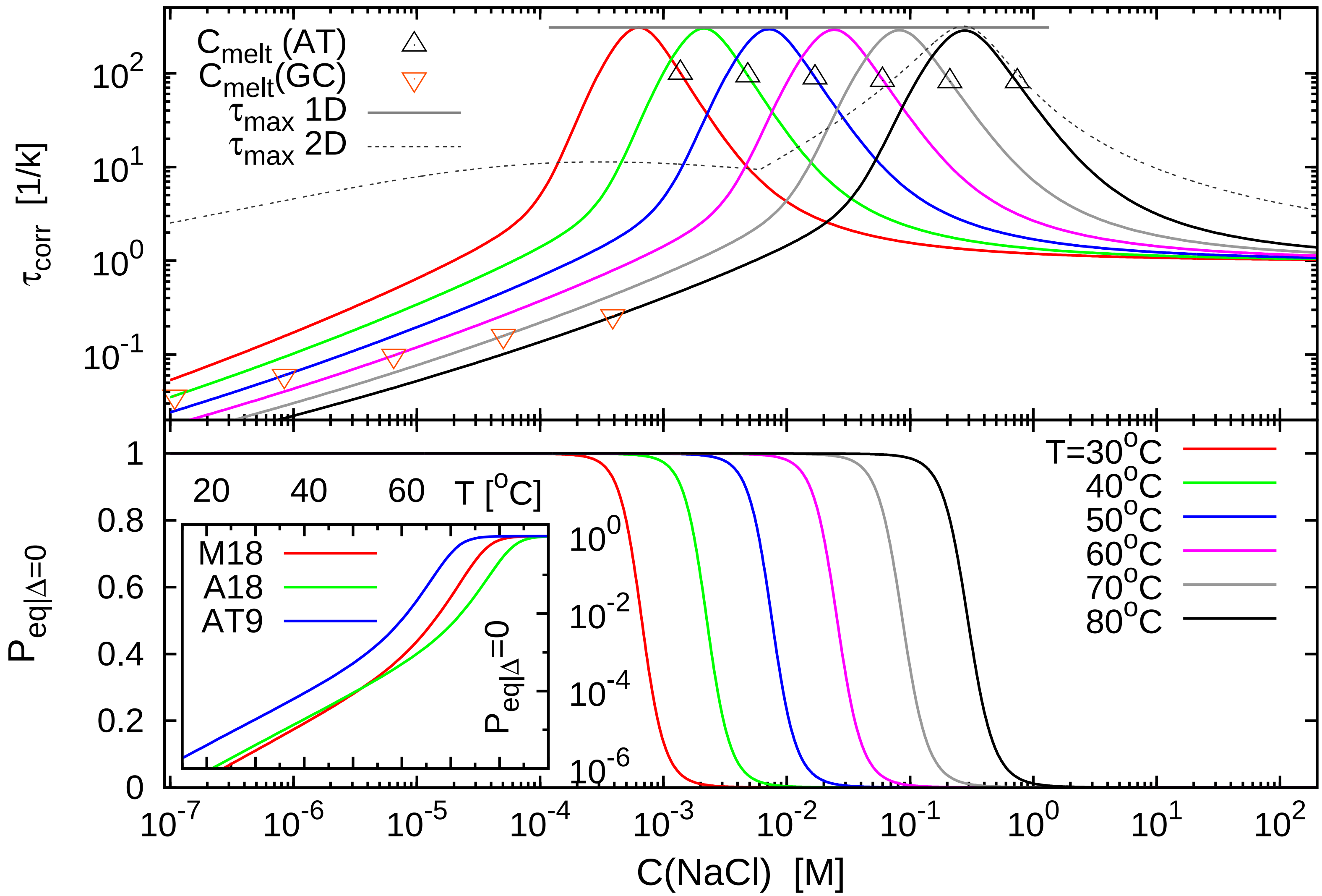


Adeno Associated Viral P5 promoter (YinYang 1 motif):

```
       1                       25
       |                       |
5'- GTGGCCATTTAGGGTATATATGGCCGAGTGAGCGA
    GCAGGATCTCCATTTTGACCGCGAAATTTGAACG -3'
             |                      |
            TSS                    67
```



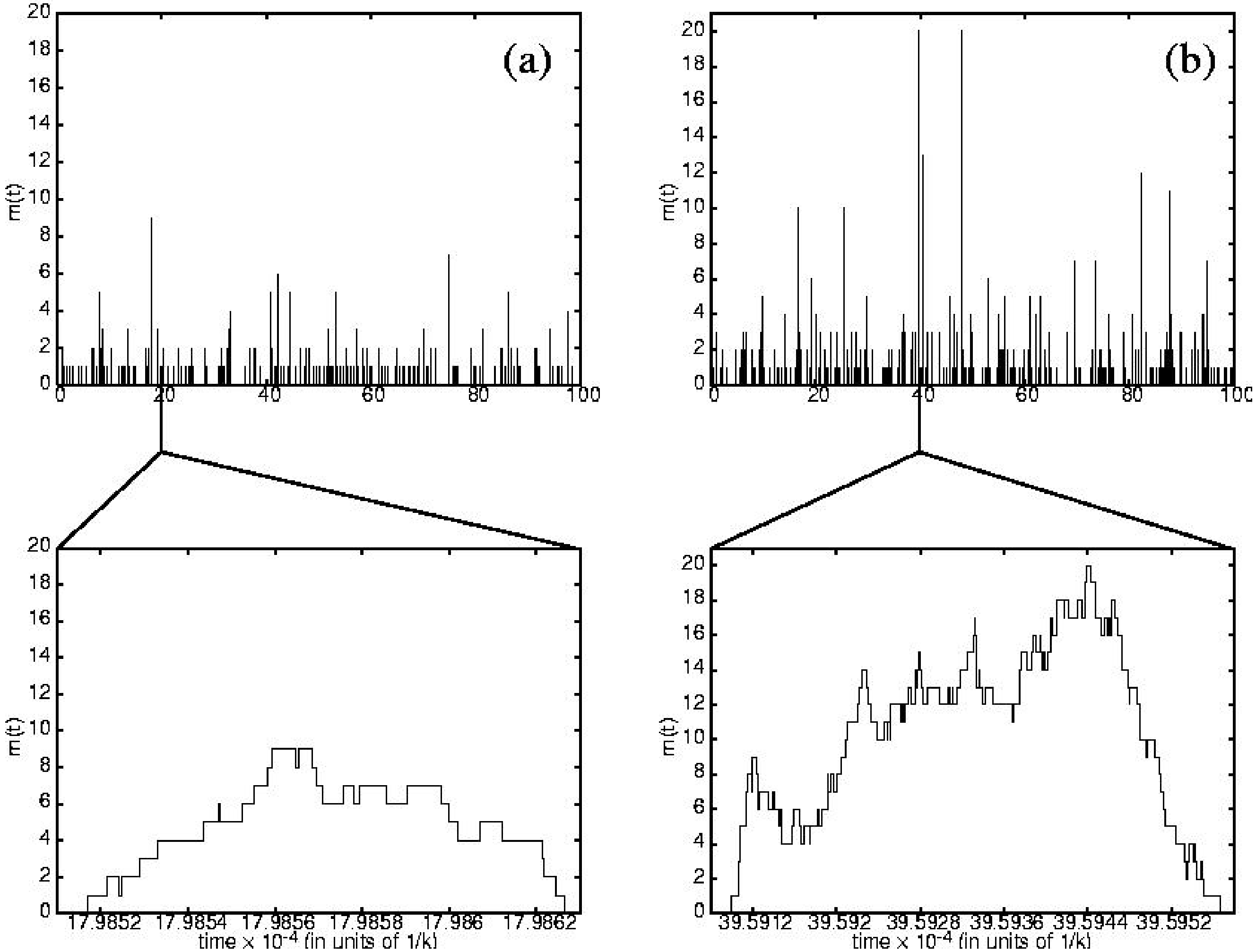
Salt dependence of correlation time (AT9)



T Ambjörnsson, SK Banik, O Krichevsky & RM, Phys Rev Lett (2006); RM & T Ambjörnsson, J Comp Theor Nanosc (2005)

Single bubble time series—Stochastic Gillespie algorithm

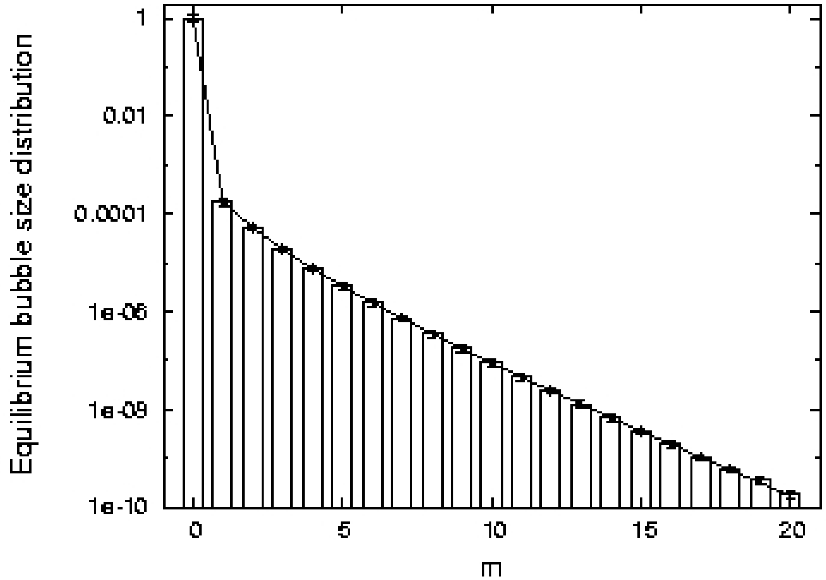
Reaction PDF $P(\tau, \mu; m) = \mathsf{t}^{\mu}(m) \exp\{-(\mathsf{t}^{+}(m) + \mathsf{t}^{-}(m))\,\tau\} \therefore \mu \in \{+,-\}$

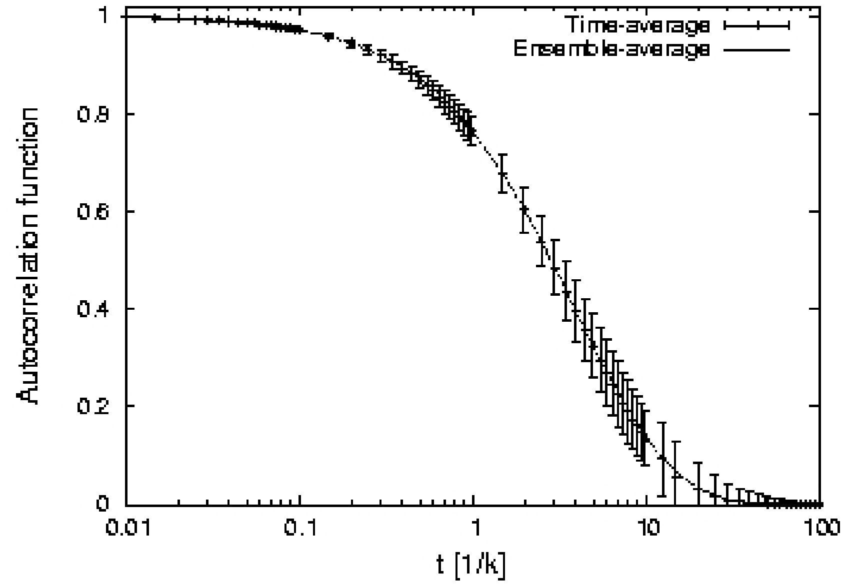


$u = 0.6$ $u = 0.9$

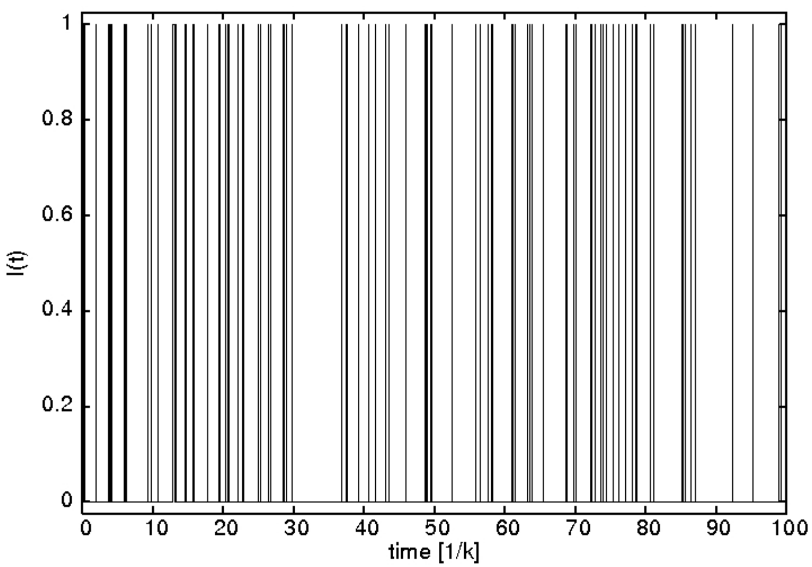
SK Banik, T Ambjörnsson & RM, Europhys Lett (2005)

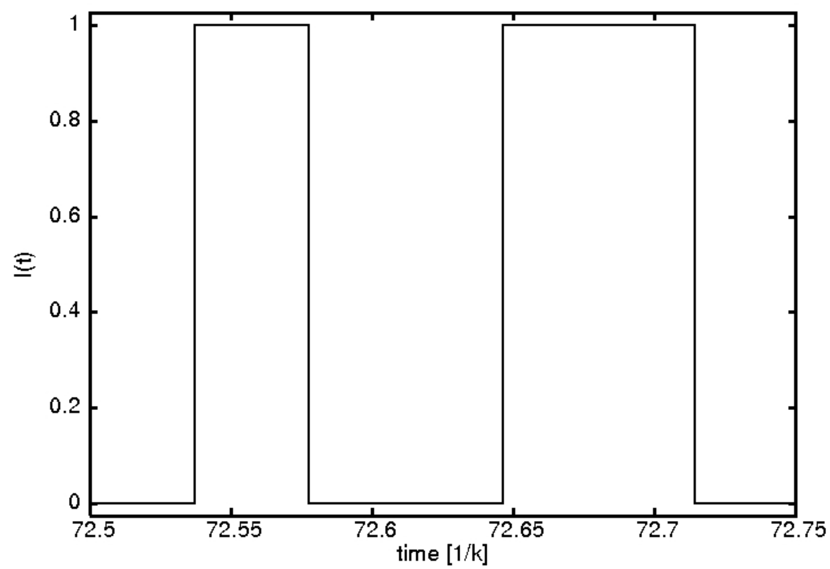
Histogram of equilibrium bubble sizes and autocorrelation:





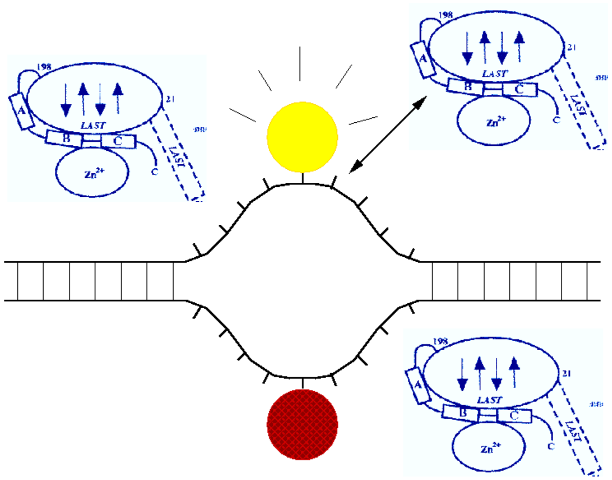
Heteropolymer domain with single tag:





SK Banik, T Ambjörnsson & RM, Europhys Lett (2005)

DNA and single-stranded DNA binding proteins (SSBs):



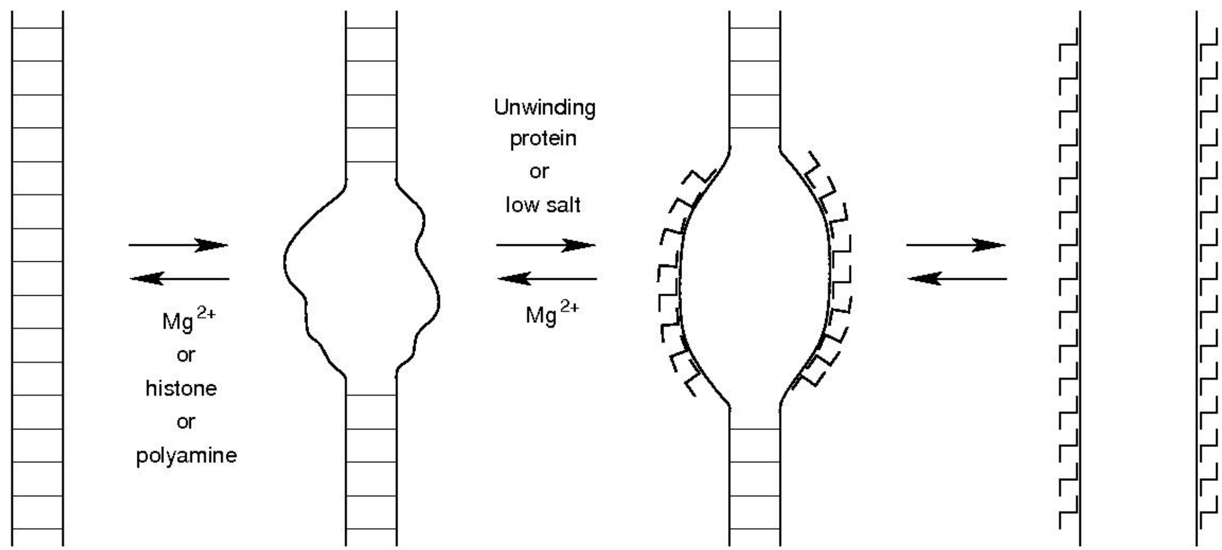
Binding strength $\kappa = c_0 K^{\text{eq}}$

Equilibrium constant K^{eq}

SSB-concentration c_0

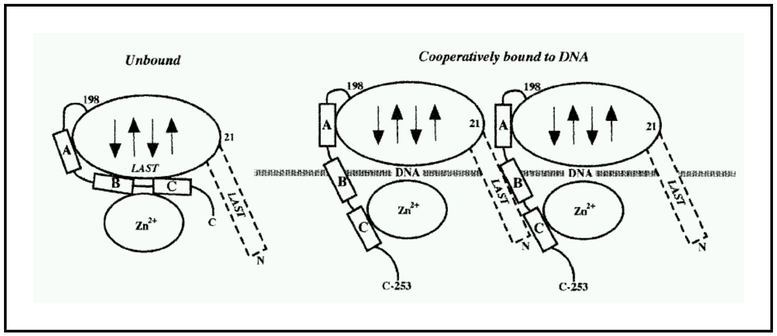
SSB-size λ in units of bp

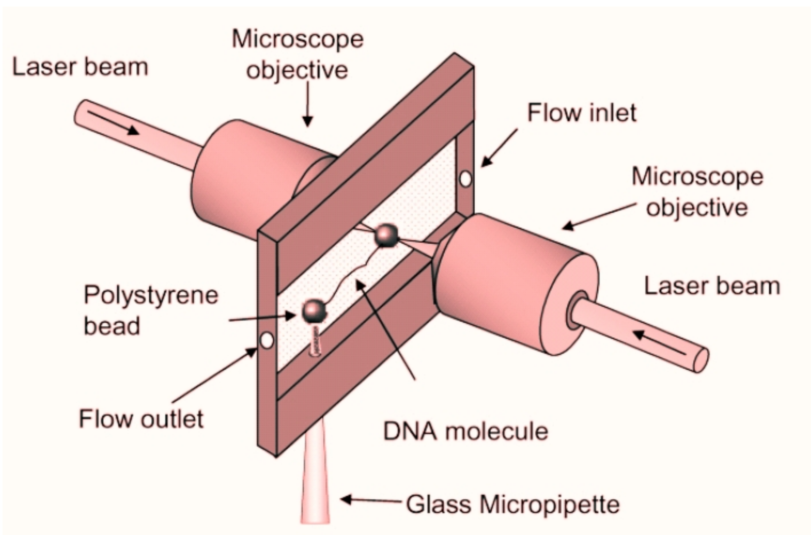
Classical view SSB-induced denaturation:

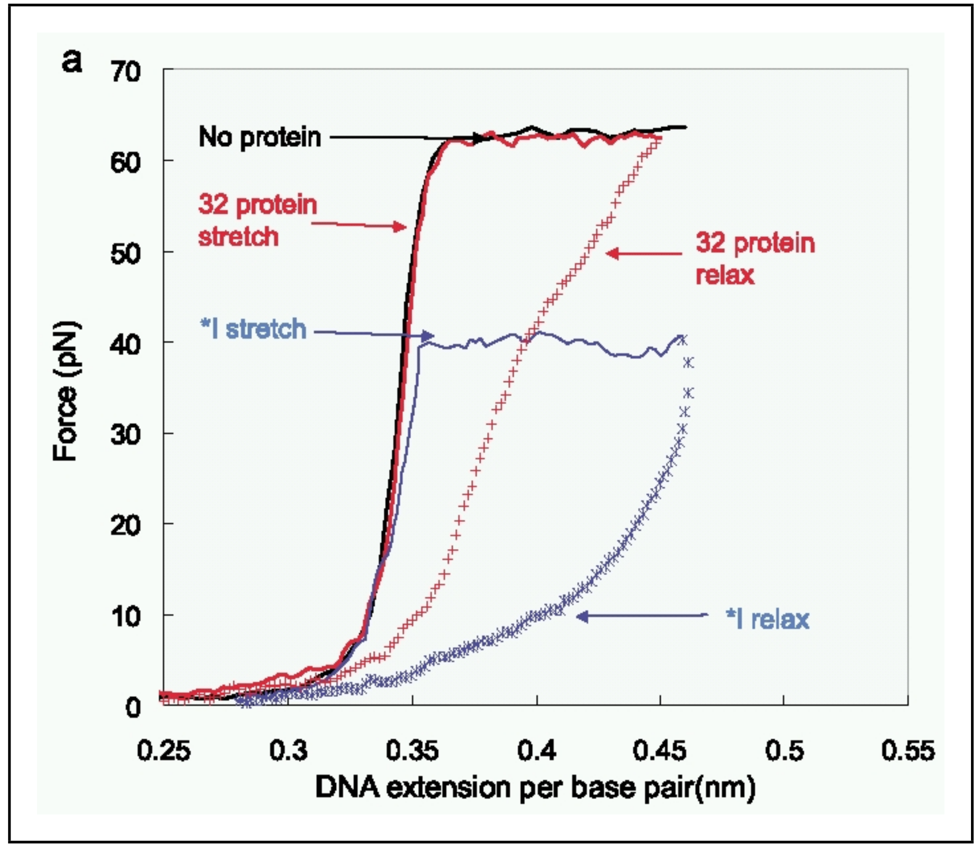


Kornberg, *DNA synthesis* (1974); R Karpel, IUBMB Life (2002)

DNA-overstretching experiments in the presence of bacteriophage T4 g32p SSBs

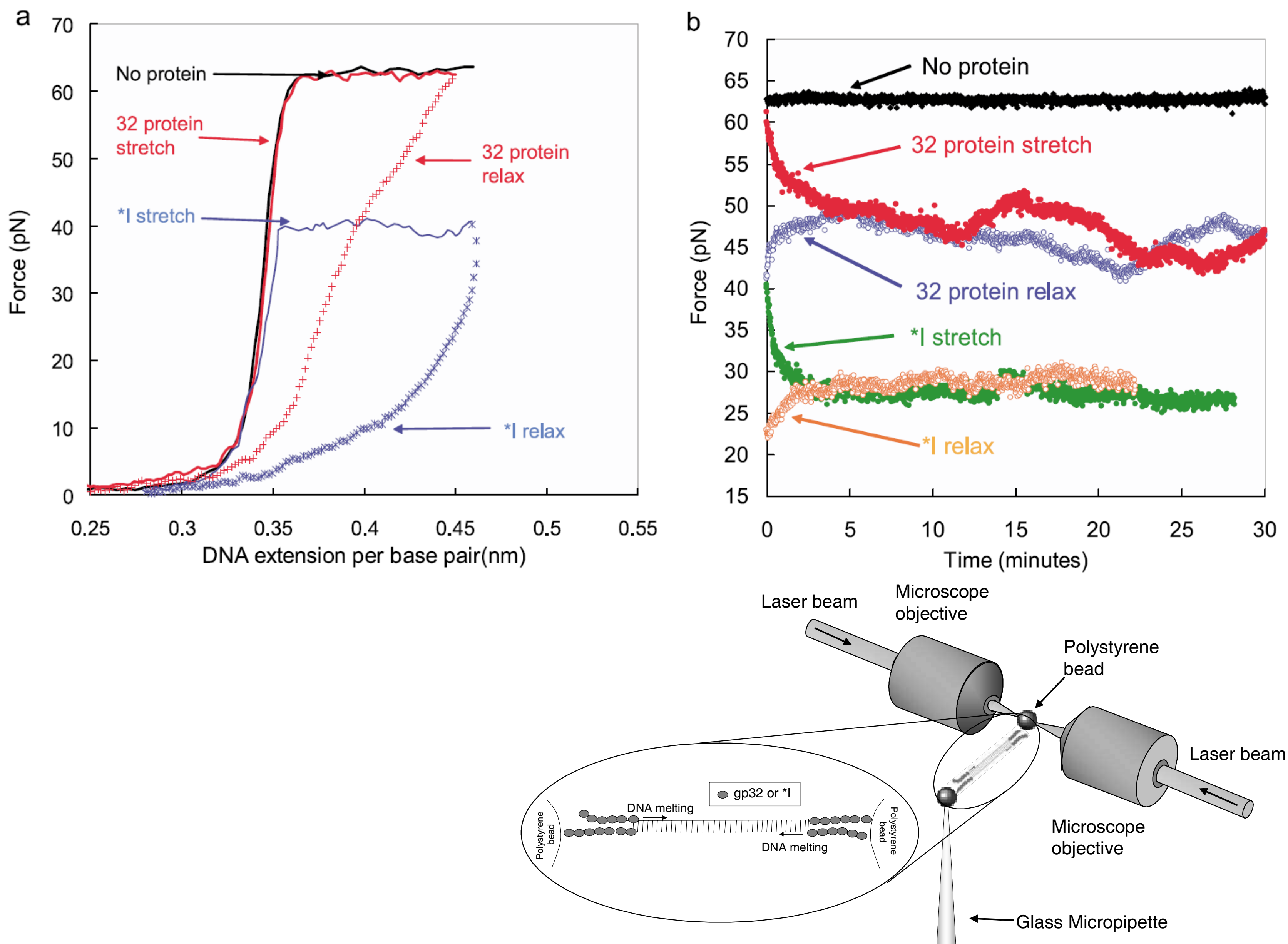






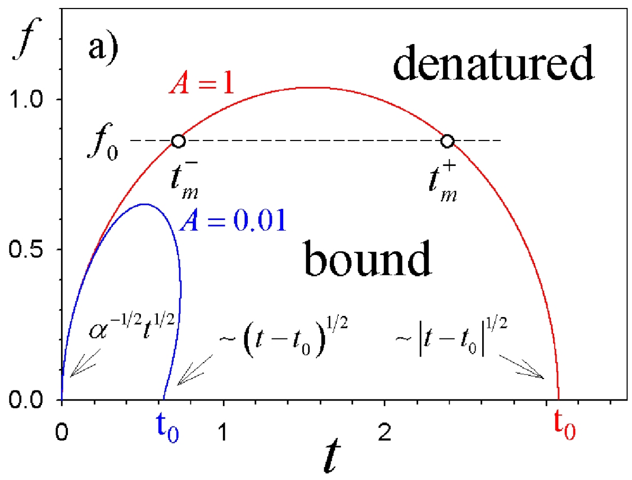
R Karpel, IUBMB Life (2002); K Pant et al, J Mol Biol (2003), (2004)

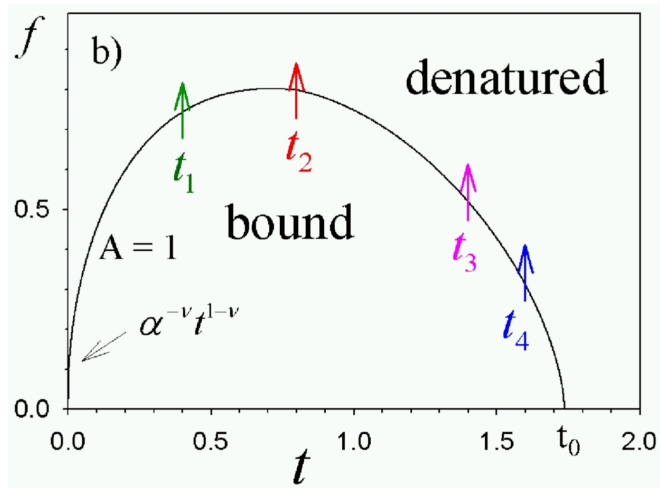
DNA and single-stranded DNA binding proteins (SSBs):

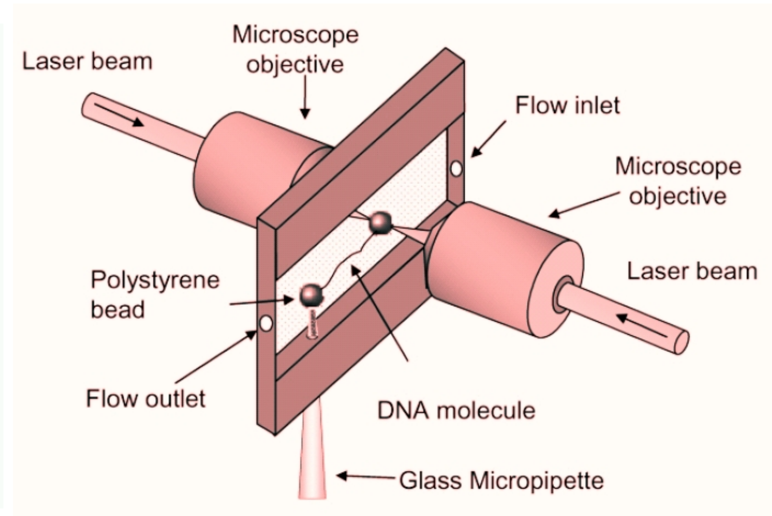


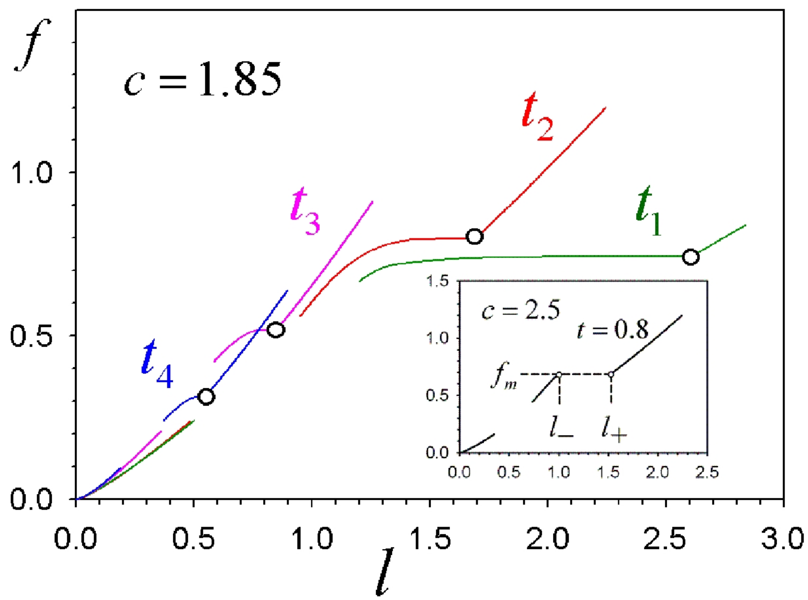
K Pant, RL Karpel, MC Williams, J Mol Biol (2003)

Force-induced melting of DNA









@ $F > 0$ new critical exponent:

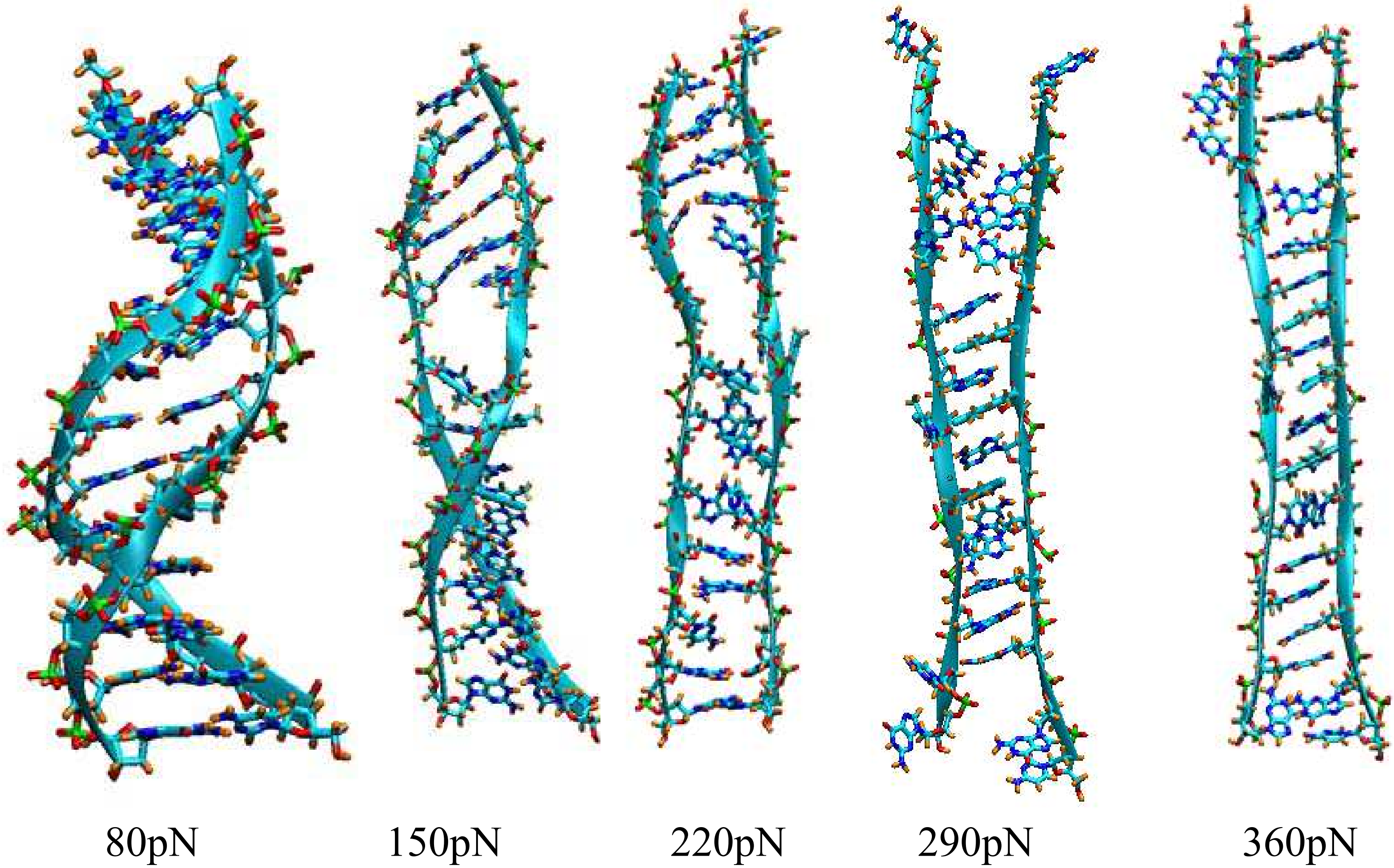
$c = 4\nu - 1/2 \approx 1.85$

$(c = 3\nu \approx 1.76$ @ $F = 0)$

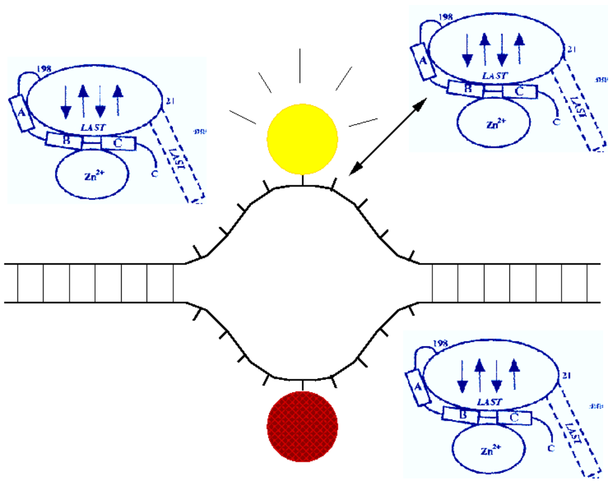
Melting temperature: $T_m = T_m(F)$

A Hanke, M Ochoa & RM, PRL (2008)

S-DNA debate



Breathing bubbles interacting with single-strand binders (SSBs):



Binding strength $\kappa = c_0 K^{\mathrm{eq}}$

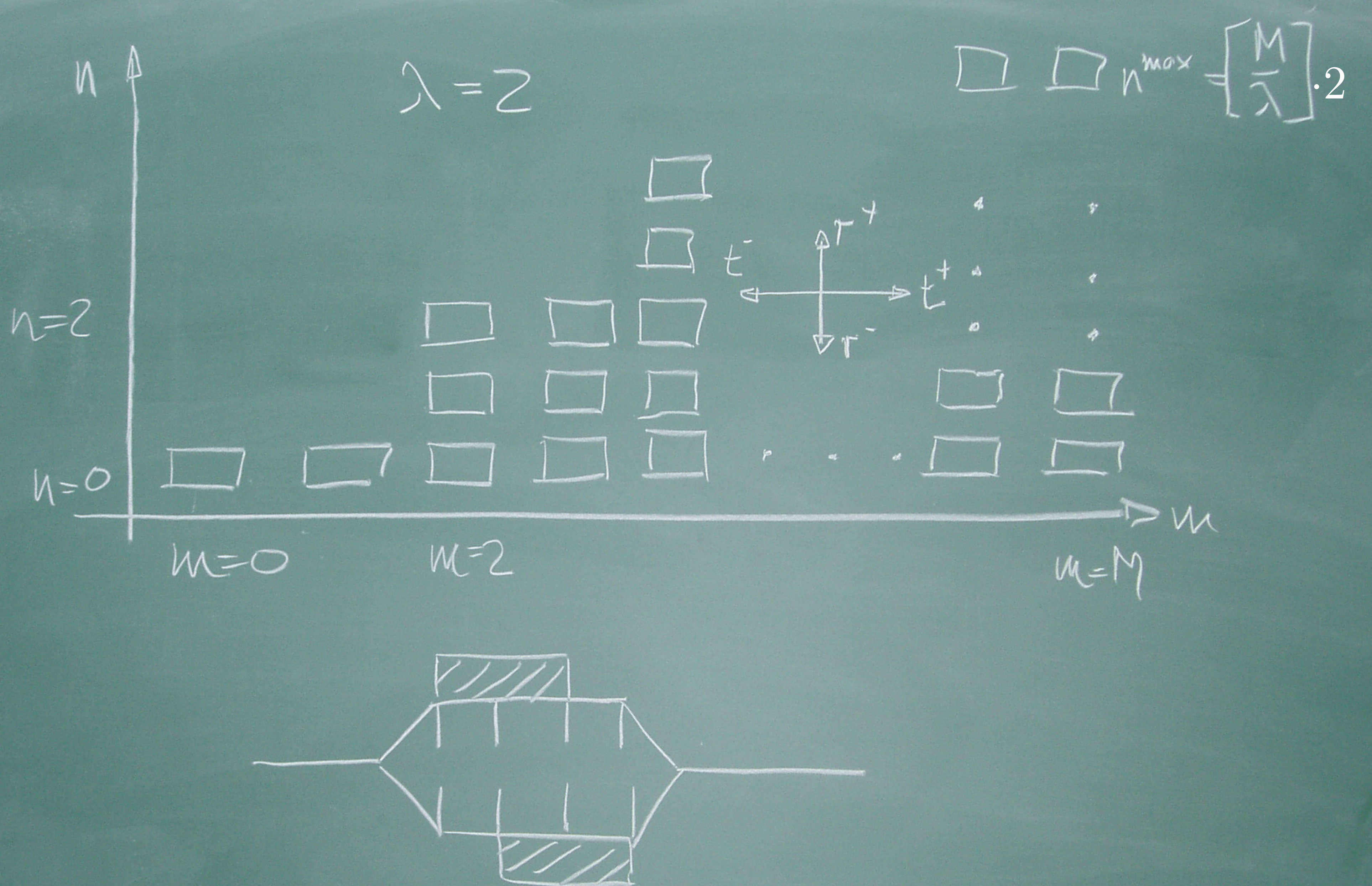
Equilibrium constant K^{eq}

SSB-concentration c_0

SSB-size λ in units of bp

$$
\begin{aligned}
\frac{\partial}{\partial t} P(m,n,t) \quad = \quad & \mathsf{t}^+(m-1,n)P(m-1,n,t) + \mathsf{t}^-(m+1,n)P(m+1,n,t) \\
& -\Big(\mathsf{t}^+(m,n) + \mathsf{t}^-(m,n)\Big)P(m,n,t) \\
& +\mathsf{r}^+(m,n-1)P(m,n-1,t) + \mathsf{r}^-(m,n+1)P(m,n+1,t) \\
& -\Big(\mathsf{r}^+(m,n) + \mathsf{r}^-(m,n)\Big)P(m,n,t)
\end{aligned}
$$

T Ambjörnsson & RM, PRE Rapid Comm (2005)



T Ambjörnsson & RM, PRE Rapid Comm (2005); J Phys Cond Mat (2005a)

Transfer rates fulfil detailed balance:

$$\mathsf{t}^+(m-1,n)\mathscr{Z}(m-1,n) = \mathsf{t}^-(m,n)\mathscr{Z}(m,n)$$
$$\mathsf{r}^+(m,n-1)\mathscr{Z}(m,n-1) = \mathsf{r}^-(m,n)\mathscr{Z}(m,n)$$

Partition function:

$$\mathscr{Z}(m,n) = \mathscr{Z}^{\text{bubble}}(m)\mathscr{Z}^{\text{bind}}(m,n)$$

$$\mathscr{Z}^{\text{bubble}}(m) = \sigma_0 u^m (1+m)^{-c} \ \therefore \ \mathscr{Z}^{\text{bubble}}(0) = 1, \ u = e^{-\beta(E-\theta_0\tau)}$$

$$\mathscr{Z}^{\text{bind}}(m,n) = \Omega^{\text{bind}}(m,n)\kappa^n \ \therefore \ \kappa = c_0 v_0 e^{\beta|E_{\text{bind}}|}$$

$$\Omega^{\text{bind}}(m,n) = \sum_{n'=0}^{n} \omega^{\text{bind}}(m,n')\omega^{\text{bind}}(m,n-n')\Big|_{n'\leq n^{\max}/2}^{n-n'\leq n^{\max}/2}$$

$$\omega^{\text{bind}}(m,n) = \binom{m-(\lambda-1)n}{n}$$

Bubble breathing-SSB binding transfer rates

$$\mathsf{t}^{+}(m,n) = \mathsf{t}^{+}(m) = ku\left(\frac{1+m}{2+m}\right)^{c} \;\therefore\; \mathsf{t}^{+}(0) = 2^{-c}k\sigma_0 u$$

$$\mathsf{t}^{-}(m,n) = k\frac{\Omega^{\mathrm{bind}}(m-1,n)}{\Omega^{\mathrm{bind}}(m,n)} \equiv k \times \Pr\left\{ \begin{array}{c} \text{no SSB} \\ \text{at fork} \end{array} \right\}$$

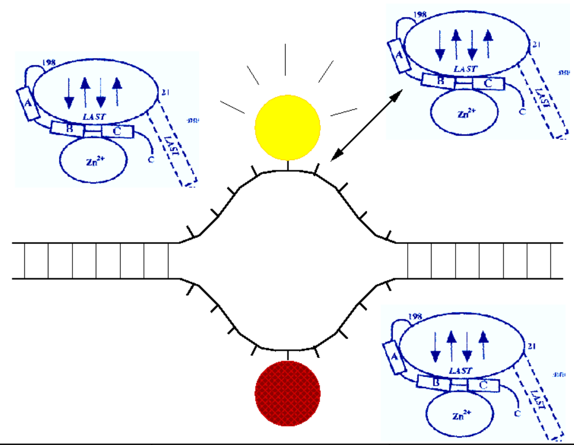
$$\mathsf{r}^{+}(m,n) = \gamma k\kappa(n+1)\frac{\Omega^{\mathrm{bind}}(m,n+1)}{\Omega^{\mathrm{bind}}(m,n)} \equiv \gamma k\kappa \times \Pr\left\{ \begin{array}{c} \exists \text{ slot for} \\ \text{additional SSB} \end{array} \right\}$$

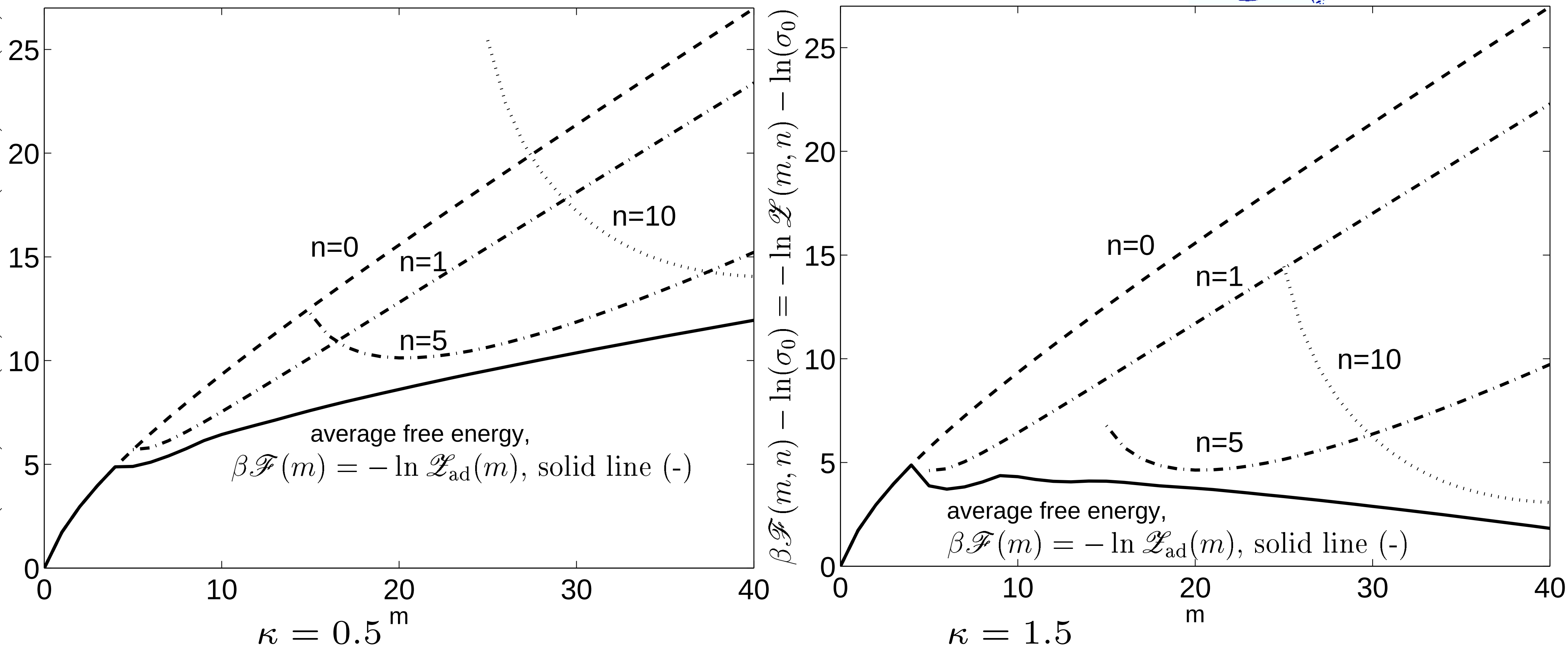
$$\mathsf{r}^{-}(m,n) = n\gamma k$$

$$\gamma \equiv \frac{\{\text{Protein unbinding rate}\}}{\{\text{Bubble zipping rate}\}} = \frac{q}{k}$$

Breathing bubbles and ssDNA binding proteins (SSBs)

$$\frac{\partial}{\partial t}P(m,n,t) = \mathscr{M}(m,n)P(m,n,t)$$



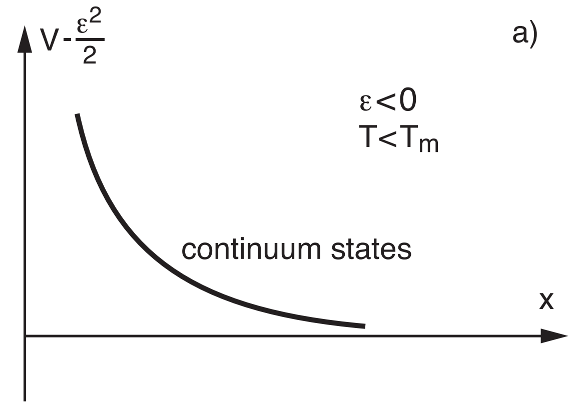


T Ambjörnsson & RM, PRE Rapid Comm (2005)

DNA bubble dynamics as quantum Coulomb problem

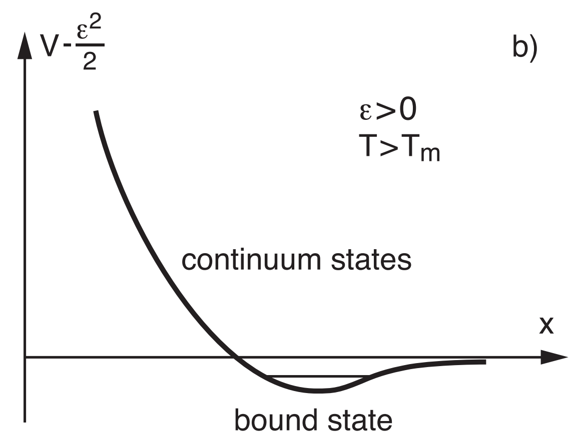
Continuum form of the Poland-Scheraga free energy:

$$\mathscr{F} = \gamma_0 + \gamma_1 \left(1 - \frac{T}{T_m}\right) x + c \ln x$$



Langevin equation for bubble breathing

$$\frac{dx}{dt} = -D\frac{d\mathscr{F}}{dx} + \xi(t), \ \ \langle \xi(t)\xi(t')\rangle = 2Dk_B T\delta(t-t')$$



Fokker-Planck equation ($\mu = c/2k_B T$):

$$\frac{\partial P}{\partial t} = \frac{\partial}{\partial x}\left(\frac{\mu}{x} - \underbrace{\frac{\gamma_1}{2k_B T}\left[\frac{T}{T_m} - 1\right]}_{\epsilon}\right) P + \frac{1}{2}\frac{\partial^2 P}{\partial x^2}$$

With $P = e^{\epsilon x} x^{-\mu} \tilde{P}$, obtain imaginary time Schrödinger Eq:

$$-\frac{\partial \tilde{P}}{\partial t} = -\frac{1}{2}\frac{\partial^2 \tilde{P}}{\partial x^2} + \left(\frac{\mu(\mu+1)}{2x^2} - \frac{\mu\epsilon}{x} + \frac{\epsilon^2}{2}\right)\tilde{P}$$

HC Fogedby & RM, Phys Rev Lett (2007), Phys Rev E (2007)

Bubble lifetime distribution:

(i) $T < T_m$: asymptotically,

$$\wp(t) \simeq x_0^{1+c} e^{|\epsilon| x_0} e^{-\epsilon^2 t/2} t^{-3/2-c/2}$$

$$\mathcal{T} = \int_0^\infty t\wp(t)dt \simeq \frac{x_0}{|\epsilon|}\frac{K_{(c-1)/2}(x_0|\epsilon|)}{K_{(c+1)/2}(x_0|\epsilon|)}$$

(ii) $T = T_m$: exact,

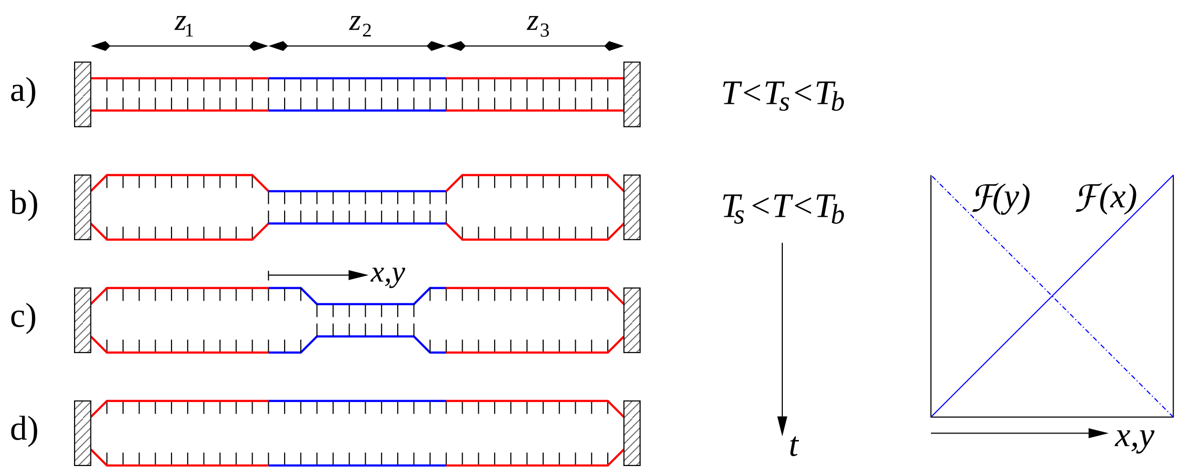
$$\wp(t) = \frac{2x_0^{1+c}}{\Gamma(1/2 + c/2)} e^{-x_0^2/2t}(2t)^{-3/2-c/2}$$

$$\mathcal{T} = \frac{x_0^2}{c-1}, \quad \forall c > 1$$

(iii) $T > T_m$: denaturation, bubble coalescence, grand ensemble

HC Fogedby & RM, Phys Rev Lett (2007), Phys Rev E (2007)

Bubble coalescence in breathing DNA

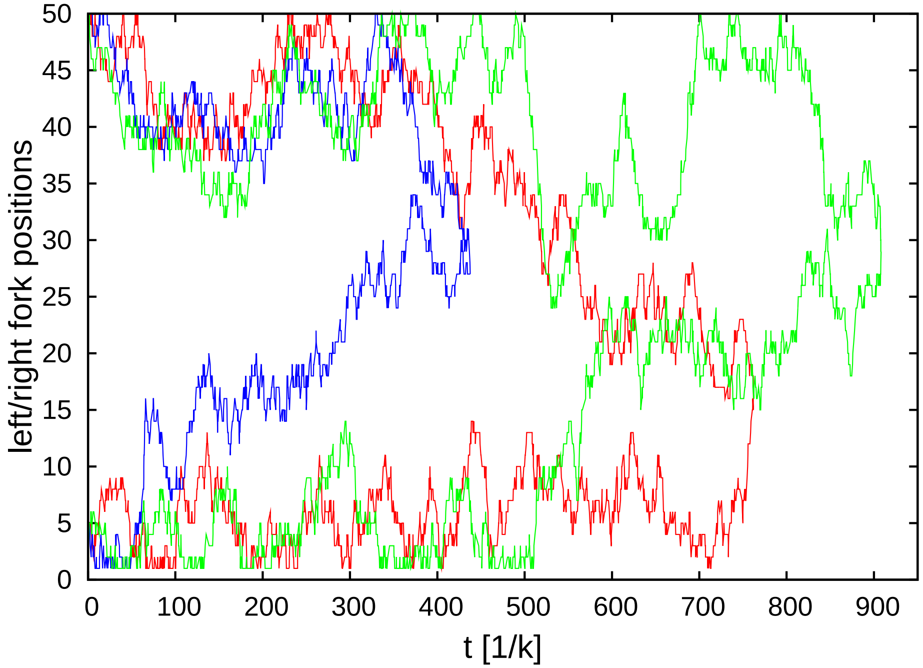


The problem of two viscious walkers in opposite potentials:

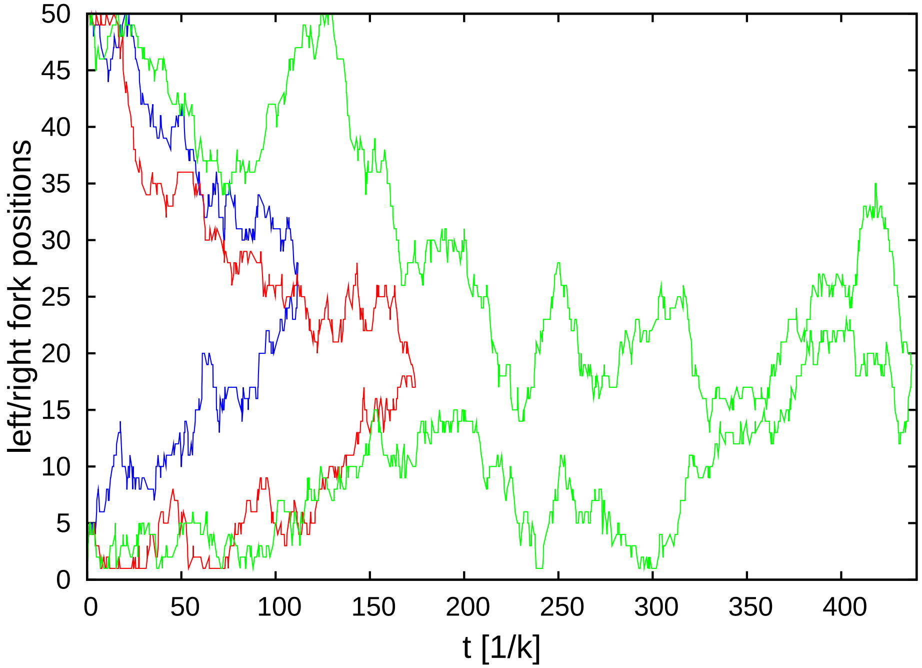
$$\frac{\partial}{\partial t}P(x,y,t) = \left(D\left[\frac{\partial^2}{\partial x^2} + \frac{\partial^2}{\partial y^2}\right] - F\frac{\partial}{\partial x} + F\frac{\partial}{\partial y}\right) P(x,y,t)$$

T Novotný, JN Pedersen, MS Hansen, T Ambjörnsson & RM, Europhys Lett (2007); E-print cond-mat/0610752

Zipping fork trajectories



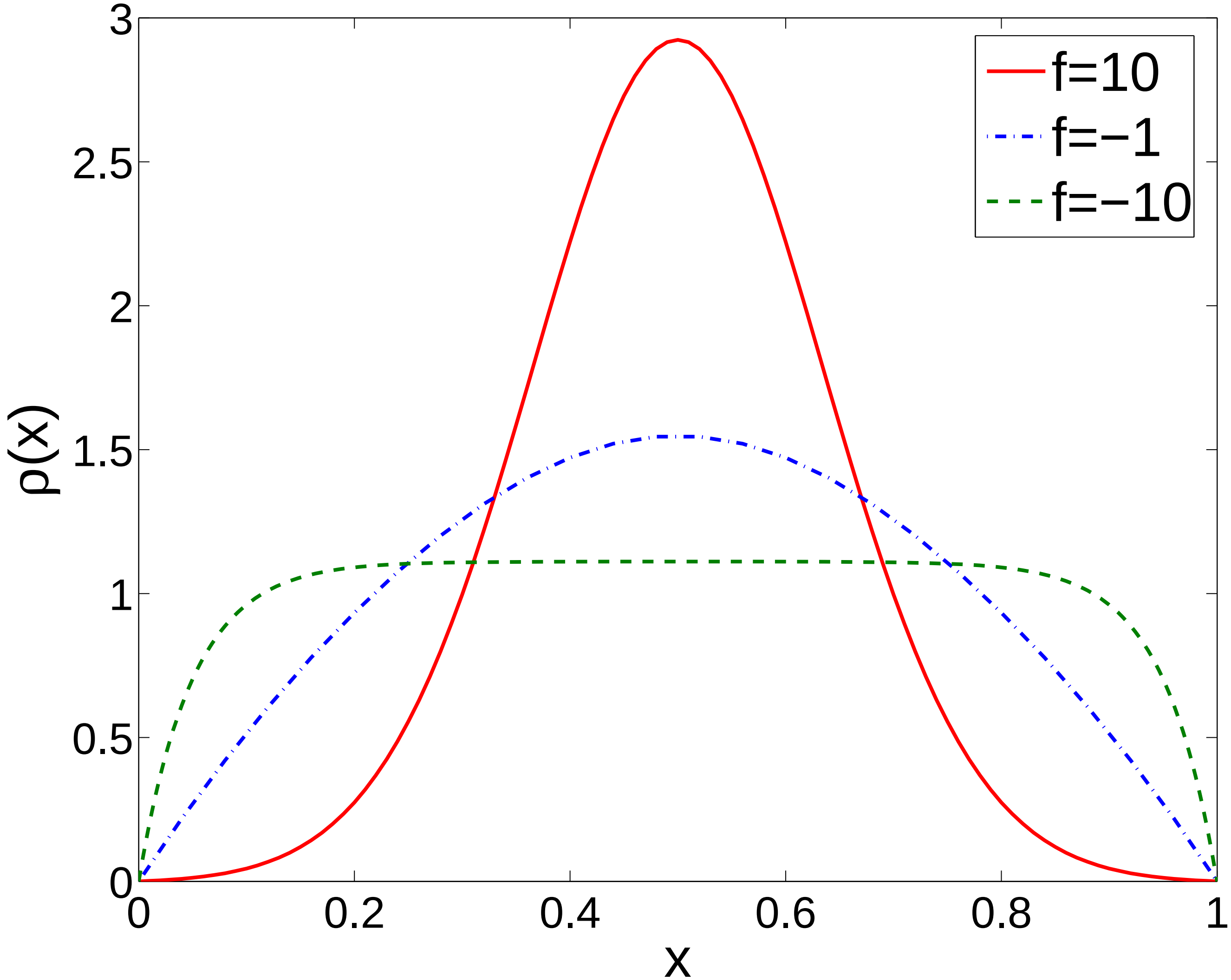
$u = 0.98$



$u = 1.10$

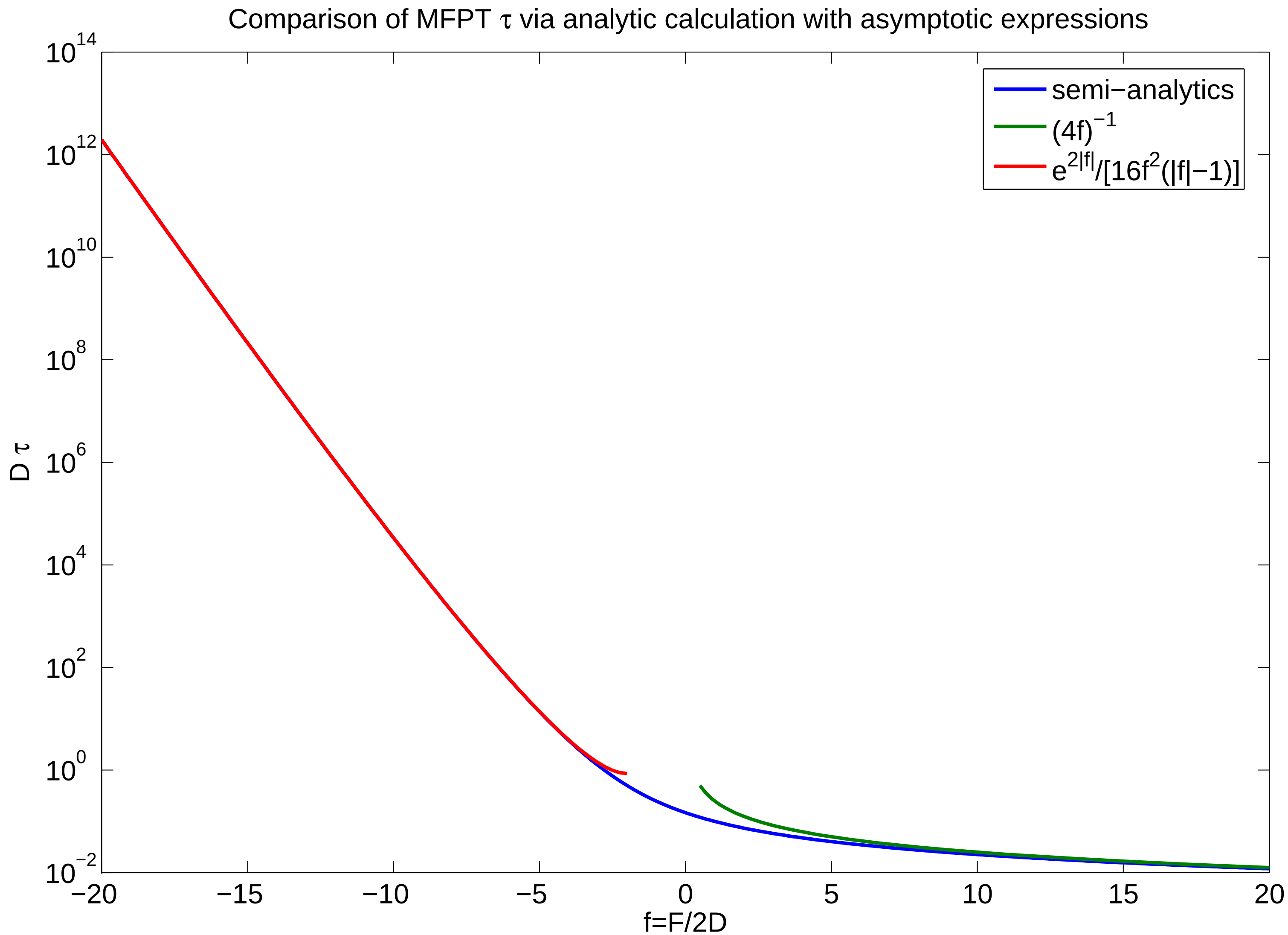
Meeting position:

$f \ll -1$: Pr to be @ x is $\propto \exp(-\beta\phi(x))$ with free energy $\phi(x) = -\int^x f(x')dx' \Longrightarrow$ joint Pr @ x: $\exp(-\beta[\phi_L(x) + \phi_R(x)]) \approx \text{const}$



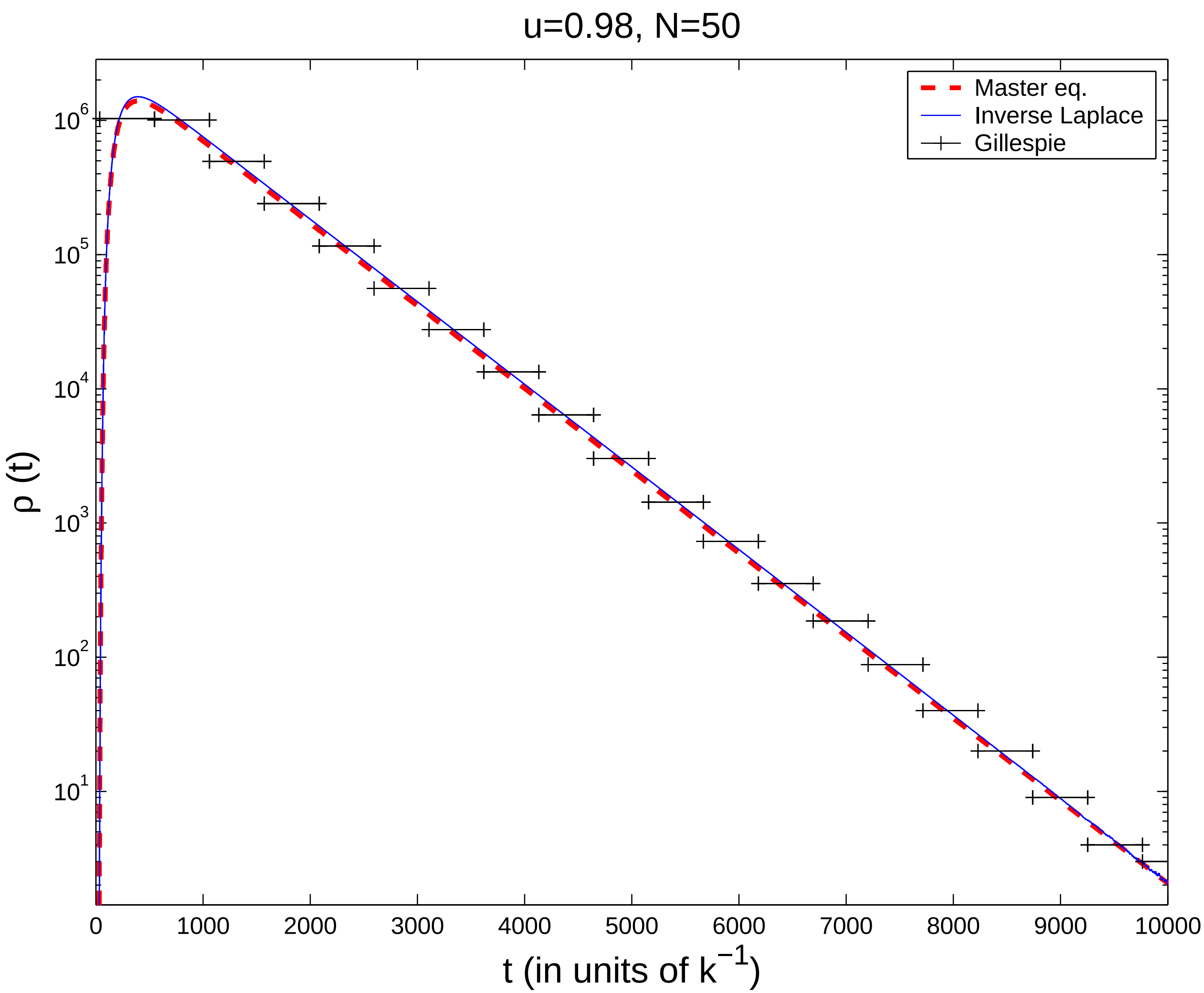
T Novotný, JN Pedersen, MS Hansen, T Ambjörnsson & RM, Europhys Lett (2007); E-print cond-mat/0610752

Mean coalescence time



T Novotný, JN Pedersen, MS Hansen, T Ambjörnsson & RM, Europhys Lett (2007); E-print cond-mat/0610752

Coalescence time density, comparison of methods



T Novotný, JN Pedersen, MS Hansen, T Ambjörnsson & RM, Europhys Lett (2007); E-print cond-mat/0610752