Denaturation of circular DNA

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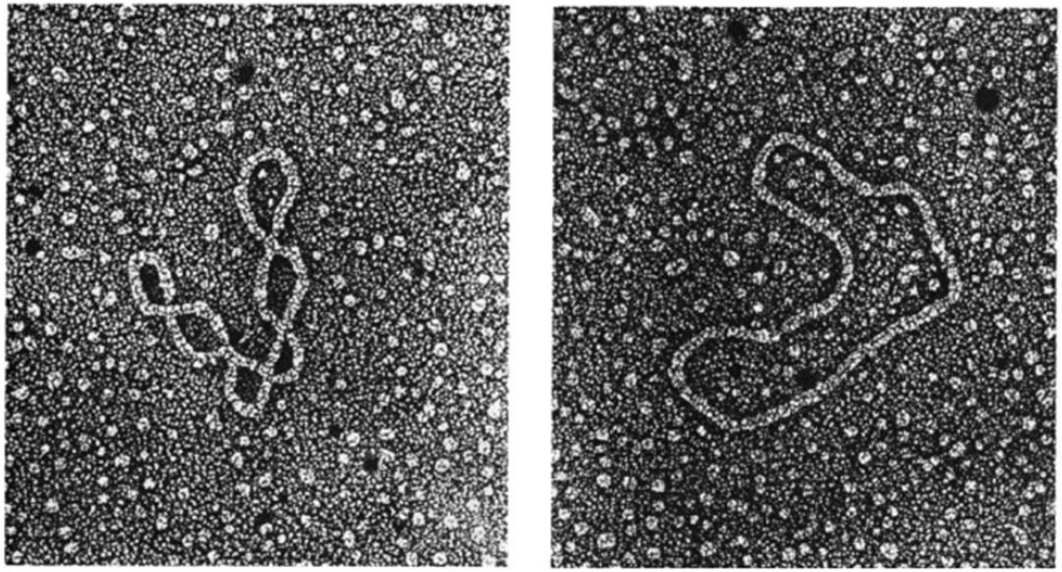

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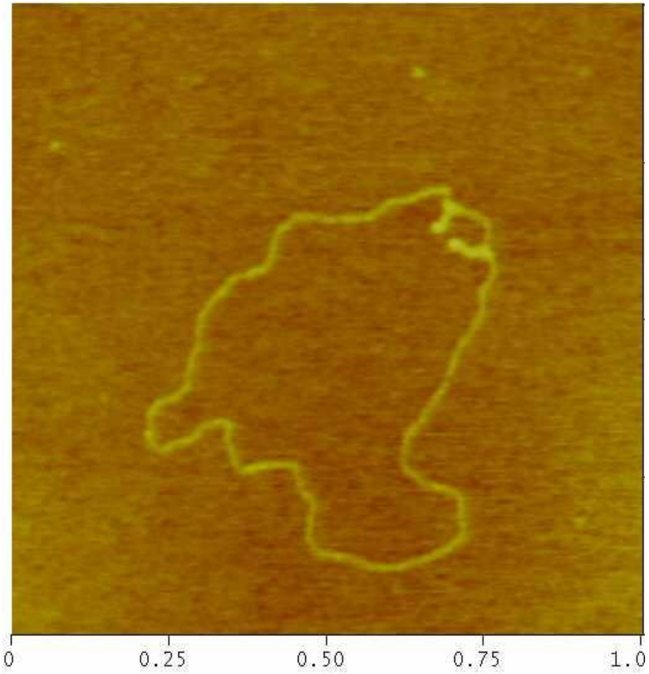
Form II



STM and AFM experiments

Outline

- Summary of the main results
- DNA denaturation
- The Poland-Scheraga model linear DNA
- Linear DNA – canonical vs grand canonical approach
- Circular DNA

Summary of the main results

The nature of the melting transition is determined by the loop entropy parameter c

$$\Omega(l) \sim \frac{s^l}{l^c}$$

in DNA $c \approx 2.1$

linear DNA

$c \leq 1$ no transition

$1 < c \leq 2$ continuous transition

$c > 2$ first order transition

Circular DNA

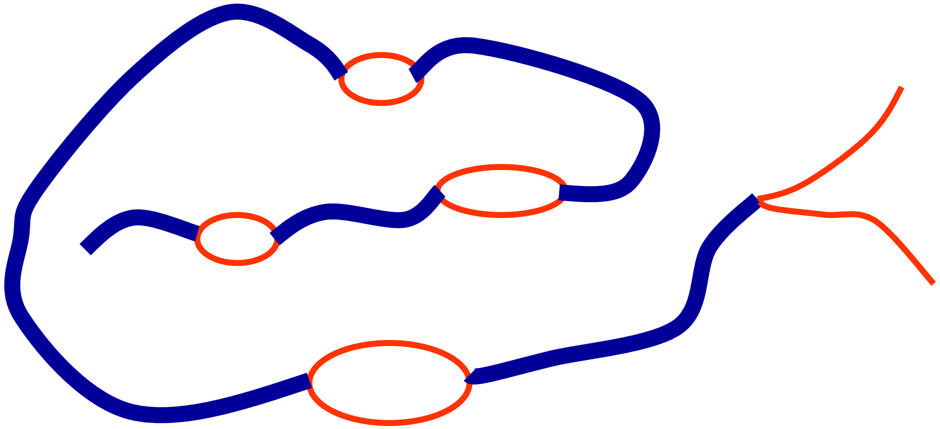
$c \leq 2$ no transition

$2 < c \leq 3$ transition of high order $\left\lceil \frac{c-1}{c-2} \right\rceil$

$c > 3$ second order transition

DNA denaturation

fluctuating DNA



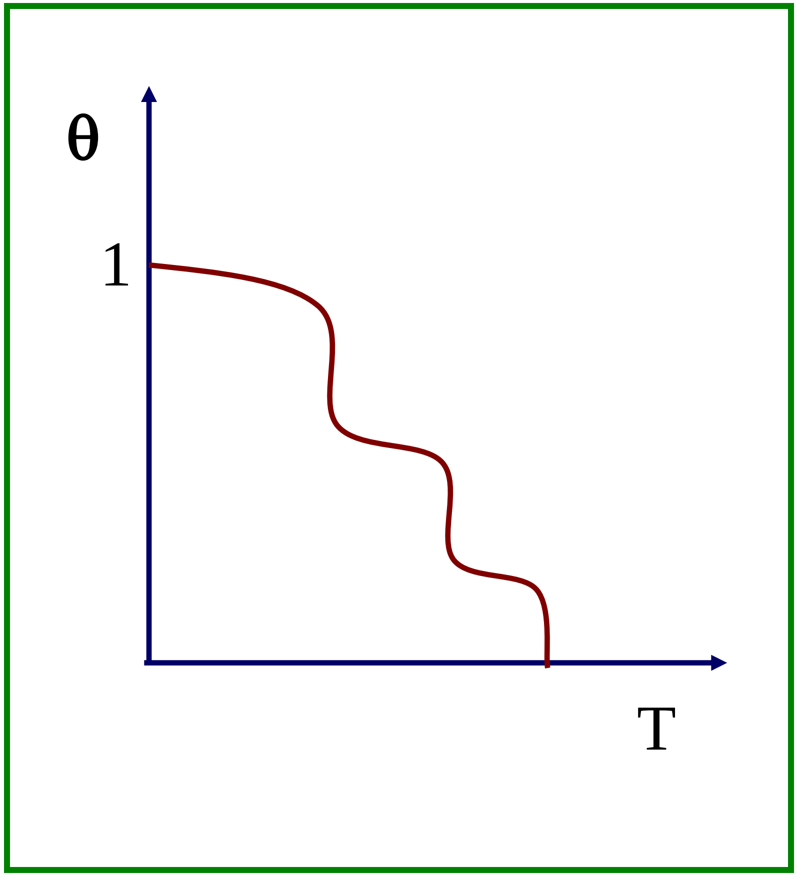
Persistence length l_p

double strands $l_p \sim$ 100-200 bp

Single strands $l_p \sim$ 10 bp

Schematic melting curve

θ = fraction of bound pairs



Melting curve is measured directly by optical means

absorption of uv line 268nm

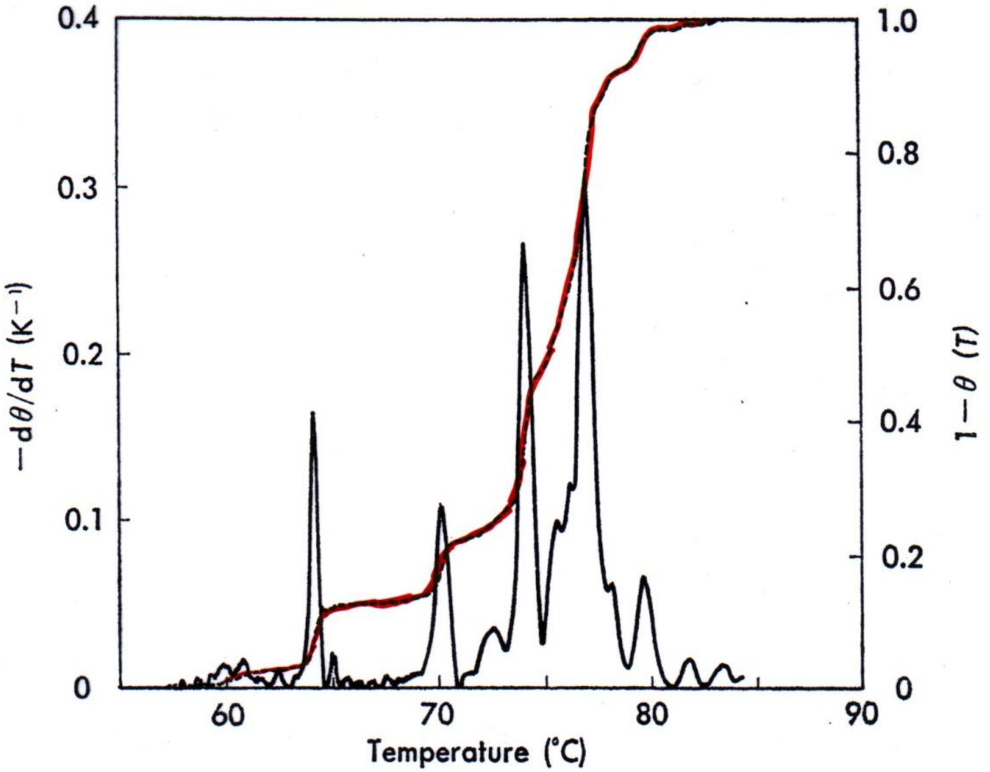


Fig. 1. Melting profile of pNT1 DNA in 0.1×SSC. Closed circular DNA of plasmid pNT1 was linearized by cutting it at the unique *Eco* RI site. Melting profile was measured as described previously (*5*, *9*). Details will be reported elsewhere (*21*). Dotted line: cumulative profile. Solid line: differential profile.

Linearized
Plasmid pNT1
3.83 Kbp

O. Gotoh, Adv. Biophys. 16, 1 (1983)

Melting curve of yeast DNA 12 Mbp long

Bizzaro et al, Mat. Res. Soc. Proc. 489, 73 (1998)

Linearized Plasmid pNT1 3.83 Kbp

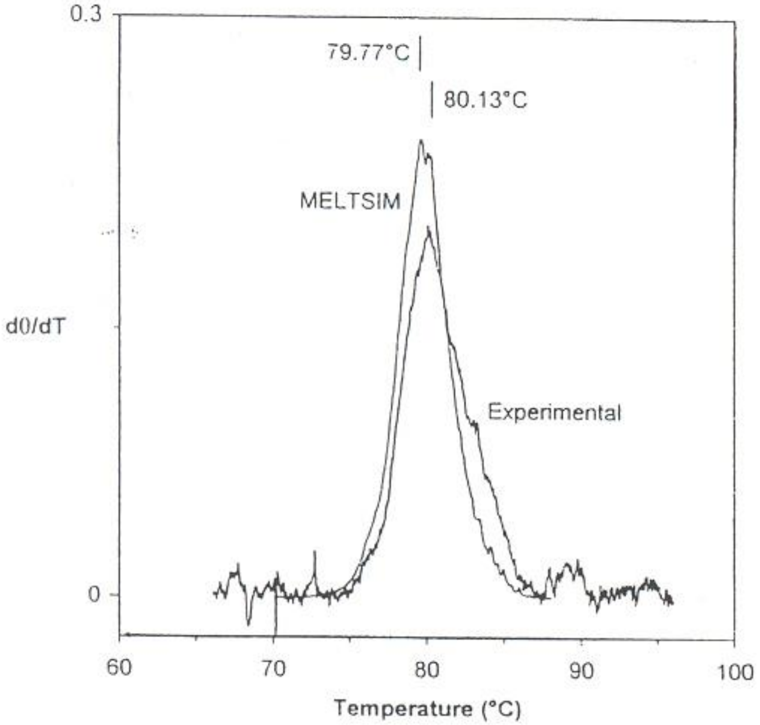
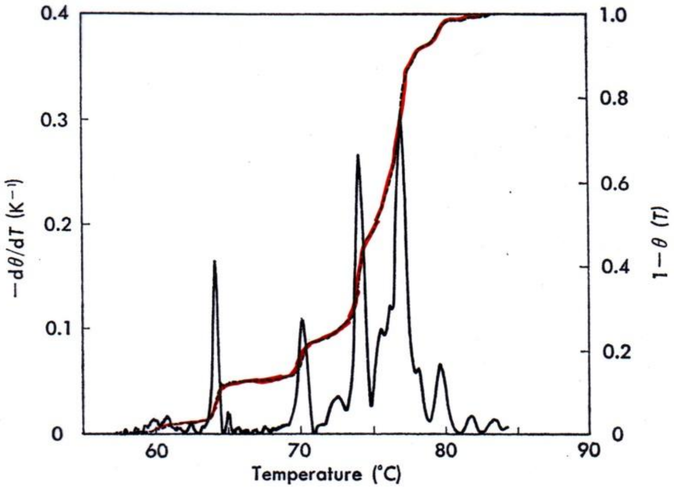


Figure 1: Experimental and MELTSIM Derivative Melts of Total Yeast DNA. Vertical Lines Indicate Tm's Calculated from Gaussian Fits.

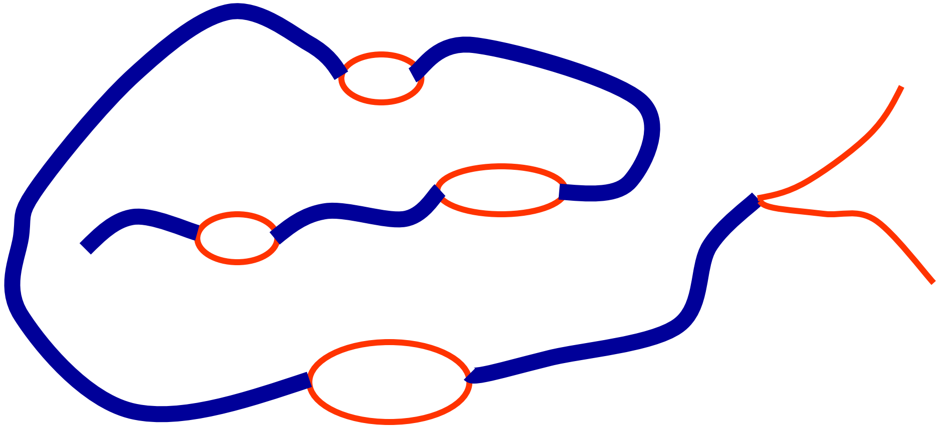


One is interested in features like

- Loop size distribution $P(l)$
- Order of the denaturation transition
- Inter-strand distance distribution $P(r)$
- Effect of heterogeneity of the chain
- Loop dynamics

Theoretical Approach

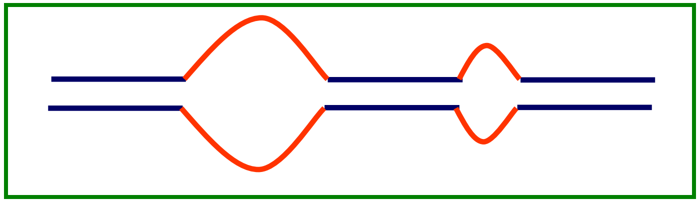
fluctuating microscopic configurations



approaches:

- Poland-Scheraga
- Peyrard-Bishop

Poland-Scheraga model



Bound segment:

- Energy ε per bond (complementary bp)

Loops:

- Degeneracy

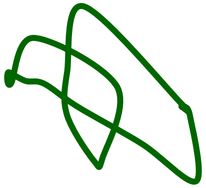
$$\boxed{\Omega(l) \propto \frac{s^l}{l^c}}$$

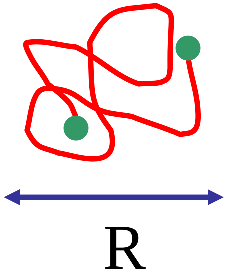
s - geometrical factor

c=universal exponent

$$\Omega(l) \propto \frac{s^l}{l^c}$$

random loop: $c=d/2$ $c= 3/2$ $(d=3)$





$$R \sim \sqrt{l}$$

$$V = R^d$$

Poland, Scheraga (1966)

$$\Omega(l) \propto \frac{s^{l}}{l^{c}}$$

self-avoiding loop c=dν c=1.76 (d=3)

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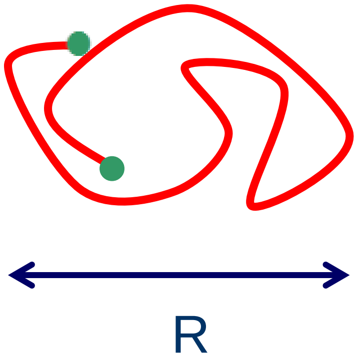


$$R \sim l^{\nu}$$

$$V = R^{d}$$

$$c = d\,\nu$$

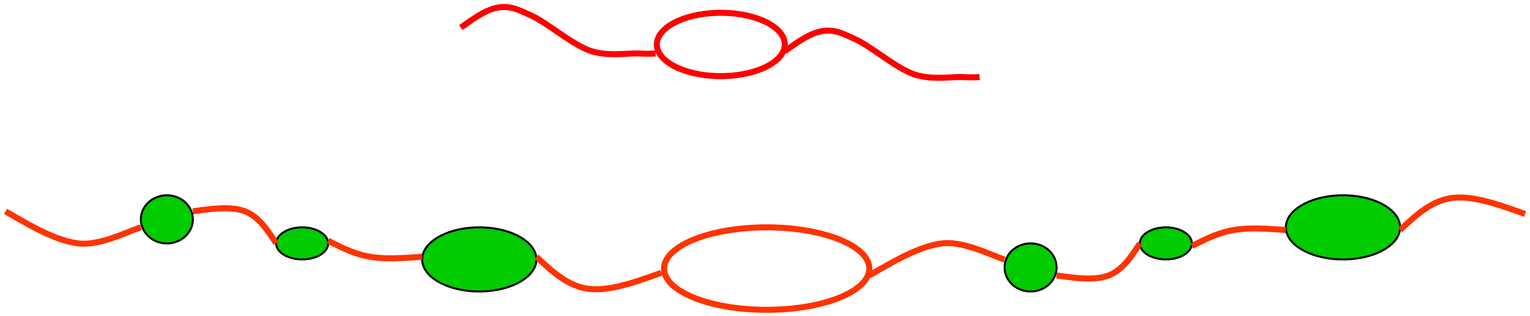
Fisher (1966)

$$\Omega(l) \propto \frac{s^l}{l^c}$$

Self avoiding loop embedded in a chain:

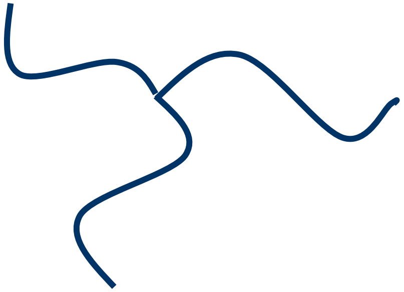
$$c = d\nu - 2\sigma_3$$

$c=2.11$ ($d=3$)



Kafri, Mukamel, Peliti, (2000)

$$\Omega(L) \sim s^L L^{2\sigma_1}$$



$$\Omega(L) \sim s^L L^{3\sigma_1+\sigma_3}$$

$$\Omega(L) \sim s^L L^{-d\nu}$$

the main results of the PS model

$c \leq 1$	no transition
$1 < c \leq 2$	continuous transition
$c > 2$	first order transition

for $1 < c \leq 2$ $\quad \beta = \frac{2-c}{c-1}$

$m_b \sim t^{\beta}$ where m_b is the fraction of bound pairs and $t = \frac{T_M - T}{T_M}$

Loop-size distribution

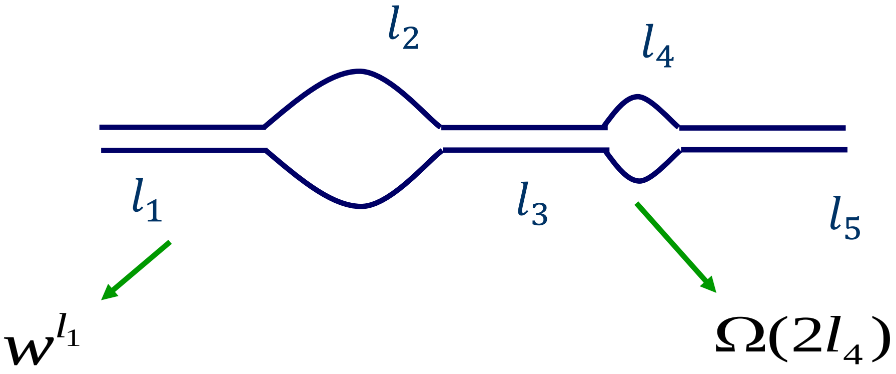
$$P(l) \propto \frac{e^{-l/\xi}}{l^c}$$

$$\xi \propto \frac{1}{(T_M - T)^{\frac{1}{c-1}}} \qquad 1 < c \le 2$$

$$\xi \propto \frac{1}{T_M - T} \qquad c > 2$$

PS model – grand canonical approach

typical configuration



$$\boxed{w^{l_1}\ \Omega(2l_2)\ w^{l_3}\ \Omega(2l_4)\ w^{l_5}\dots}$$

$$\boxed{w = e^{\beta E} \qquad \Omega(l) = \frac{s^l}{l^c}}$$

$$\boxed{\begin{gathered} Z(L) = \sum_k \sum_{l_1} \sum_{l_2} \cdots \sum_{l_{2k+1}} \\ l_1 + \dots + l_{2k+1} = L \end{gathered}}$$

Grand partition sum

$$Q(z)=\sum_{l=1}^{\infty} Z(L) z^{l}$$

z - fugacity

$$L=\frac{\partial \ln Q(z)}{\partial \ln z}$$

$$\boxed{Q(z)=\frac{1}{1-V(\omega z)U(sz)}}$$

$$V(\omega z)=\sum_{l=1}^{\infty} w^{l} z^{l}$$

GPS of a segment

$$w=e^{-\beta E}$$

$$U(sz)=\sum_{l=1}^{\infty} \frac{s^{l}}{l^{c}} z^{l}$$

GPS of a loop

$$Q(z) = \frac{1}{1 - V(\omega z)U(sz)}$$

$$L = \frac{\partial \ln Q(z)}{\partial \ln z}$$

$$L \to \infty \longrightarrow V(\omega z)U(sz) = 1$$

solve for the fugacity $z(\omega)$

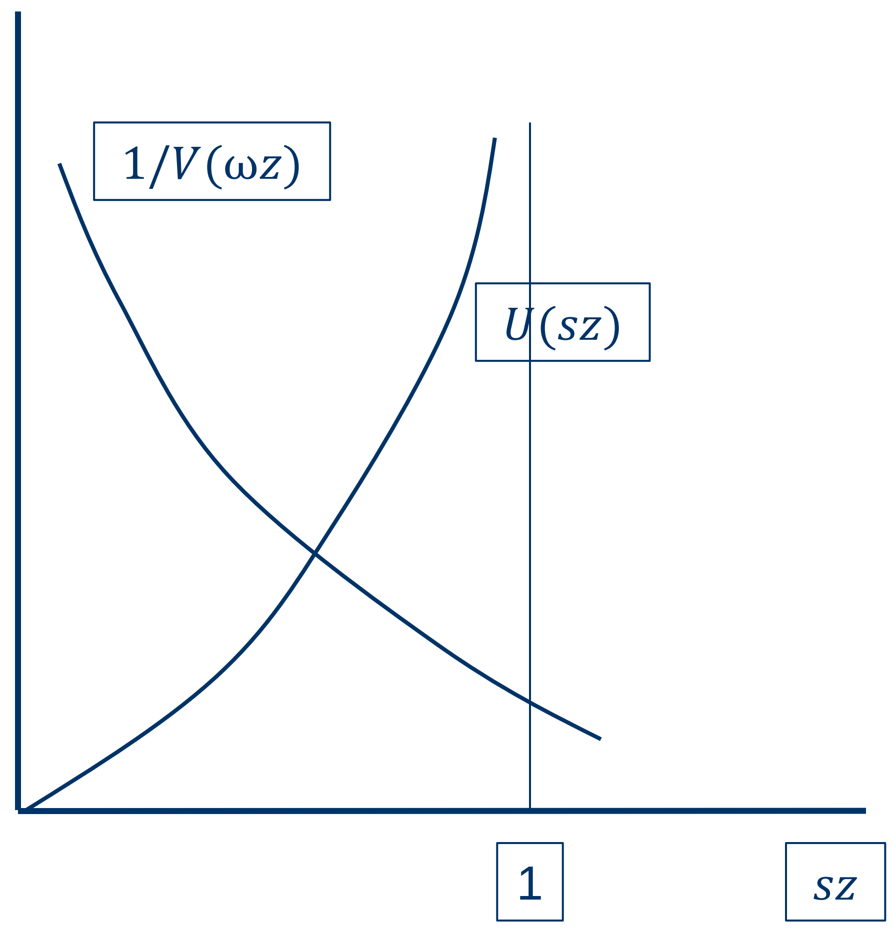
Thermodynamic potential $z(\omega)$

$$w = e^{\beta E}$$

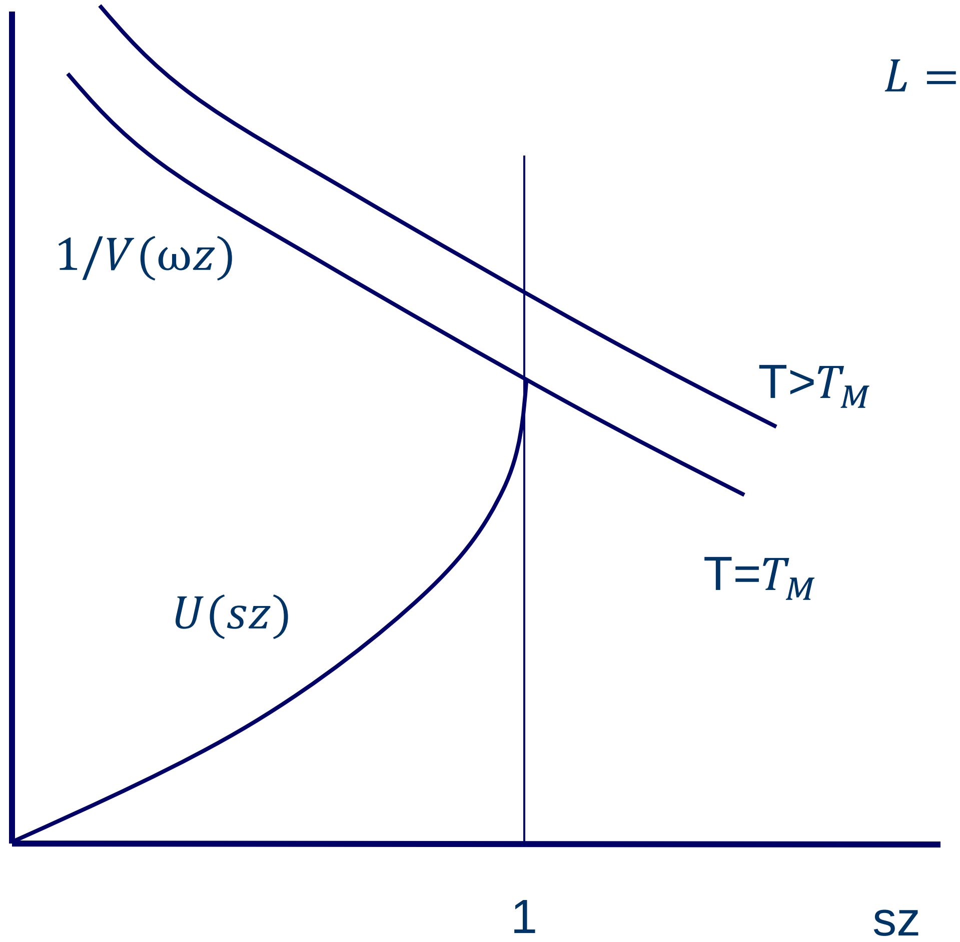
Order parameter

$$\theta = \frac{\partial \ln z}{\partial \ln w}$$

for $T < T_M$



$1 < c \leq 2$



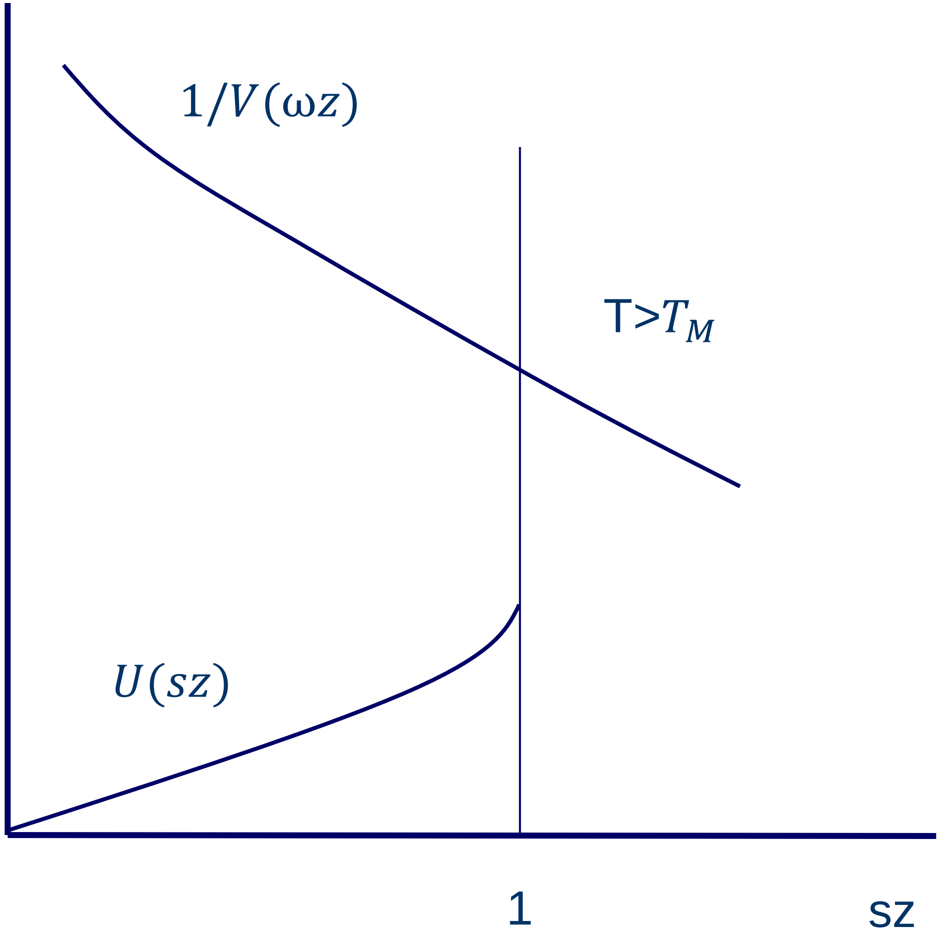
$$L = \frac{\partial \ln Q}{\partial \ln z} = z\frac{U'(sz)V(\omega z)+U(sz)V'(\omega z)}{1-U(sz)V(\omega z)}$$

$$U'(sz) \xrightarrow{sz\to 1} \infty$$

$$U(sz) = \Phi_c(sz) = \sum \frac{(sz)^n}{n^c}$$

C>2

$$L = \frac{\partial \ln Q}{\partial \ln z} = \mathrm{z}\,\frac{U'(sz)V(\omega z) + U(sz)V'(\omega z)}{1 - U(sz)V(\omega z)}$$



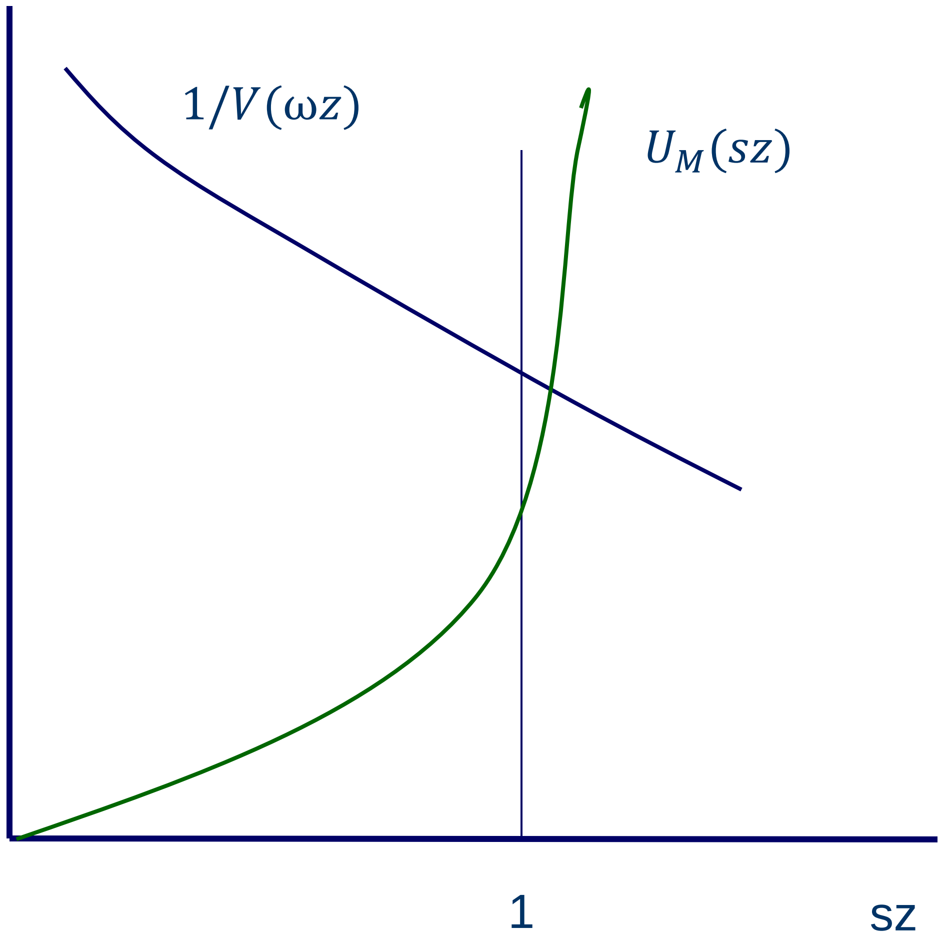
$$U'(sz) \xrightarrow{\mathrm{sz}\to 1} const < \infty$$

How does one solve for $L \to \infty$?

One needs to regularize Φ_c(sz) by inserting a cutoff

$$\Phi_c^M(sz) = \sum_{n=1}^{M} \frac{(sz)^n}{n^c}$$

The truncated function is defined also for $sz > 1$



C>2

$$\epsilon \equiv sz - 1 = (c-2)\frac{\ln M}{M} + O\left(\frac{\ln \ln M}{M}\right) \qquad M \to \infty\,, \epsilon \to 0$$

Canonical analysis

$Z(L_b, L_l) =$ partition sum of the model

L_b overall length of the bound segments

L_l overall length of the loops

$$L_b + L_l = L$$

$$Q(z_b, z_l) = \sum_{L_l, L_b} Z(L_b, L_l) z_b^{L_b} z_l^{L_l} = \left[\frac{1}{\omega z_b} - 1 - A\Phi(s z_l)\right]^{-1}$$

$$Z(L_b, L_l) = \frac{1}{2\pi i} \oint \frac{Q(z_b, z_l)}{z_b^{L_b+1} z_l^{L_l+1}} dz_b dz_l$$

the integral over z_b can be readily carried out

$$Z(L_b, L_l) = \frac{1}{2\pi i} \oint \frac{\omega^{L_b} \left(1 + A\Phi_c(s z_l)\right)^{L_b+1}}{z_l^{L_l+1}} dz_l$$

$$= \frac{1}{2\pi i} \oint e^{-LF(z_l, m_b)} dz_l$$

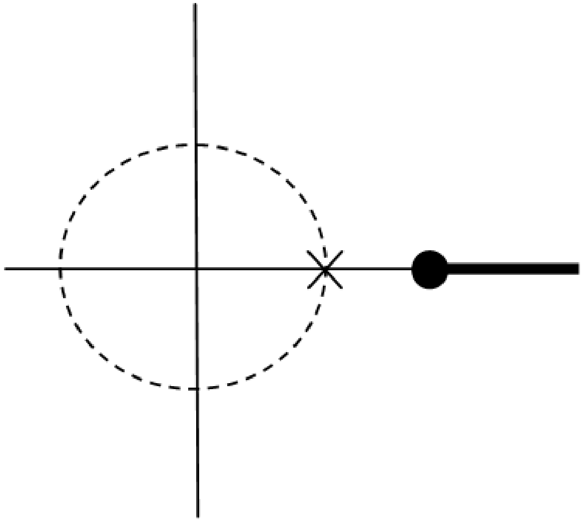
$$F(z_l, m_b) = -m_b \ \ln[\omega(1 + A\Phi_c(s z_l)] + (1 - m_b) \ln z_l$$

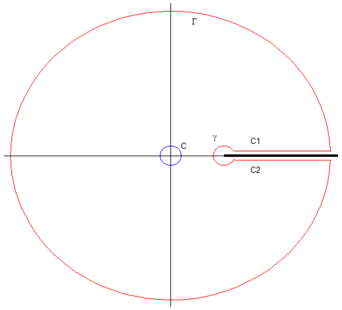
$$m_b = L_b / L \qquad\qquad L_l = L - L_b$$

$$Z(L_b, L_l) = \frac{1}{2\pi i} \oint e^{-LF(z_l, m_b)} dz_l$$

saddle point

$$\frac{dF}{dz} = \frac{1}{z}\left[1 - m\left(1 + \frac{A\Phi_{c-1}(sz)}{1 + A\Phi_c(sz)}\right)\right] = 0$$





$m > m_c$

$m < m_c$

$m_c = 0$ for $c \le 2$

$m_c < 1$ for $c > 2$

$$m = \left(1 + \frac{A\Phi_{c-1}(sz(m))}{1 + A\Phi_c(sz(m))}\right)^{-1}$$

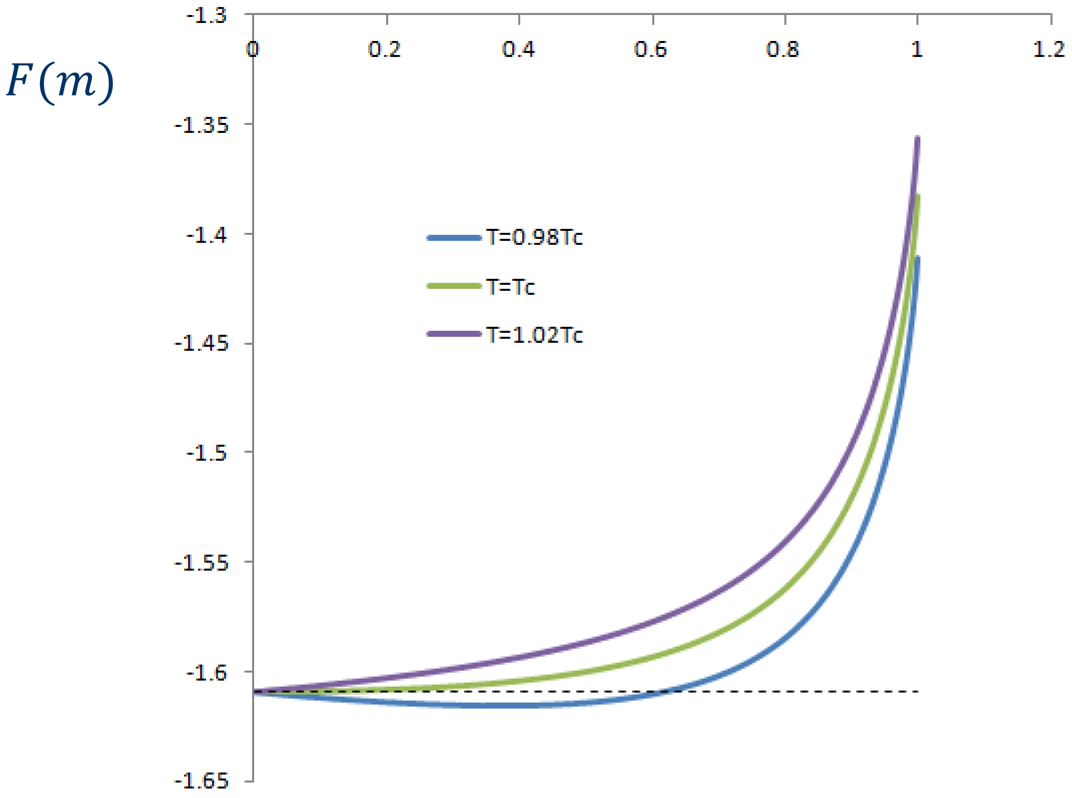
$$m_c = \left(1 + \frac{A\zeta_{c-1}}{1 + A\zeta_c}\right)^{-1}$$

$$F(z(m),m) = \begin{cases} -\ln\left[\omega\left(1 + A\Phi_c\left(sz(m)\right)\right) + (1-m)\ln z(m)\right] & m \geq m_c \\ -m\ln[\omega(1 + A\zeta_c)] - (1-m)\ln s & m < m_c \end{cases}$$

$$m_c = \left(1 + \frac{A\zeta_{c-1}}{1+A\zeta_c}\right)^{-1}$$

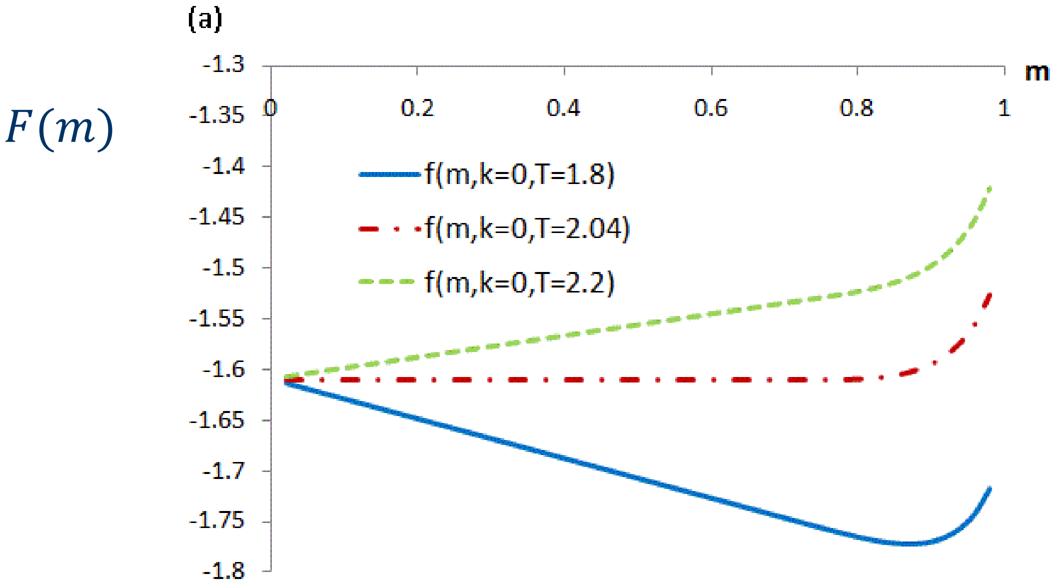
minimize $F(z(m),m)$

$c = 1.5 < 2$



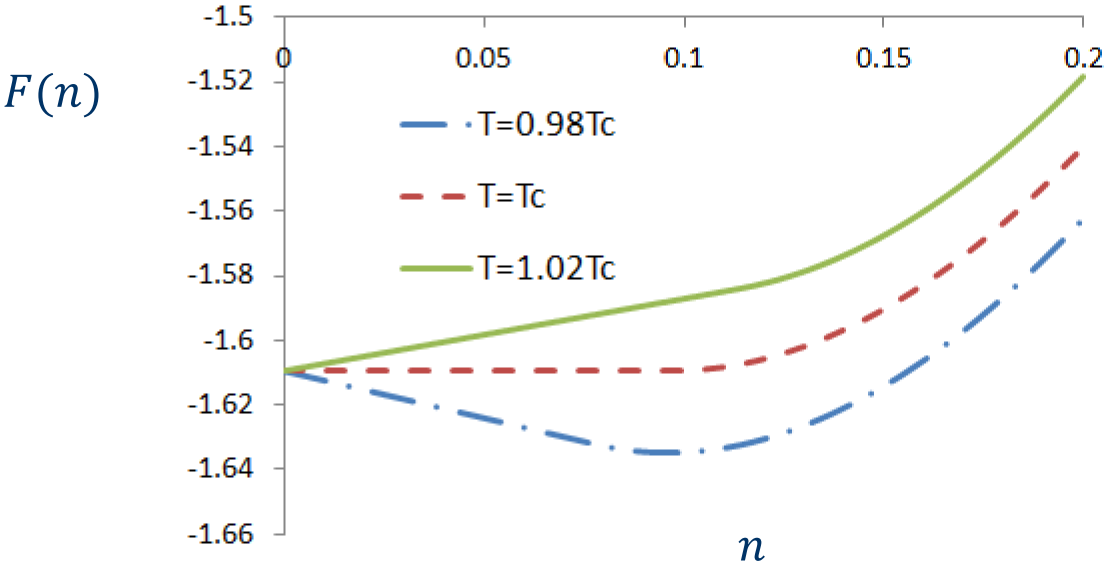
$$m_c = \left(1 + \frac{A\zeta_{c-1}}{1+A\zeta_c}\right)^{-1} = 0 \quad \text{for } c < 2$$

$$c = 2.5 > 2$$



$$m_c = \left(1 + \frac{A\zeta_{c-1}}{1+A\zeta_c}\right)^{-1} > 0 \quad \text{for } \ c > 2$$

n - density of loops N/L



c=2.5

Denaturation of circular DNA

one needs to absorb the extra twist generated by the denaturated loops

two mechanisms:

- over-twist of the rest of the chain with an energy cost:

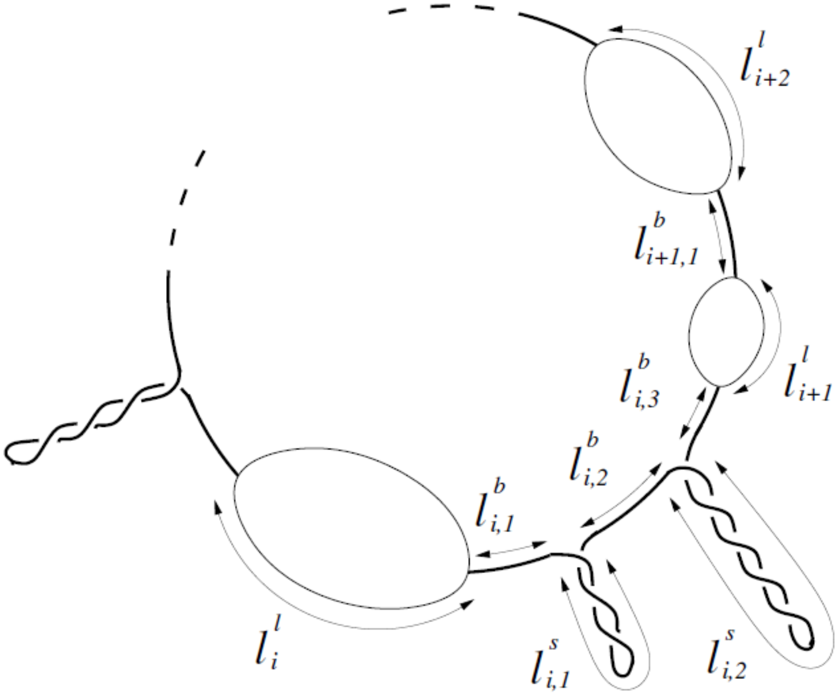
$$H = \kappa \frac{L_l^2}{L_b} + \epsilon L_b$$

- introduce wirthe (a twist of the backbone)

Rudnick, Bruinsma (2002); Garel, Orland, Yeramian (2004); Kabacioglu Orlandini, DM (2009); Bar, Kabacioglu, DM (2011)

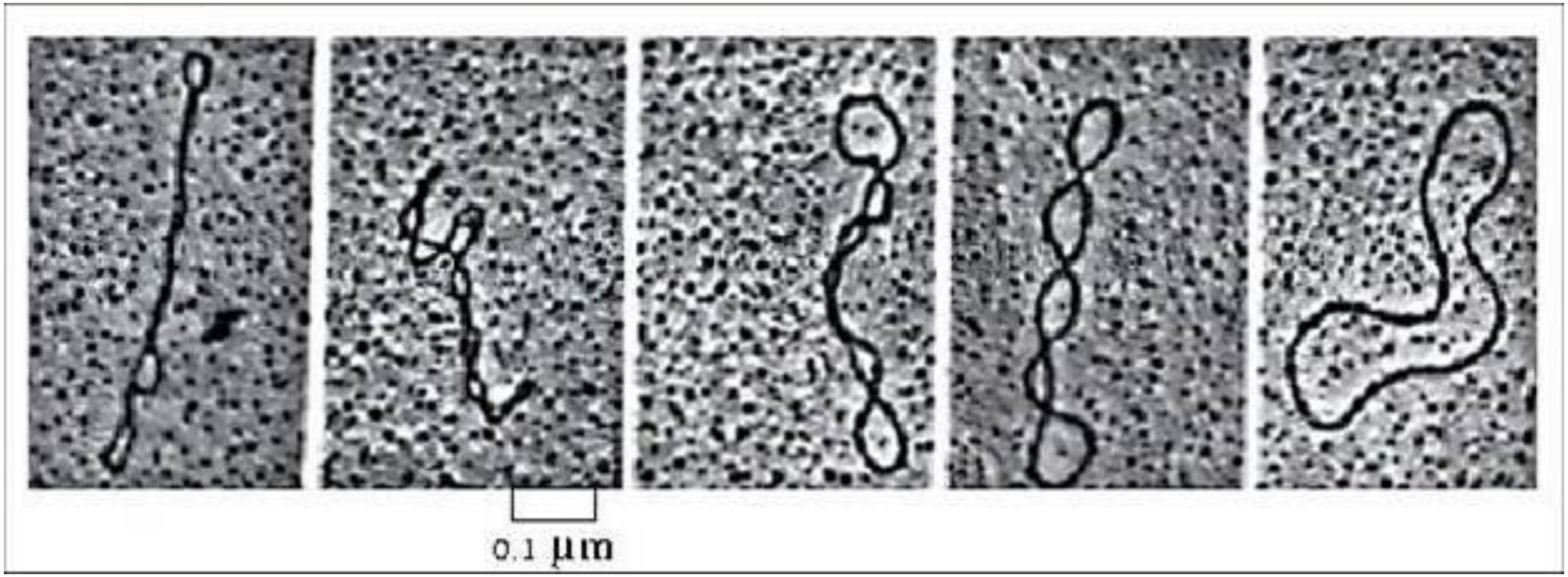
writhe mechanism

typical configuration



the linking number is a topological invariant:

linking number = twist + writhe


0.1 µm

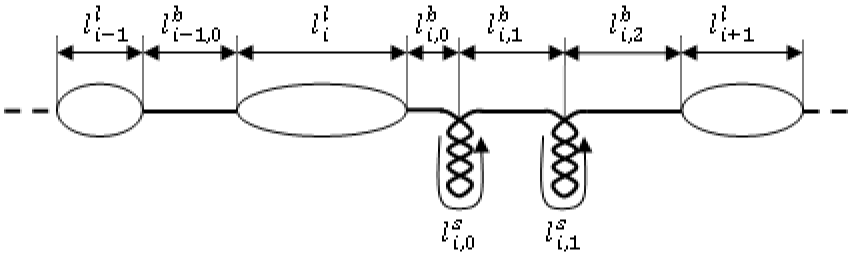
L_l – overall length of loops

L_b – overall length of bound segments

L_s – overall length of supercoiled segments

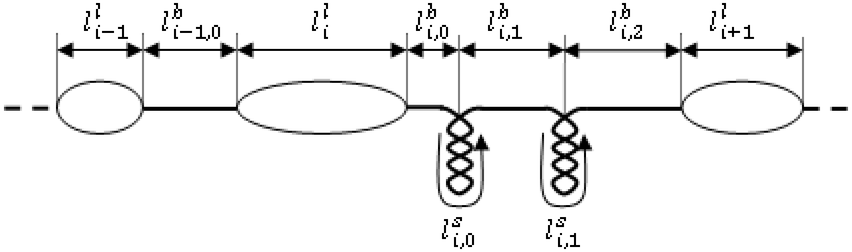
$$L_b + L_l + L_s = L$$

$$L_l - L_s = 0$$ (conserved linking number)



Boltzmann weight of a configuration

ω Boltzmann weight of a bound pair
ν Boltzmann weight of a supercoiled pair



$$\Omega\left(l^l_{i-1,0}\right) \times \omega^{l^b_{i-1,0}} \times \Omega\left(l^l_{\mathrm{i}}\right) \times \omega^{l^b_{i,0}} \times \nu^{l^{l^S_{i,o}}} \ldots\ldots$$

Grand canonical approach

$$Q(z,\mu) = \sum Z(L_b, L_l - L_s) z^{L_b + L_l + L_s} \mu^{L_l - L_s}$$

$$\boxed{Q(z,\mu) = \frac{\tilde{V}(z,\mu)}{1 - \tilde{V}(z,\mu) U(z,\mu)}}$$

$$\tilde{V}(z,\mu) = \frac{V(z)}{1 - V(z) W(\frac{Z}{\mu})}$$

$$V(z) = \sum_{n=1}^{\infty} (\omega z)^n = \frac{\omega z}{1 - \omega z}$$

$$W\left(\frac{z}{\mu}\right) = \sum_{n=1}^{\infty} \left(\frac{\nu z}{\mu}\right)^n = \frac{\nu z}{\mu - \nu z}$$

$$U(z\mu) = \sum_{n=1}^{\infty} A \frac{(sz\mu)^n}{n^c} = \mathrm{A}\Phi_c(sz\mu)$$

$$Q(z,\mu) = \left[\left(\frac{1}{\omega z} - 1\right) - \frac{\nu z}{\mu - \nu z} - A\Phi_c(sz\mu)\right]^{-1}$$

$$L = \frac{\partial \ln Q}{\partial \ln z} \qquad L_l - L_s = \frac{\partial \ln Q}{\partial \ln \mu}$$

$L = \infty$

$$\left(\frac{1}{\omega z} - 1\right) - \frac{\nu z}{\mu - \nu z} = \mathrm{A}\Phi_c(sz\mu)$$

$L_l - L_s = 0$

$$\frac{\nu z}{(\mu - \nu z)^2} = \frac{A}{\mu}\Phi_{c-1}(sz\mu)$$

$$\left(\frac{1}{\omega z}-1\right)-\frac{\nu z}{\mu-\nu z}=\mathrm{A}\Phi_c(sz\mu)$$

$$\frac{\nu z}{(\mu-\nu z)^2}=\frac{A}{\mu}\Phi_{c-1}(sz\mu)$$

$\underline{c \leq 2 \qquad\qquad\qquad \text{no phase transition}}$

for example for $1<c\leq 2$

$\Phi_{c-1}(x)\rightarrow\infty$ while $\Phi_c(x)<\infty$ for $x\rightarrow 1$

thus $sz\mu=1$ is not a solution of these equations.

$c > 2$

for $T > T_M$

$$\left(\frac{1}{\omega z} - 1\right) - \frac{\nu z}{\mu - \nu z} = \mathrm{A}\Phi_c(sz\mu)$$

$$\frac{\nu z}{(\mu - \nu z)^2} = \frac{A}{\mu}\Phi_{c-1}(sz\mu)$$

$$sz\mu = 1$$

three equations for the two fugacities z, μ

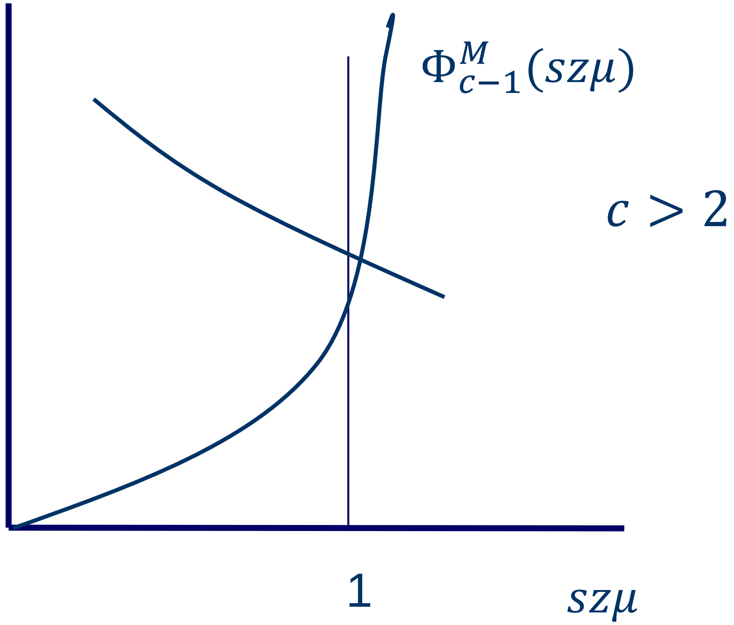
one needs to treat the limit $sz\mu$ more carefully by regularizing $U(sz\mu)$

$$U_M(sz\mu) = A\sum_{n=1}^{M}\frac{(sz\mu)^n}{n^c} \equiv A\Phi_c^M(sz\mu)$$

$$\left(\frac{1}{\omega z}-1\right)-\frac{\nu z}{\mu-\nu z}=\mathrm{A}\Phi_c(sz\mu)$$

$$\frac{\nu z}{(\mu-\nu z)^2}=\frac{A}{\mu}\Phi_{c-1}(sz\mu)$$

$$sz\mu=1$$



$$\left(\frac{1}{\omega z}-1\right)-\frac{\nu z}{\mu-\nu z}=\mathrm{A}\zeta_c$$

$$sz\mu=1$$

$$\frac{\nu z}{(\mu-\nu z)^2}=\frac{A}{\mu}(\zeta_{c-1}+a_0)$$

$$a_0=\frac{\nu}{As(\mu-\nu z)^2}-\zeta_{c-1}$$

nature of the denaturation transition

$$m_b = \frac{\partial \ln z}{\partial \ln \omega} \qquad m_s = \frac{\partial \ln z}{\partial \ln \nu} \qquad m_l = \frac{\partial \ln z}{\partial \ln s}$$

$$m_b = \left[1 + \frac{2\omega\nu\mu z^2}{(\mu - \nu z)^2}\right]^{-1}$$

$T > T_M$

$$\left(\frac{1}{\omega z} - 1\right) - \frac{\nu z}{\mu - \nu z} = \mathrm{A}\zeta_c$$

$$sz\mu = 1$$

$T < T_M$

$$\left(\frac{1}{\omega z} - 1\right) - \frac{\nu z}{\mu - \nu z} = \mathrm{A}\Phi_c(sz\mu)$$

$$\frac{\nu z}{(\mu - \nu z)^2} = \frac{A}{\mu}\Phi_{c-1}(sz\mu)$$

$$\Phi_{c-1}(1-\epsilon) = \zeta_{c-1} + \zeta_{c-2}\epsilon + \cdots + \Gamma(c-2)\epsilon^{c-2} + \cdots$$

$$\delta m_b(t) = \begin{cases} g(t) & t < 0 \\ g(t) + \alpha t^{\eta} & t > 0 \end{cases}$$

$$\eta = \begin{cases} \dfrac{1}{c-2} & 2 < c \leq 3 \\ 1 & c \geq 3 \end{cases}$$

$$t = \frac{T_M - T}{\mathrm{T_M}}$$

$g(t)$ −analytic function near $t = 0$

free energy: $$F(t) = \begin{cases} f(t) \\ f(t) + \gamma t^{\eta+1} \end{cases}$$

order of the transition $$\begin{cases} \left\lceil \dfrac{c-1}{c-2} \right\rceil & 2 < c \leq 3 \\ 2 & c \geq 3 \end{cases}$$

typical curves

$$c = 4.5 \qquad s = 5 \qquad A = 0.1$$

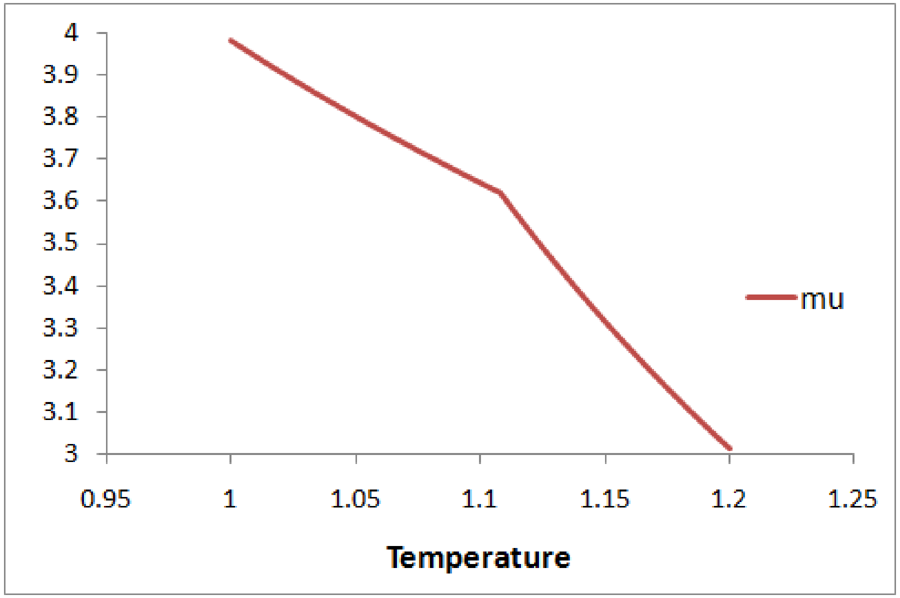
N_b –number of bound segments

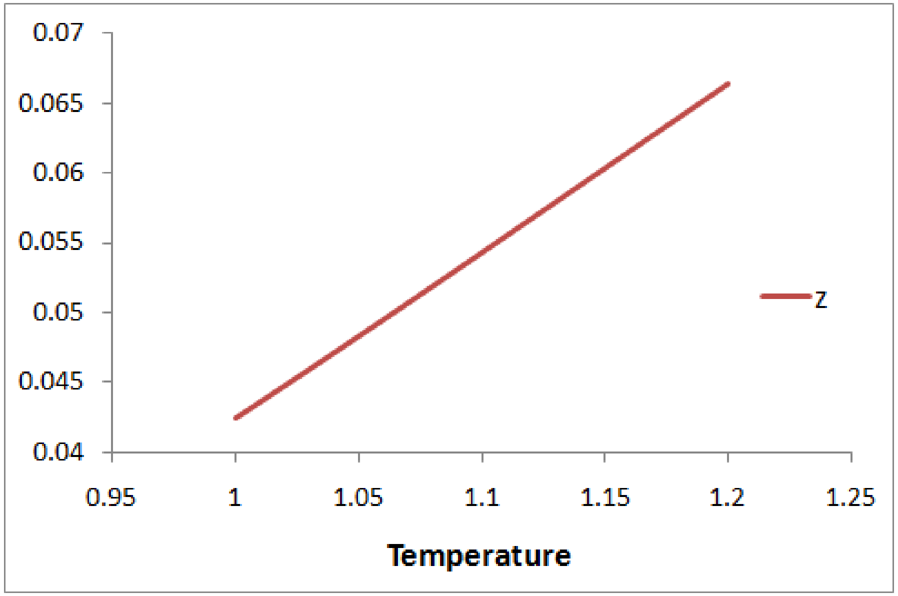
L_b –total length of bound segments

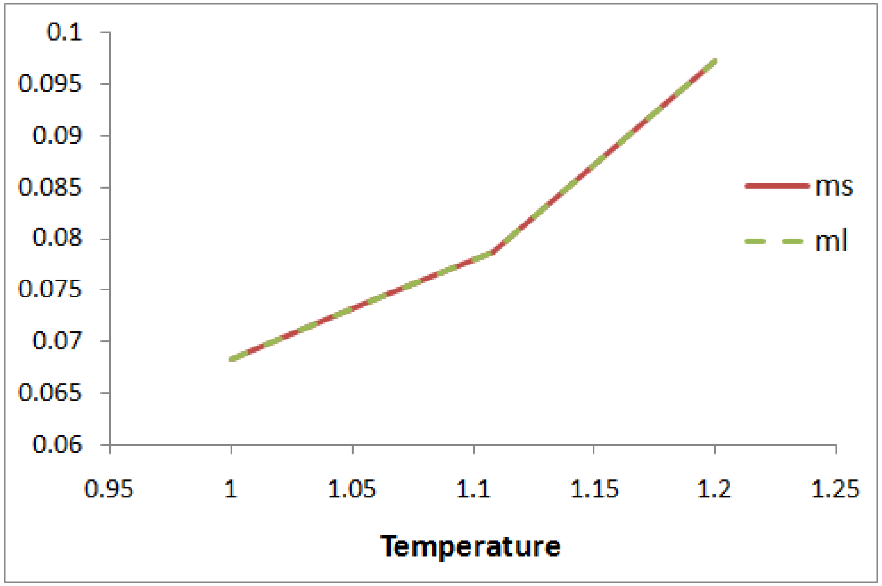
$m_b = \frac{L_b}{L}$ fraction of the overall bound pairs

$l_b = \frac{L_b}{L}$ average length of a bound segment

$n_b = \frac{m_b}{l_b} = \frac{\mathrm{N_b}}{\mathrm{L}}$ density of bound segments



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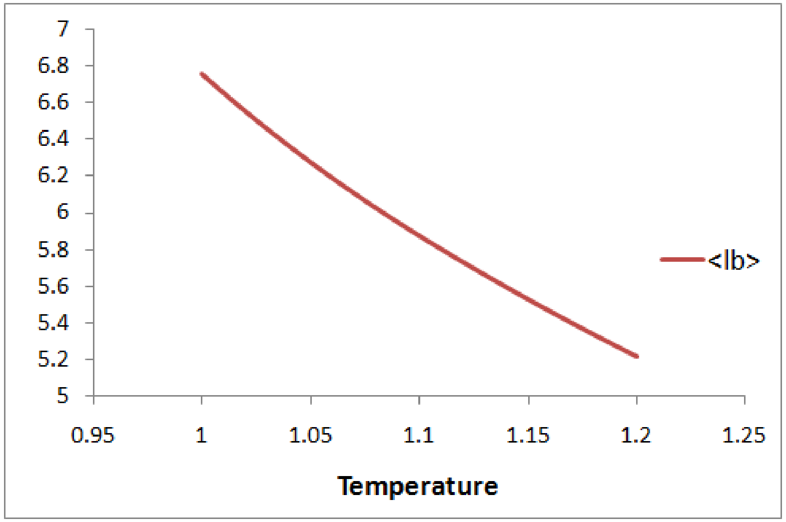
1.15
1.2
1.25
Temperature
ms
ml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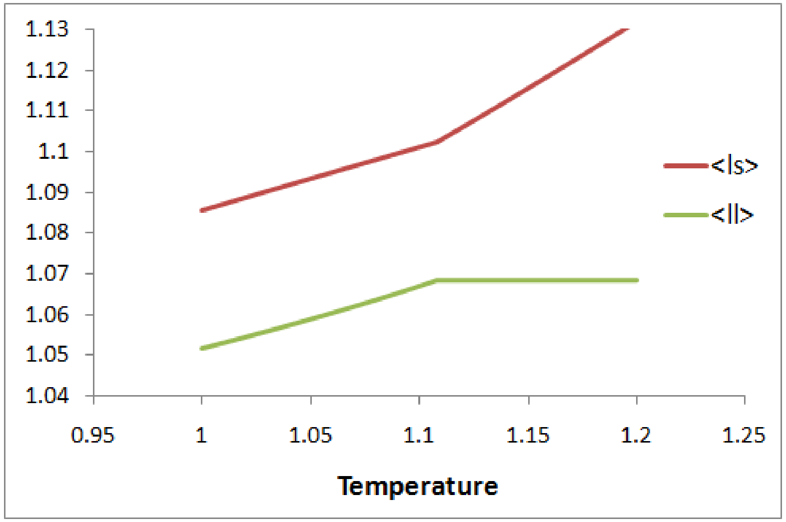
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Temperature
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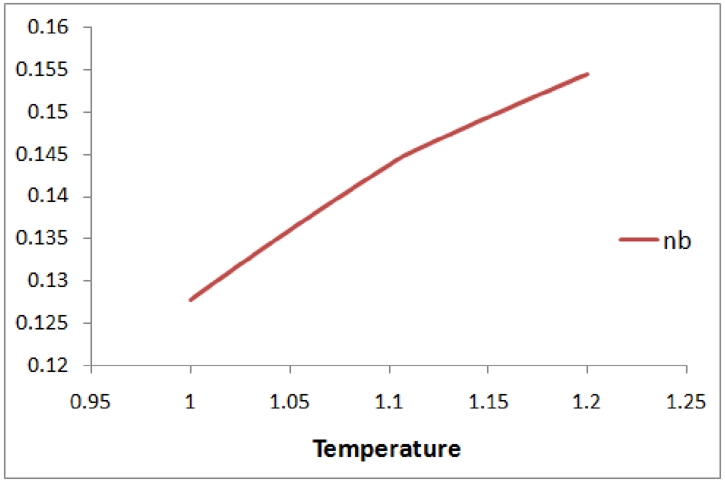
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1.15
1.2
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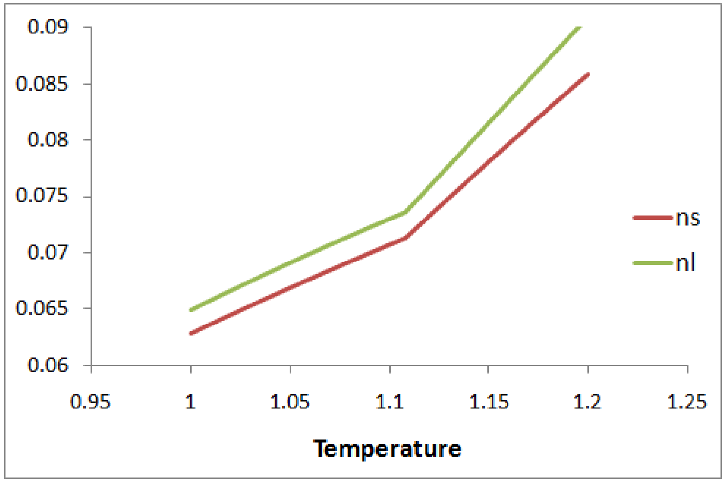
Temperature
nb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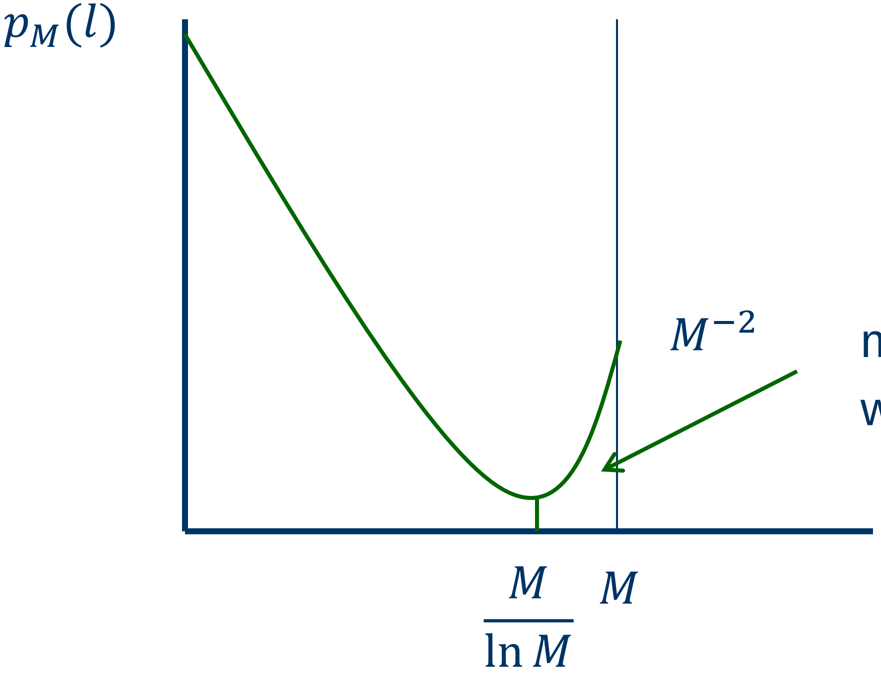
0.075
0.07
0.065
0.06
0.95
1
1.05
1.1
1.15
1.2
1.25
Temperature
ns
nl

loop-length distribution and existence of a macroscopic loop at $T > T_M$

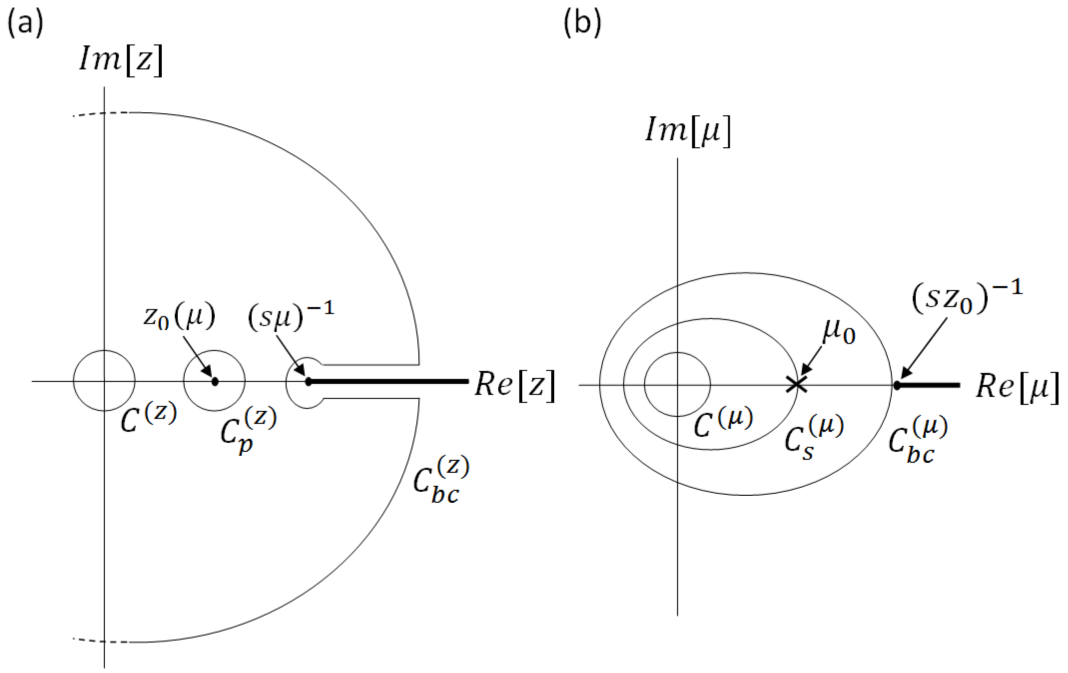
$$p_M(l) = \frac{1}{\Phi_c^M(sz\mu)} \frac{(sz\mu)^l}{l^c} \qquad sz\mu = e^{\epsilon} \quad \epsilon = (c-2)\frac{\ln M}{M}$$



macroscopic loop weight $O(\frac{1}{M})$

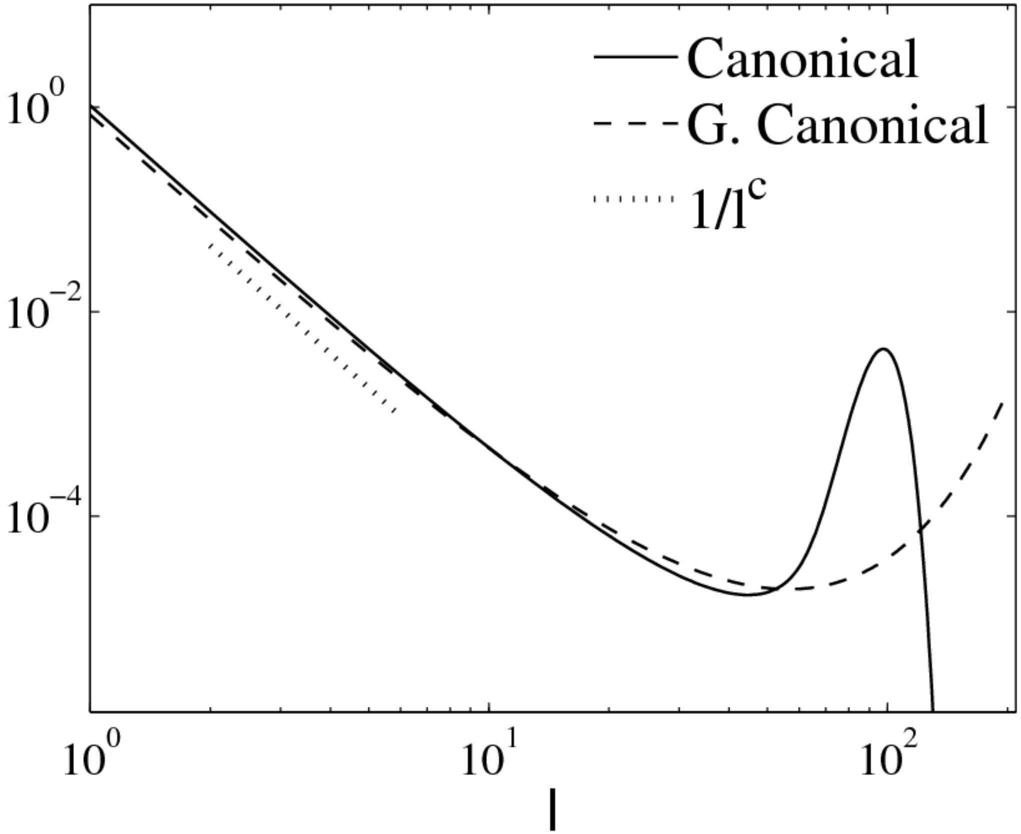
Canonical approach

$$Z(L, L_l - L_s) = \frac{1}{(2\pi i)^2} \oint d\mu \oint dz \frac{Q(z,\mu)}{z^{L+1} \mu^{L_l - L_s + 1}}$$



calculate the loop size distribution

$$P(l) = A\frac{s^l}{l^c}\frac{Z(L-l,-l)}{Z(L,0)} \qquad (L \to L-l \quad ; \quad L_l - L_s \to -l)$$



summary

linear DNA		Circular DNA	
$c \leq 1$	no transition	$c \leq 2$	no transition
$1 < c \leq 2$	continuous transition	$2 < c \leq 3$	transition of high order $\left\lceil \frac{c-1}{c-2} \right\rceil$
$c > 2$	first order transition	$c > 3$	second order transition

canonical vs grand-canonical results for the PS model