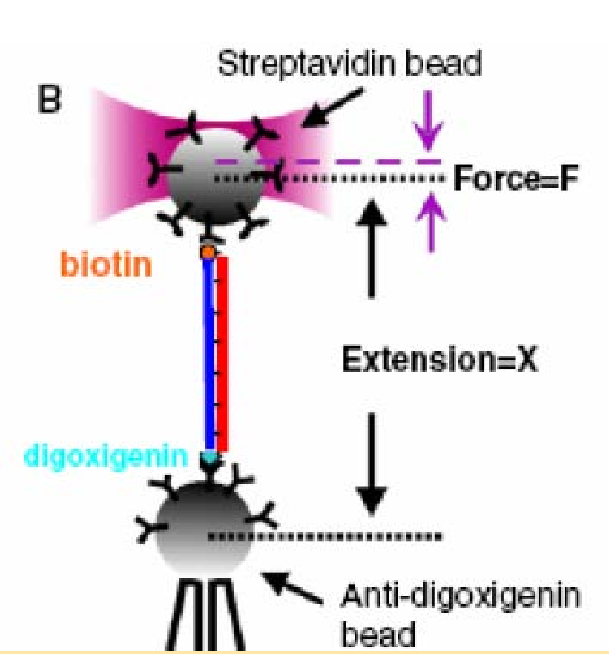
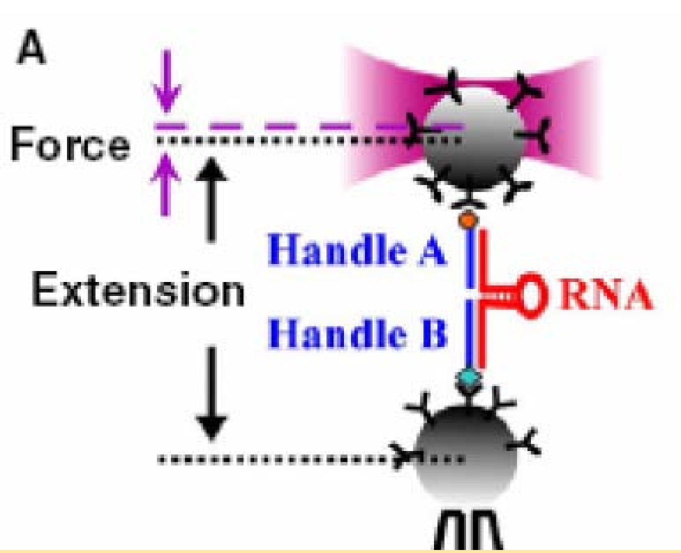
Models for Single Molecule Experiments





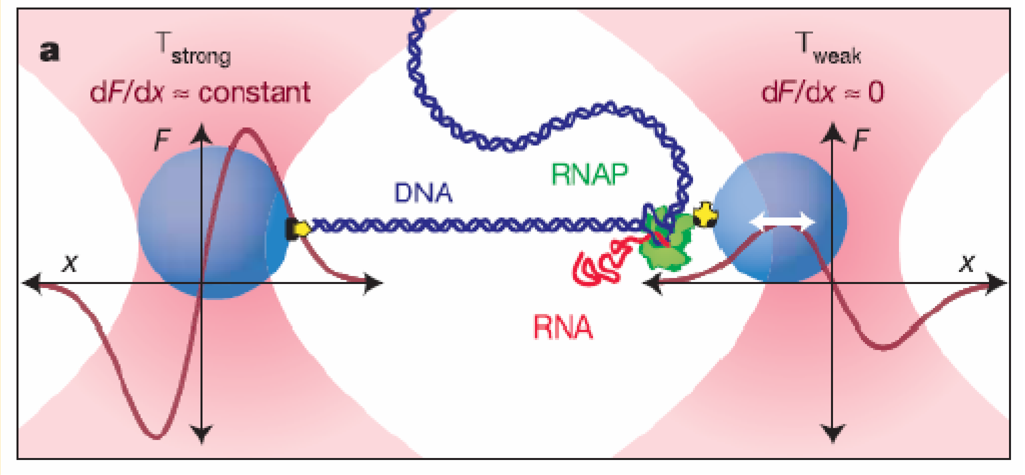
2 Binding – Unbinding

Folding - Unfolding

-> Kramers – Bell theory

1 Polymer elasticity:

- Hooke's law (?)
- FJC (Freely joined chain)
- WLC (Worm-like chain)



3 Molecular Motors

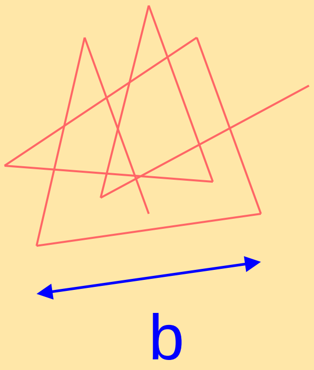
- Brownian Ratchet model
- Power-Stroke model

The Elasticity of a Polymer Chain

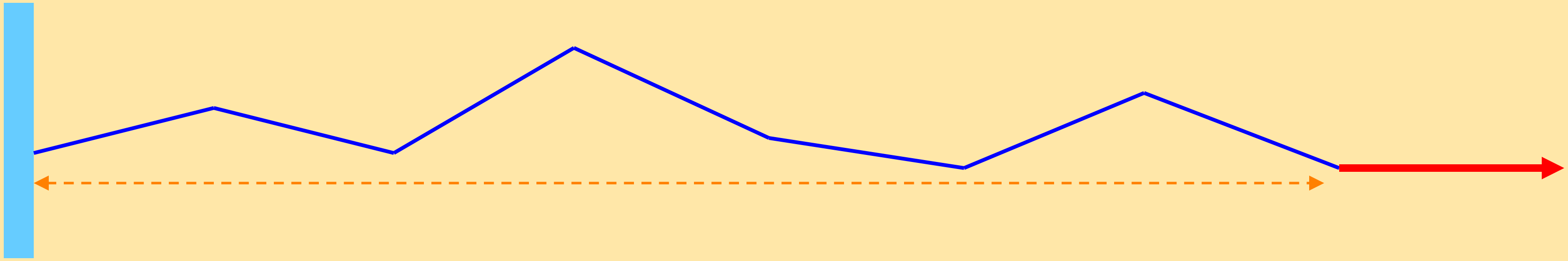
The freely jointed chain (FJC) model:

Treats the polymer as made up of orientationally independent statistical segments (known as Kuhn segments)

Can think of the chain as a 3D random walk.



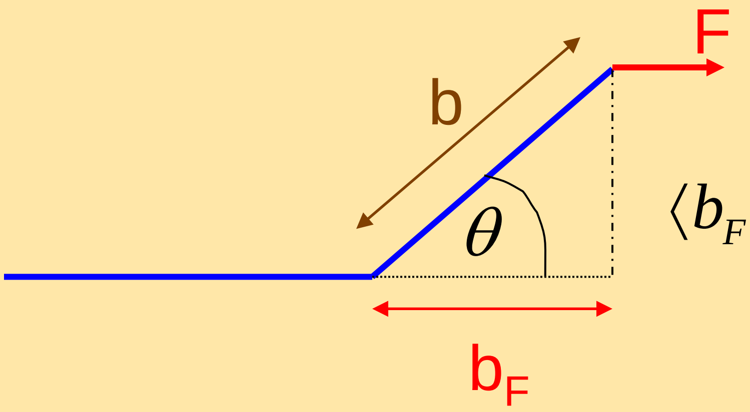
The Elasticity of a Polymer Chain



In the presence of a force, F, the segments tend to align in the direction of the force.

Opposing the stretching is the tendency of the chain to maximize its entropy. Extension corresponds to the equil. Point between the external force and the entropic elastic force of the chain.

The Freely Jointed Chain Model



$$\langle b_F \rangle = \frac{\int_0^{\pi} b\cos(\theta) 2\pi b^2 \sin(\theta) d\theta \, e^{\frac{Fb\cos(\theta)}{k_B T}}}{\int_0^{\pi} 2\pi b^2 \sin(\theta) d\theta \, e^{\frac{Fb\cos(\theta)}{k_B T}}}$$

Where -F x b cos(θ) is the potential energy acquired by a segment aligned along the direction θ with an external force F. Integration leads to:

$$\langle b_F \rangle = b\left[\coth\left(\frac{Fb}{k_B T}\right) - \frac{k_B T}{Fb}\right] = b\mathfrak{I}\left(\frac{Fb}{k_B T}\right)$$ *Langevin Function*

And for a polymer made up of N statistical segments its ave. end-to-end distance is:

$$\langle R(F) \rangle = Nb\mathfrak{I}\left(\frac{Fb}{k_B T}\right) = L\mathfrak{I}\left(\frac{Fb}{k_B T}\right)$$

The Freely Jointed Chain Model

At low forces:

$$x/L = \coth\left(\frac{Fb}{k_BT}\right) - \frac{k_BT}{Fb}, \qquad \text{for } Fb << k_BT:$$

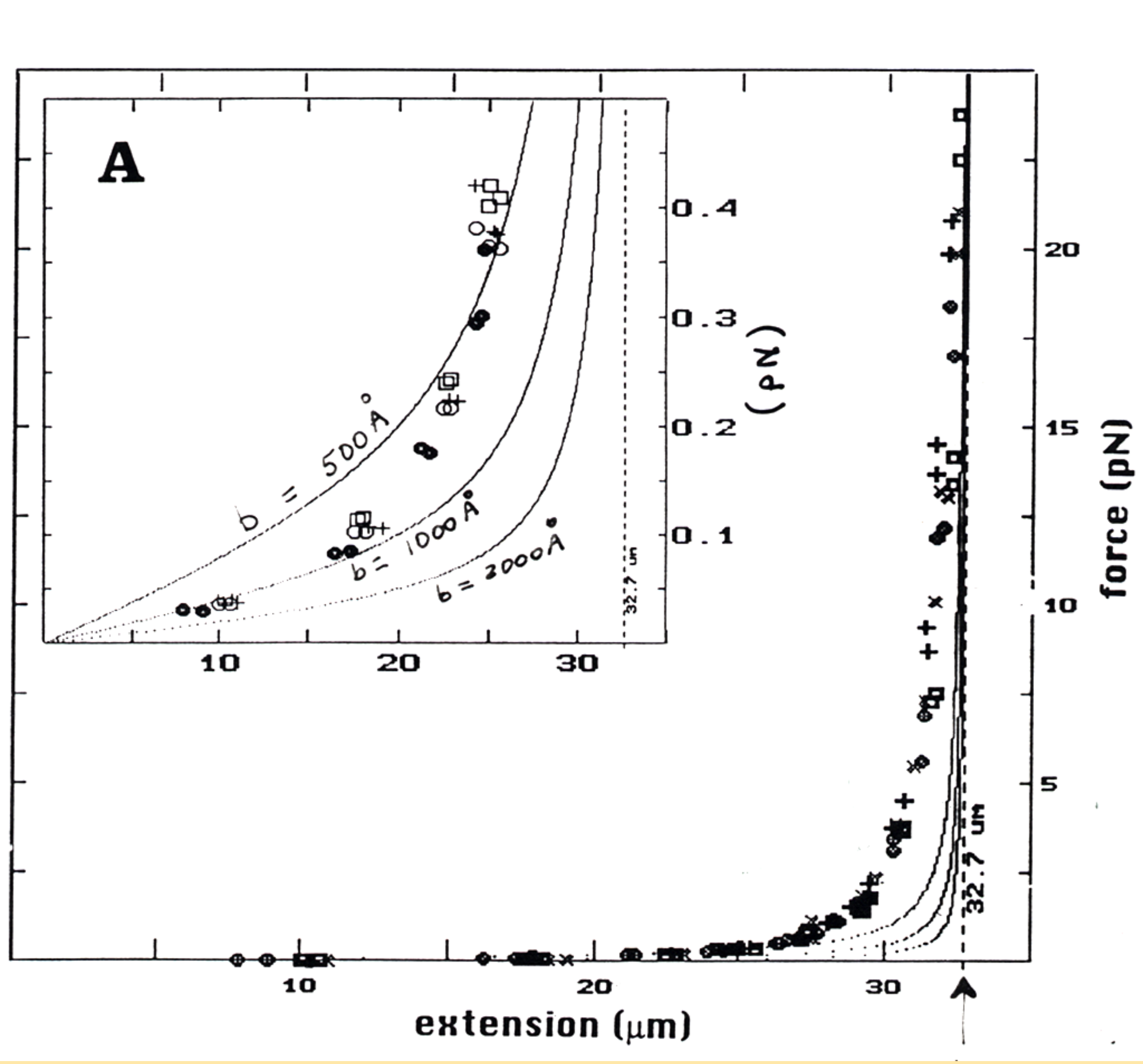
$$\coth(a) = \frac{1}{a} + \frac{a}{3} + \ldots, \text{ therefore:}$$

$$\text{x/L} = \frac{1}{a} + \frac{a}{3} - \frac{1}{a} + \ldots. = \frac{Fb}{3k_BT}$$

$$F = \frac{3k_BT}{b}\left(\frac{x}{L}\right)$$

Thus, at low forces, the chain behaves as a hookian spring with a spring constant, $\kappa = \partial F/\partial x =$ $3k_BT/b$ = $3k_BT/2P$, where P is the persistence length of the chain. Lets see its physical meaning…..

Limitations of the Freely-Jointed Chain Model



Limitations of the Freely-Jointed Chain Model

At very low (< 100 fN) and at high forces (> 5 pN), the FJC does a good job.

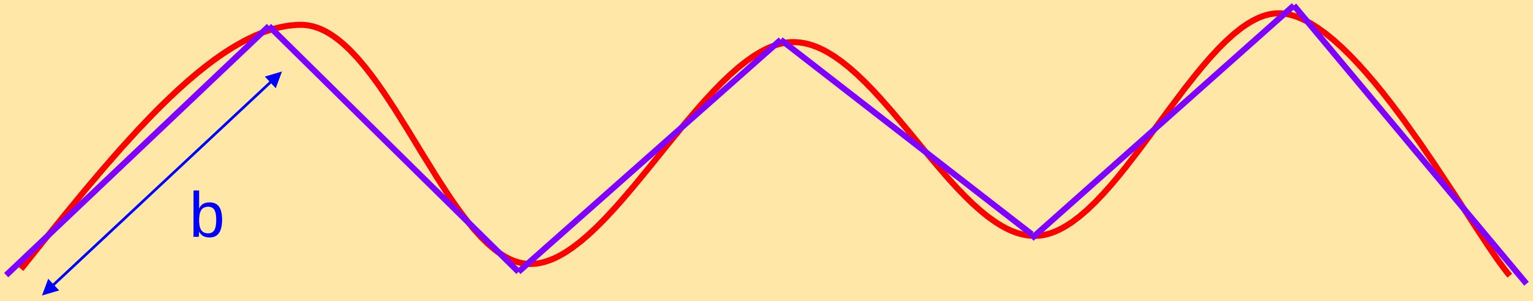
At intermediate forces, however, it is seen that it takes more force than that predicted by the FJC to stretch the molecule the same amount In fact the experimental data "cuts the corner" on the theoretical curve.

One problem with the FJC is that it tends to *COARSE GRAIN* the description of the polymer molecule: Each Kuhn segment has a fixed length, is unstretchable and completely straight.

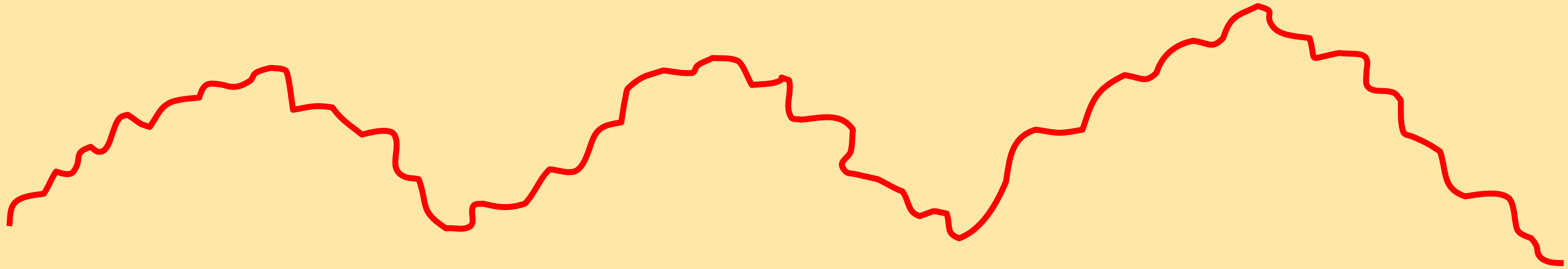
No thermal fluctuations away from the straight line are allowed
The polymer can only disorder at the joints between segments

Coarse Grained Description in the FJC Model

Idealized FJC:



Realistic Chain:



The Worm-like Chain Model

Uses a continuum description of the chain to address these limitations:

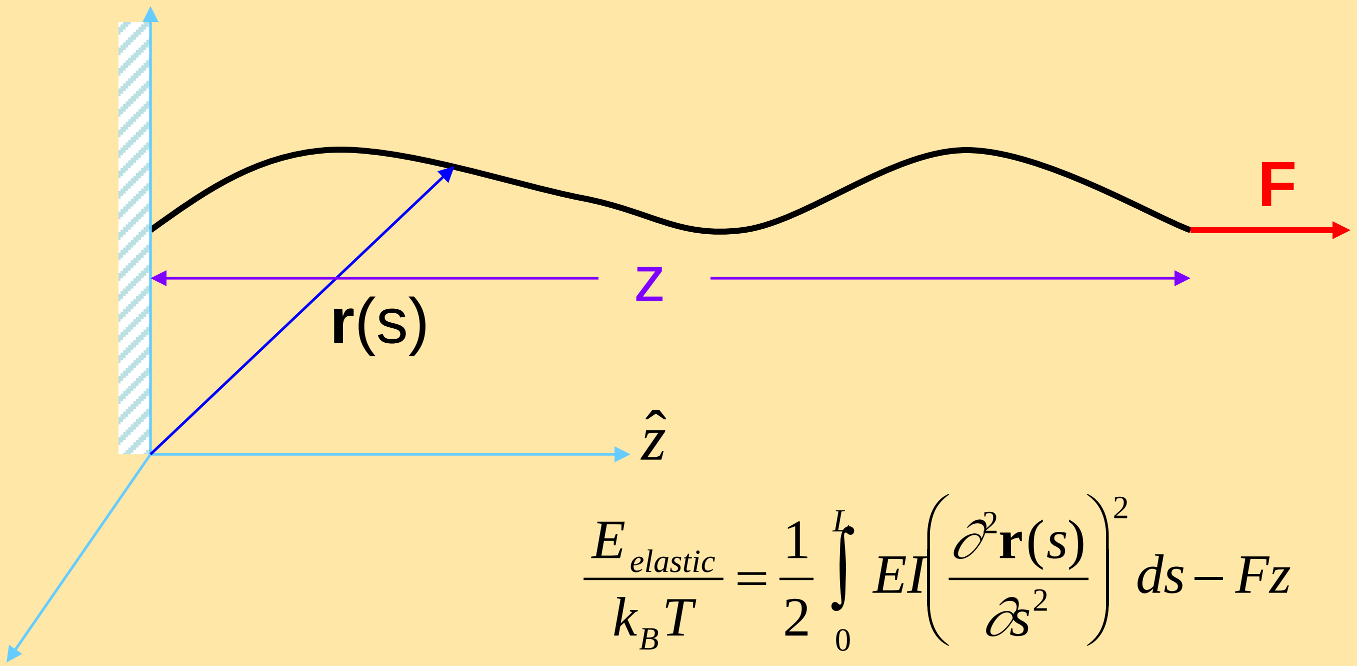
- The entropic elasticity of the DNA chain involves small deviations of the molecular axis due to thermal fluctuations.

- The direction of the chain is correlated over a distance, called the *persistence length* of the chain. For DNA, at 10 mM NaCl, P_{DNA} = 150 bp or 550 nm.

- Thus, forces of the order of k_BT/P are needed to align and straighten elastic units of these dimensions.

The Worm-like Chain Model

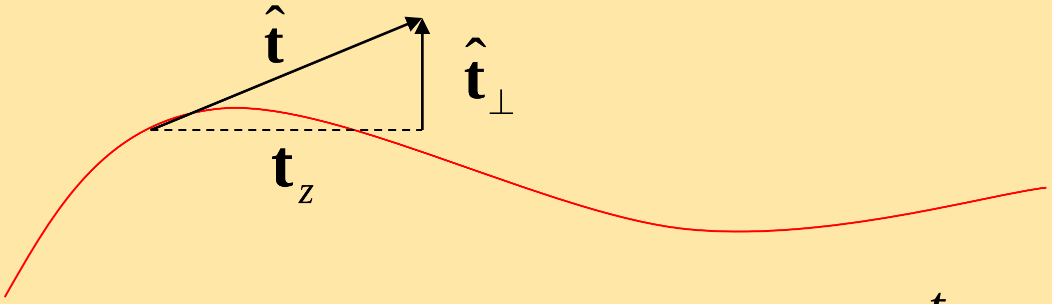
For forces > $k_B T/P$, the force-extension behavior of the chain can be obtained from the effective energy of a stretched WLC:



$$\frac{E_{elastic}}{k_B T} = \frac{1}{2}\int_0^L EI\left(\frac{\partial^2 \mathbf{r}(s)}{\partial s^2}\right)^2 ds - Fz$$

At high forces, the extension of the WLC approaches its coutour length, L, and the tangent vectors $\hat{t}$ fluctuate only slightly around the pulling axis, $\hat{z}$

The Worm-like Chain Model



$$t_{\mathbf{z}} = \sqrt{1 - \mathbf{t}_{\perp}^{2}} \cong 1 - \frac{\mathbf{t}_{\perp}^{2}}{2} + \vartheta(\mathbf{t}_{\perp}^{4}) + ...$$

Now:

$$\mathbf{k} \equiv \frac{\partial^2 \mathbf{r}(s)}{\partial s^2} = \frac{\partial \hat{\mathbf{t}}}{\partial s} = \frac{\partial}{\partial s}(t_z \hat{\mathbf{z}} + \hat{\mathbf{t}}_{\perp})$$

$$\mathbf{k} = \frac{\partial}{\partial s}\left[(1 - \frac{\mathbf{t}_{\perp}^{2}}{2})\hat{\mathbf{z}} + \mathbf{t}_{\perp} \right]$$

$$\mathbf{k} = -\left(\mathbf{t}_{\perp} \bullet \frac{\partial \mathbf{t}_{\perp}}{\partial s} \right)\hat{\mathbf{z}} + \frac{\partial \mathbf{t}_{\perp}}{\partial s}$$

Since $\mathbf{t}_{\perp} \bullet \frac{\partial \mathbf{t}_{\perp}}{\partial s} = t_{\perp} \frac{\partial \mathbf{t}_{\perp}}{\partial s}$, then

$$|\mathbf{k}| = \sqrt{\frac{\partial \mathbf{t}_{\perp}}{\partial s}^{2} \left(1 + t_{\perp}^{2}\right)} \cong \frac{\partial \mathbf{t}_{\perp}}{\partial s}$$

$$\mathbf{k}^2 = \left(\frac{\partial \mathbf{t}_{\perp}}{\partial s} \right)^2$$

Therefore, we can write the energy of the stretched molecule:

The Worm-like Chain Model

$$\mathrm{E}_{elastic} = \frac{1}{2}\int_0^L \left[EI\left(\frac{\partial \mathbf{t}_\perp}{\partial s}\right)^2 + F\mathbf{t}_\perp{}^2 \right] ds - FL$$

Where we have written:

$$z = \int_0^L t_z ds, \quad \text{and} \quad L = \int_0^L ds$$

Next, we switch to Fourier space to decompose the energy into normal modes. Using:

$$\tilde{\mathbf{t}}_\perp(q) = \int e^{iqs}\mathbf{t}_\perp(s) ds$$

we can write

$$E_{elastic} = \frac{1}{2}\left(\frac{1}{2\pi}\int \left[Aq^2 + F\right] \left|\tilde{\mathbf{t}}_\perp(q)\right|^2 dq\right) - FL$$

where $A \equiv EI$

The Worm-like Chain Model

Each normal mode has associated with it energy $k_B T/2$ by equipartition, thus:

$$\frac{1}{2}\left(Aq^2 + F\right)\left\langle \left|\tilde{\mathbf{t}}_\perp\right|^2 \right\rangle = 2 \times \frac{k_B T}{2}$$

The factor of 2 accounting for the x and y components of $\tilde{\mathbf{t}}_\perp$

$$\left\langle \left|\tilde{\mathbf{t}}_\perp\right|^2 \right\rangle = \frac{2k_B T}{Aq^2 + F}$$

Next we calculate $\mathbf{t}_\perp{}^2(s)$

$$\mathbf{t}_\perp{}^2(s) = \frac{1}{2\pi}\int e^{-iqs}\tilde{\mathbf{t}}_\perp(q)dq \times \frac{1}{2\pi}\int e^{-iq's}\tilde{\mathbf{t}}_\perp(q')dq'$$

$$\mathbf{t}_\perp{}^2(s) = \frac{1}{4\pi^2}\int\int e^{-i(q-q')s}\tilde{\mathbf{t}}_\perp(q)\bullet\tilde{\mathbf{t}}_\perp(q')dqdq'$$

The Worm-like Chain Model

$$\mathbf{t}_\perp{}^2(s) = \frac{2\pi}{4\pi^2} \int \int \delta(q-q')\tilde{\mathbf{t}}_\perp(q) \bullet \tilde{\mathbf{t}}_\perp(q')\,dq\,dq'$$

$$\mathbf{t}_\perp{}^2(s) = \frac{1}{2\pi} \int \tilde{\mathbf{t}}_\perp(q) \bullet \tilde{\mathbf{t}}_\perp(q)\,dq$$

Then,

$$\langle \mathbf{t}_\perp{}^2(s) \rangle = \frac{1}{2\pi} \int \langle \left| \tilde{\mathbf{t}}_\perp(q) \right|^2 \rangle\, dq$$

$$\langle \mathbf{t}_\perp{}^2(s) \rangle = \frac{2}{2\pi} \int \frac{k_B T}{Aq^2 + F}\, dq$$

$$\langle \mathbf{t}_\perp{}^2(s) \rangle = \frac{k_B T}{\sqrt{FA}}$$

The Worm-like Chain Model

Now, the extension of the chain in the z-direction is:

$$\frac{z}{L} = \frac{1}{L}\int_0^L (\hat{\mathbf{t}} \bullet \hat{\mathbf{z}})\, ds = \frac{1}{L}\int_0^L \left(1 - \frac{\langle \mathbf{t}_\perp{}^2 \rangle}{2}\right) ds$$

$$\frac{z}{L} = 1 - \frac{\langle \mathbf{t}_\perp{}^2 \rangle}{2} = 1 - \frac{k_B T}{\sqrt{4FA}}$$

From this expression we can obtain:

$$\frac{FA}{(k_B T)^2} = \frac{1}{4\left(1 - \frac{z}{L}\right)^2}$$

Now, $A = EI = k_B TP$, where P is the persistence length of the chain. Then:

The Worm-like Chain Model

$$\frac{FP}{k_B T} = \frac{1}{4\left(1-\frac{z}{L}\right)^2}$$

Expanding the term on the rhs for small extensions (small force limit):

$$\cong \frac{1}{4}\left(1+2\frac{z}{L}+\ldots\right)$$

Now, we have seen that at low forces, the chain follows the FJC model with behavior like:

$$\frac{FP}{k_B T} = \frac{3}{2}\left(\frac{z}{L}\right)$$

Thus, we can obtain an interpolation formula that describes The whole range of forces and extensions of the polymer simply by adding to the high force regime the terms:

The Worm-like Chain Model

$$\frac{z}{L} \quad - \quad \frac{1}{4}$$

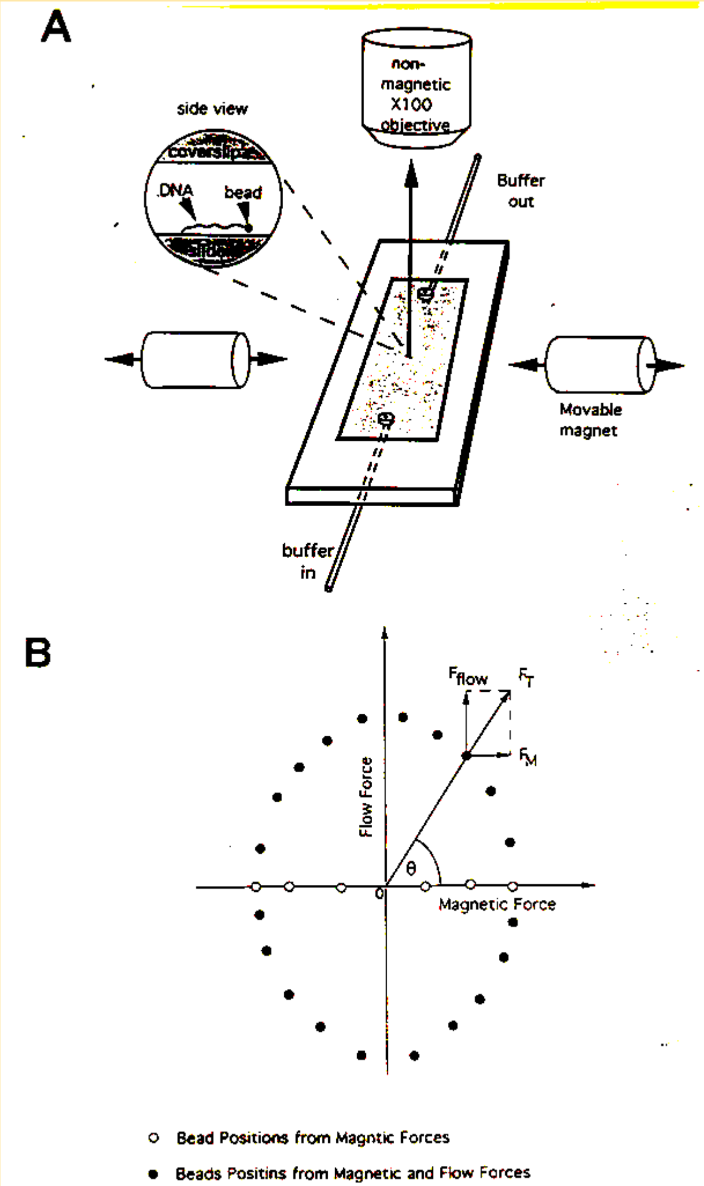
So that:

$$\frac{FP}{k_B T} = \frac{1}{4\left(1 - \frac{z}{L}\right)^2} + \frac{z}{L} - \frac{1}{4}$$

Bustamante *et al. Science*, **265**, 1599-1600 (1994)

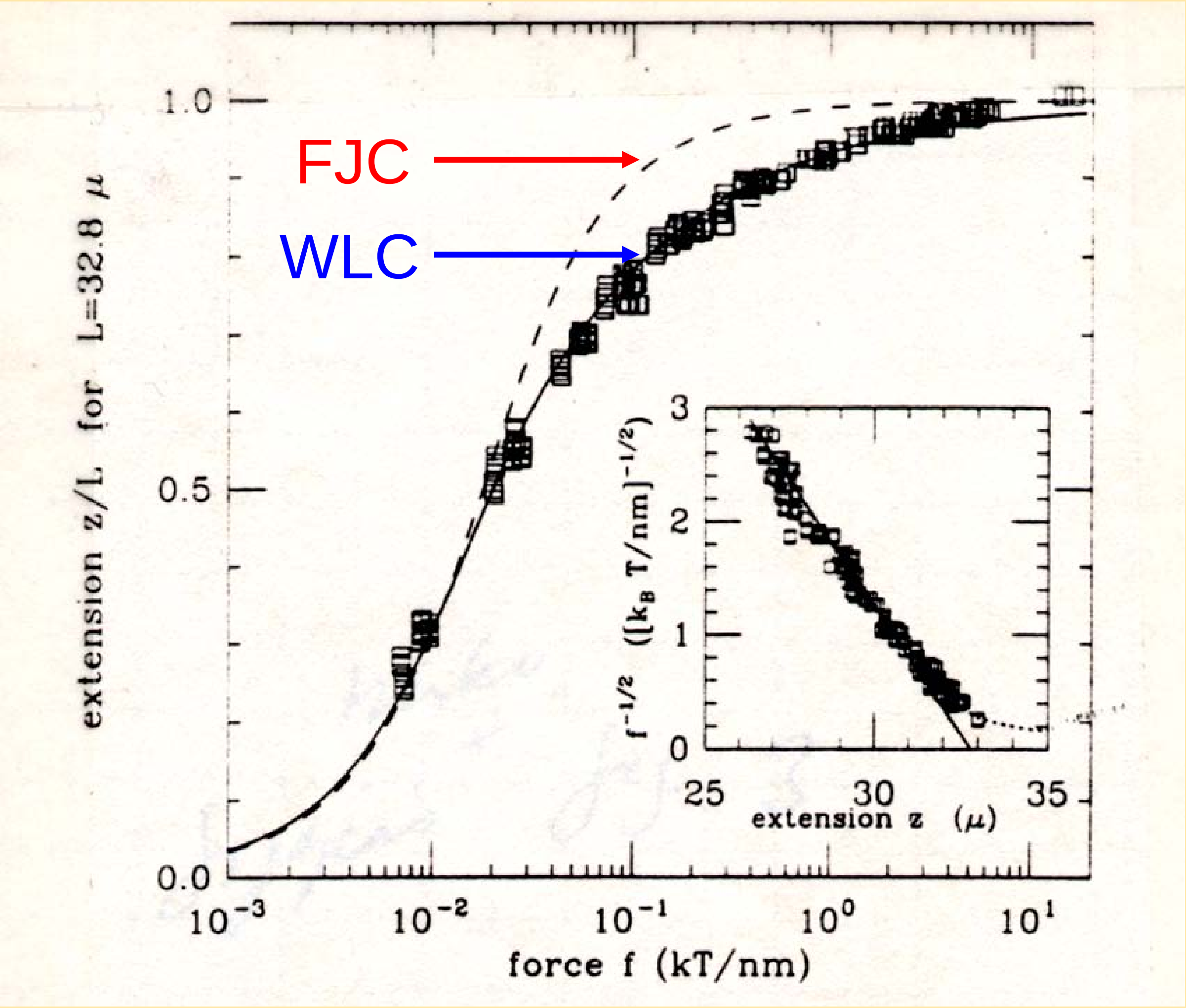
This interpolation formula is an excellent approximation to the exact solution throughout most of the range of forces investigated by the magnetic bead experiment.

DNA Elasticity: Experimental Design



Smith et al, Science (1992)

Fitting the WLC Model to the Data



Asymptotic Behavior of the Interpolation Formula

At high forces and large extensions, the force-extension behavior of the WLC model is dominated by the quadratic term:

$$\frac{FP}{k_B T} \approx \frac{1}{4\left(1-\frac{z}{L}\right)^2} \qquad \text{For } FP > k_B T$$

and:

$$z = L - L\sqrt{\frac{k_B T}{4FP}}$$

Thus, at high forces, a plot of $1/\sqrt{F}$ vs. z, should be linear linear with an intercept in the abscisa of L and an intercept in the ordinate equal to $2\sqrt{P/k_B T}$

The $1/F^{1/2}$ *vs* Z Plot