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Dynamics of semiflexible polymer solutions in the *tightly entangled* concentration regime.

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Abstract

I corroborate an important experimental evidence reported by [Schuldt *et al., PRL,* 2016, **117**, 197801] revealing the incapability of the current theoretical framework to fully describe the dynamics of semiflexible polymer solutions in the *tightly-entangled* concentration regime. These results have been endorsed here by means of previously published, but overlooked, data by [Tassieri *et al., PRL,* 2008, **101**, 198301; Tassieri *et al., Biophysical J.,* 2008, **94**, 2170]. The ensemble of information provides a strong evidence that the scaling law of the plateau modulus as function of polymer's concentration and persistence length, i.e. $G^0 \propto c^\alpha L_p^\beta$, should have both the exponents positive; in contrast with all the existing predictions converging on similar values of $\alpha > 0$ and $\beta < 0$. Hence, the need of new theoretical models able to better interpret the contribution of the polymer bending rigidity to the viscoelastic properties of the polymer network.

1 Introduction

Sitting between two well established theoretical frameworks describing the dynamics of linear flexible polymers and rigid rods,[1–4] there exists a number of extrapolated models[5–14] aimed at describing the *yet not* fully understood dynamics of linear semiflexible polymers. Indeed, despite their importance to soft-matter physics, biology, and industrial processing, a basic analytical model aimed at predicting the viscoelastic properties of semiflexible polymer solutions has not yet been agreed upon. This is because solutions of semiflexible polymers have many regimes of viscoelastic behaviour, depending on the polymers' degree of rigidity (commonly described in terms of persistence length L_p), on their contour length L and on the concentration c.

In spite of that, in the case of concentrated solutions (i.e., in the *tightly-entangled* regime), where the geometrical mesh size ξ_m of the polymer network is much smaller than L_p, all theoretical models converge to either identical or similar scaling law predictions of the solution's plateau modulus G^0 as function of both c and L_p. In particular, while most theories[10–13] predict a scaling law of $G^0 \propto c^{7/5} L_p^{-1/5}$, competing studies[8] propose $G^0 \propto c^{4/3} L_p^{-1/3}$. Ergo, all the existing models agree on (i) very similar concentration-dependence of the plateau modulus, with fractional exponents so close to each other (i.e., $G \propto c^{1.4}$ vs. $G \propto c^{1.3\bar{3}}$) to be indistinguishable experimentally,[8,15,16] and (ii) an inverse proportionality of G^0 to the polymer's persistence length, with significantly different fractional exponents (i.e., $G \propto L_p^{-0.2}$ vs. $G \propto L_p^{-0.3\bar{3}}$) to allow critical studies[16] on which of the above predictions (*if either*) better describes the viscoelastic behaviour of semiflexible polymer solutions in the *tightly-entangled* concentration regime.

Interestingly, thanks to new experimental findings reported by Schuldt *et al.*,[17] here augmented by experimental data previously published by Tassieri *et al.*,[16,18] it is actually possible to assert that *none* of the above mentioned scaling laws is actually suitable to fully describe the viscoelastic properties of concentrated solutions of semiflexible polymers. Indeed, when these data[16–18] are drawn together, they provide a strong evidence that the

scaling law of the plateau modulus as function of c and L_p, i.e. $G^0 \propto c^\alpha L_p^\beta$, must have both the exponents positive; in contrast with all the current theoretical predictions. In particular, while nothing new can be said on α (i.e., in *tightly-entangled* regime it remains undetermined on whether it is 7/5 or 4/3), it is actually possible to assert that β is a positive number, possibly ranging between 1 and 5, depending on yet unknown factors; however, certainly different from the two negative values (i.e., $G \propto L_p^{-0.2}$ or $G \propto L_p^{-0.3\bar{3}}$) proposed by current theories. In addition, upon entering the *tightly-entangled* regime, some semiflexible polymer solutions show a scaling behaviour of $G^0 \propto c^2 L_p$, which is here *semi-empirically* obtained by combining a general expression of the plateau modulus, valid for entangled solutions of semiflexible polymers,[12] with a relationship describing the topological confinement of semiflexible polymers in the *tightly-entangled* regime.[8]

Hence, the need of new models that better interpret the contribution of the polymer bending rigidity to the viscoelastic properties of the polymer network and the concentration-dependence of the characteristic length scales of this latter, as highlighted in this paper.

2 Theoretical background

2.1 The persistence length L_p

To a first approximation a polymer with aspect ratio $p \gg 1$ (defined as the ratio between the polymer contour length L and its diameter d) can be described as a homogeneous elastic filament with circular cross-section and mechanical properties proportional to its Young's modulus (E).[19] Once the latter is known, the elastic stiffness for specific deformations, such as stretch, bending and twist can be calculated. For sufficiently small amplitudes of excitation, additional simplifying assumptions can be made; e.g., if a constant cross-section is assumed during bending, then all the different types of deformations can be considered as independent from each other. A further hypothesis that can be made is the inextensibility constraint of the filament. These assumptions form the basis of the worm-like chain (WLC)

model. Moreover, since the energy per unit length required to change the length of such a filament by a specified fractional strain is proportional to Ed^2 and the energy per length required to bend it into an arc of radius R is of order of Ed^4/R^2, the ratio between the arc bending energy and the strain energy is of order of $(d/R)^2$; therefore, the adoption of the WLC model is a valid option as long as $d/R << 1$.

The smallest excitations to which a polymer chain is subject are caused by thermal energy. These represent an important feature for microscopic structures of semiflexible polymer solutions. The conventional measure of the degree of thermal flexing of a polymer is the persistence length L_p.[20] This is defined by the correlation function Equation 1, as the characteristic distance along the polymer chain over which the direction of unit vector $\vec{u}(s)$ drawn tangent to the polymer contour become uncorrelated:

$$\langle \vec{u}(s) \bullet \vec{u}(s') \rangle = e^{-|s-s'|/L_p}, \tag{1}$$

where the brackets $\langle \cdots \rangle$ denote the average over thermal fluctuations, and s is the distance along the chain contour, measured from an arbitrary point s' (Figure 1). The persistence length is proportional to the filament rigidity and it is inversely proportional to the temperature:

$$L_p = \frac{EI}{k_B T}, \tag{2}$$

where k_B is the Boltzmann's constant, T is the absolute temperature and I is the moment of inertia of the filament cross-section, which is different for solid (I_s) or annular (I_a) shapes:

$$I_s = \frac{\pi}{64}d^4 \quad or \quad I_a = \frac{\pi}{64}(d_2^4 - d_1^4), \tag{3}$$

where d_1 and d_2 are the inner and the outer diameters. For a thin annular tube of thickness ϵ, where $d = d_1 \simeq d_2$ and $d_2 = d_1 + \epsilon$, one obtains $I \cong (\pi d^3 \epsilon)/21$. The product EI is also defined as the "flexural rigidity" or "bending modulus" (κ).
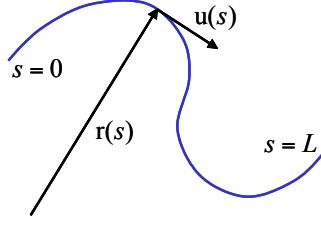
Figure 1: Schematic representation of a linear semiflexible polymer.

Under the WLC model (for which $L \gg d$), the ration $L/L_p \sim 1$ represents a crossover between two opposing asymptotic *ideal* configurations towards which semiflexible polymers may tend to depending on the relative value of L and L_p: (i) *flexible phantom* chain of Kuhn length[21] $b = 2L_p$ (for $L \gg L_p$) and (ii) stiff *rod* (for $L \ll L_p$). This can be further elucidated by considering the polymer's mean-square end-to-end distance $\langle R^2 \rangle$, which for a worm-like chain can be expressed in terms of L and L_p, with clear asymptotic values:[22]

$$\langle R^2 \rangle = 2L_p^2 \left(\frac{L}{L_p} - 1 + e^{-L/L_p} \right) \Rightarrow \begin{cases} \langle R^2 \rangle \cong 2LL_p \ \ (L \gg L_p) \\ \\ \langle R^2 \rangle \cong L^2 \ \ (L \ll L_p) \end{cases} \tag{4}$$

where the brackets $\langle \cdots \rangle$ denote an average over all possible states of the system (obtained either by considering many chains or numerous different conformations of the same chain). It follows that, in the first asymptotic case, the unit tangent vector will change direction several times along the polymer contour length and the chain configuration will look like a "*random coil*"; whereas, in the second asymptotic case, the unit tangent vector will be almost invariant and the polymer will look like a "*rod*".

2.2 Biological models of semiflexible polymer

Synthetic (*flexible*) linear polymer melts and their blends[23–25] have served the field of rheology as ideal models for the development of constitutive equations (e.g.,[26–31]) aimed at predicting the rheological behaviour of complex materials (e.g.,[32–34]). Similarly, biological systems such as protein filaments,[35,36] self-assembling peptides[37,38] and viruses,[39,40] have been explored by

biophysicists as potential models[41] of either semiflexible or rod-like polymers, with a dual aim of gathering new insights on their dynamics during biological processes. For instance, *in vitro* reconstituted actin filament has served for many years as biological model of linear semiflexible polymers and plays a key role in many biological processes, including cell motility, migration and invasion.

Below I summarise the key features of three biological models of semiflexible polymers that are relevant to this work.

Actin filament. At low ionic strength, actin exists in the monomeric (globular) G-actin form. This is roughly spherical with a diameter of about 5 nm. When the ionic strength of a G-actin solution is increased to a physiological value (0.1 M), G-actin self-associates to form the backbone of the thin filament, actin filament (F-actin), which can be viewed as either a two-stranded long-pitch ($\sim$ 37 nm) helical structure or a single short-pitch ($\sim$ 5.9 nm) helical structure, which is related to the size of the monomeric G-actin. F-actin is a semiflexible polymer characterised by an average contour length of $\sim$ 15 μm,[15,42] a persistence length ranging from $\sim$ 2 μm to $\sim$ 30 μm (depending on the buffer used)[43,44] and a diameter of $\sim$ 9 nm.[18,43] However, despite its ideal structure, *in vitro* reconstituted actin filaments come with devious issues that are difficult to deal with; these include, to mention but a few, (i) the unidirectional "treadmilling" of monomers from one end toward the other, (ii) contamination by either cross-linking or capping proteins and (iii) buffer pH and salinity.

Intact cardiac thin filament. An alternative to F-actin is represented by the (cardiac) thin filament (CTF), which is made of three constituents: F-actin, Tropomyosin (Tm) and Troponin (Tn). Tm is an extended molecule that is $\sim$ 42nm long formed as a homodimer or heterodimer of two α-helical chains arranged as a coiled coil. Stability of the coiled coil is produced by hydrophobic interactions between nonpolar side chains contributed by amino acids in each chain. Each chain is 284 residues long and spans 7 actin monomers on each strand of the F-actin filament. Neighbouring Tm overlap in a head-to-tail configuration with periodicity of 38.5nm along the thin filament.[45] Troponin is composed of three interacting

subunits each receiving its identifying letter from the first identified property: troponin C (TnC) binds Ca^{2+}, troponin I (TnI) binds to actin and inhibits the actomyosin ATPase in a Ca^{2+}-insensitive manner on a one-to-one basis with actin, and troponin T (TnT) links the Tn complex to Tm.[45] It has been shown[18] that, solutions of intact cardiac thin filaments have a bell-shaped distribution of contour lengths that contained a population of filaments of much greater length than the *in vivo* sarcomere size ($\sim 1\mu m$) due to a one-dimensional annealing process. Moreover, by combining these results with microrheology and dynamic light scattering measurements, it has been possible to reveal the rod-like nature of the CTF,[18] which has a persistence length of the order of a few microns (i.e., $L/L_p \simeq 1$) and concentration-dependence of the viscosity of $\eta \propto c^3$, in agreement with Doi and Edwards' model of rod-like polymers.[2,3]

DNA microtubes. A valuable alternative to both the above mentioned systems is given by "tiled-based DNA tubes".[17,46,47] These are engineered microtubes made up by n distinct and partially complementary DNA oligonucleotides, which hybridise to a half overlapping ring of n interconnected DNA helices. Depending on the chosen set of n strands, n-helix tubes (nHTs) with a uniform circumference can be obtained. Once formed in water solutions at a specific pH and ionic strength, they are extremely stable. The resulting filiform ($p \gg 1$) structures have a contour length distribution comparable to actin (up to $20\mu m$),[46] but with tunable (annular) cross-section and persistence length ranging from $\sim 9nm$ to $\sim 60nm$[47] and from $\sim 1\mu m$ to $\sim 20\mu m$,[17] respectively.

2.3 Measuring L_p via dynamic light scattering

The first fundamental contribution towards the understanding of semiflexible polymer dynamics by dynamic light scattering (DLS) measurements was thanks to Farge and Maggs.[48] They found that, with the following length scale conditions satisfied: $qL_p >> 1$, $q^{-1} < \xi_m$ and $L_p^{1/3}q^{-2/3} < \xi_m$ (where q is the wave vector), the dynamic structure factor follows a

universal scaling law:

$$g^{(1)}(q,t) \propto exp(-q^2 t^{3/4}). \tag{5}$$

Later on, the same result was achieved by other authors.[49–51] They all provided more explicit equations describing the dynamic structure factor behaviour in different time windows. These models[49–51] essentially differ from each other on the basic assumptions related to the microscopic hydrodynamic interactions and the inextensibility constraint of the polymer chain. Although these models[49,50] seem to approximate very well the experimental data,[49,52] only Kroy and Frey[50] provided, in the limit of a *"weakly bending rod"* ($L/L_p << 1$) and with the following length scale conditions satisfied $a_h << q^{-1} << L_p \cong L$ (where a_h is the hydrodynamic lateral diameter of the polymer chain) for sufficiently long time $t >> \gamma_q^{-1}(qL_p)^{-3/4}$, a *simple* analytical expression for the time decay of the dynamic structure factor in semidilute solution ($q\xi_m >> 1$):

$$g^{(1)}(q,t) = g^{(1)}(q,0)exp\left[-\frac{\Gamma(1/4)}{3\pi}(\gamma_q t)^{3/4}\right], \tag{6}$$

where γ_q is the decay rate defined as

$$\gamma_q = \frac{k_B T q^{8/3}}{4\pi\eta_s L_p^{1/3}}\left[C - ln(qa_h)\right], \tag{7}$$

η_s is the solvent viscosity and C is a constant (i.e. $5/6$ in Eq.(3.4) of Ref.[50]) that can be evaluated by laborious calculations describing the microscopic hydrodynamic interactions.

In addition, based on the same assumption as before, Kroy and Frey[50] provided the expression of the dynamic structure factor in the limit of $t \to 0$:

$$g^{(1)}(q,t) = g^{(1)}(q,0)exp(-\gamma^0 t), \tag{8}$$

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where γ^0 is the initial decay rate defined as

$$\gamma^0 = \frac{k_B T q^3}{6\pi^2 \eta_s} \left[C - ln \left(q a_h \right) \right].$$

(9)

Equations 7 and 9 (with $C = 5/6$) have been successfully used in literature[16,18,38] to *estimate* two characteristic polymer lengths: the microscopic lateral diameter d ($\cong a_h$) and the persistence length L_p. In particular, it has been shown,[16,18,38] by direct comparison with transmission electron microscopy (TEM) image analysis results, that Equation 9 provides a good estimation of the lateral diameter with an uncertainty of order of a few percent depending on how well the investigated system satisfies the "*weakly bending rod*" model conditions.

It is important at this point to specify that, as correctly pointed out by Nyrkova and Semenov,[53] the constant C in Equation 9 can be replaced by anything ~ 1 and one can still obtain a reasonable agreement between DLS estimations and direct (e.g., TEM) measurements of the microscopic lateral diameter. This uncertainty is mainly due to both the weak logarithm dependency of γ^0 on $q a_h$ and the small range of experimentaly accessible q values.[54]

2.4 Linear viscoelasticity of semiflexible polymer solutions

The first important contribution toward understanding the viscoelastic properties of entangled polymer solutions was provided by Edwards,[55] who introduced the "*tube*" model as a mean-field description of the topological confinement exerted on a given chain by the surrounding chains. Next, de Gennes[1] solved a fundamental aspect of the entangled dynamics by describing the diffusion of a chain along its own length, a process that he named "*reptation*". Finally, Doi and Edwards, by combining the idea of the tube with reptation, developed two full theories of the entangled state for two extreme cases: completely flexible[2] and rigid-rod[3] polymers.

All current models describing the viscoelastic properties of semiflexible polymer solutions

are extrapolations of the Doi and Edwards' models.[2,3] Amongst the others, Morse's model[5–8] is the only one that interpolates smoothly between the limit of rod-like polymers ($L << L_p$) and coil-like polymers ($L >> L_p$), providing a rather detailed description of the viscoelastic properties of semiflexible polymer solutions. With respect to the WLC model, Morse identifies all the possible regimes of polymer concentration and chain length expected for solutions of semiflexible polymers. He then characterises the regimes of dynamical behaviour of an entangled solution in terms of the relative magnitudes of four length scales: L, L_p, the entanglement length L_e (defined as the contour distance between collisions of the polymer with the walls of the tube and thus depending upon both L_p and ξ_m), and the chain diameter d.

Morse calculated a general expression for the stress tensor of a solution of wormlike chains that may be applied to any concentration regime. In particular, the stress tensor of a solution subjected to a macroscopic fluid velocity $\mathbf{v}$ with a spatially homogeneous rate-of-deformation tensor $\Gamma(t) = (\nabla \mathbf{v})^T$ is given in the Kramers-Kirkwood theory[56,57] by a sum: $\sigma(t) = \eta_s(\Gamma + \Gamma^T) + \sigma_p$; where σ_p is the polymer stress and η_s is the solvent viscosity. The polymer stress tensor may be divided into two contributions: $\sigma_p = \sigma_{intra} + \sigma_{inter}$, arising from forces associated with, respectively, intramolecular and intermolecular (e.g., steric or Van der Waals) interactions. The *intermolecular stress* contribution is negligible over all the frequency ranges for concentrations up to the nematic phase for both the limit cases of coil-like and rod-like polymers. The *intramolecular stress* contribution, instead, is expected to play a much bigger role in determining the viscoelastic properties of systems of long semiflexible chains. It can be expressed as a sum $\sigma_{intra} = \sigma_{curve} + \sigma_{orient} + \sigma_{tens} - ck_BT\delta$, where the stress contributions (see Figure 2) are defined physically as it follows: (i) σ_{curve} is the curvature contribution arising from forces that oppose transverse deformation or rotation of chain segments, (ii) σ_{orient} is the orientational contribution that reduces in the appropriate limit to the Brownian stress of a rigid-rod solution. (iii) σ_{tens} is the tension contribution arising from tangential forces that resist stretching or compression of the chain; and (iv) $-ck_BT\delta$ is the ideal solution osmotic pressure arising from the translational entropy of the
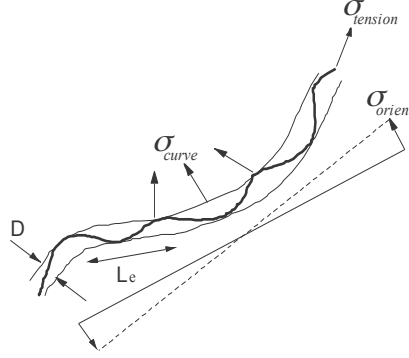
Figure 2: Schematic representation of stress tensor contributions.

molecule as a whole.

In order to calculate the linear viscoelastic properties of a fluid, Morse calculated the relaxation of σ_{intra} in a system that is subjected to an infinitesimal step strain $\delta\epsilon$ at time $t = 0$. The shear relaxation modulus $G(t)$ is defined by expressing the resulting stress as a product $\sigma(t) = G(t)(\delta\epsilon + \delta\epsilon^T)$. The complex modulus $G^*(\omega)$ is given by the Fourier transform of the time derivative of $G(t)$.[58,59] It is thus possible to express $G^*(\omega)$ as sums of contributions arising from the three different stresses: $G^*(\omega) = G^*_{curve}(\omega) + G^*_{orient}(\omega) + G^*_{tens}(\omega)$. Let us focus on the dynamics of highly entangled isotropic solutions. In particular, in the *tightly-entangled regime* (where both ξ_m and L_e are expected to be much less than L_p) and at time scales much shorter than the system's longest relaxation time (i.e., $\forall t < t_{rep}$, where t_{rep} is the "*reptation*" time), the orientational contribution $G_{orient}(t)$ is expected to make a negligible contribution to $G(t)$ and the latter may be approximated as the sum of a large but rapidly decaying tension contribution $G_{tens}(t)$ and a much smaller but more slowly decaying curvature contribution $G_{curve}(t)$; hence, the possible existence of two plateau moduli (at high and low frequencies, respectively), as also predicted by Maggs.[60] In particular, at relatively low frequencies (i.e., for $t^{-1} \gtrsim t^{-1}_{rep}$), Morse developed two analytical approximations[8] describing the confinement forces acting on a randomly chosen test chain embedded in a "*thicket*" of uncrossable chains:[8] (I) the binary collision approximation (BCA) and (II) effective medium

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approximation (EMA), which yield the following scaling laws:

$$BCA \begin{cases} R_e \propto c^{-3/5} L_p^{-1/5}, \\ L_e \propto c^{-2/5} L_p^{1/5}, \\ G^0 \propto c^{7/5} L_p^{-1/5}, \end{cases} \tag{10}$$

and

$$EMA \begin{cases} R_e \propto c^{-1/2}, \\ L_e \propto c^{-1/3} L_p^{1/3}, \\ G^0 \propto c^{4/3} L_p^{-1/3}, \end{cases} \tag{11}$$

where R_e is the radius of the tube in which the test chain is embedded ($R_e = D/2$, see Figure 2). Notice that, both the above approximations are based on the assumption that R_e and L_e obey to the following power law relationship:

$$L_e \propto R_e^{2/3} L_p^{1/3}, \tag{12}$$

which was derived by Odijk[9] in the study of the dynamics of a single worm-like chain polymer in a cylindrical pore with solid walls. Interestingly, the scaling relation resulting from the BCA had previously been obtained by several others authors,[10–13] but Morse has also estimated the prefactors.[8] So, prior to the introduction of the EMA, there was broad agreement regarding the scaling law of the solution's plateau modulus G^0 in the *tightly-entangled* concentration regime.

At relatively high frequencies (i.e., for times t such that $t_{rep}^{-1} << t^{-1} \lesssim \tau_e^{-1}$, where τ_e is the relaxation time of an undulation mode of wavelength L_e), Morse's model retrieves the scaling low predictions derived by Mackintosh *et al.*,[12] who focused exclusively on σ_{tens} contribution to the stress, but did not allow for any relaxation of the tension; an assumption

that is more applicable to cross-linked gels:

$$G^0 \propto c^{11/5} L_p^{7/5} \tag{13}$$

Notably, this is the only theoretical prediction of the plateau modulus with both positive exponents, in line with the new experimental findings reported by Schuldt et al.[17] and those previously published by Tassieri et al.;[16,18] as discussed below.

3   Discussion

In order to discriminate which of the above equations (*if any*) better represents the dynamics of semiflexible polymers in the *tightly-entangled* concentration regime, independent measurements of the solutions' plateau modulus and polymer's persistence length would be necessary, as pioneered by Tassieri et al.[16] while studying the dynamics of F-actin solutions. Driven by the same aim, Schuldt et al.[17] have recently revealed new insights on the dynamics of semiflexible polymer solutions; i.e., they have provided a strong experimental evidence indicating the existence of a direct proportionality of the plateau modulus to the polymer's persistence length $G^0 \propto L_p$.

In what follows, I wish to endorse and speculate on Schuldt et al.'s findings.[17] In particular, notwithstanding the validity of the results reported by Schuldt et al.,[17] here I argue that they cannot be considered conclusive and the dynamics of semiflexible polymers continue to be unresolved. In support of my opinion, I report several independent measurements of both the plateau modulus and the persistence length (see Figure 4) for 'three' different biological models of semiflexible polymers, in the *tightly-entangled* concentration regime. Specifically, these systems are solutions of *in vitro* reconstituted F-actin,[16] DNA *n*HT microtubules[17] and intact cardiac thin filaments (with and without addition of calcium).[18]

3.1 Concentration-dependence of the plateau modulus

Upon entering the tightly-entangled regime, Morse's model predicts a change in the concentration-dependence of the plateau modulus from $G^0 \propto c^{2.3}$ in the *loosely-entangled* regime to $G^0 \propto c^{1.4}$ for $c > c^{**}$, where c^{**} is the crossover concentration between the two regimes. This thesis has been supported so far only by simulations[61] and a single experimental evidence presented by Tassieri *et al.*,[16] leaving unaddressed Morse's dilemma that "*experiments on isotropic solutions of F-actin span concentrations near c^{**} for which the predicted asymptotic scaling of the plateau modulus $G^0 \propto c^{7/5}$ is not yet valid*".[61]

Morse's uncertainty is here elucidated by the data shown in Figure 3, in which I report measurements of the plateau modulus *versus* concentration for *nine* different systems of semiflexible polymers spanning over two decades of concentration. In particular, in Figure 3(Top) are reported the data presented in the original works,[16,17] which were obtained from either micro- or bulk-rheology measurements, respectively; whereas, in Figure 3(Bottom) the same data as above have been rescaled vertically by an arbitrary factor to allow their superimposition at high concentrations (i.e., for $c \gtrsim 3\mu M$), demonstrating their compliance to the scaling predictions for the *tightly-entangled* regime. From Figure 3(Bottom) it is possible to identify a crossover concentration at a value of $c \cong 3\mu M$, which may actually turn to be veracious due to the similar characteristic lengths (i.e., Lp, L and d) of the explored systems.[17] Nevertheless, from Figure 3(Bottom) it is clear that (i) for concentrations $c \gtrsim 3\mu M$, data can be equally well interpreted by either of the two competing scaling laws $G^0 \propto c^{7/5}$ or $G^0 \propto c^{4/3}$, leaving the dispute still open; (ii) for concentrations $c \lesssim 3\mu M$, data can be fitted by any power law $G^0 \propto c^\alpha$ having exponent α ranging between 4/3 and 2.3 (hence, Morse's dilemma). Interestingly, despite the so coveted asymptotic scaling of the plateau modulus ($G^0 \propto c^{7/5}$) is now clearly defined, it is worth noting that at least two systems in Figure 3(Bottom) (i.e., F-actin 2 and 14HT) show adherence to a power law of $G^0 \propto c^2$ over an extended range of concentrations (i.e., $0.2 \lesssim c \lesssim 10\mu M$). This peculiar feature is discussed in the following paragraph.
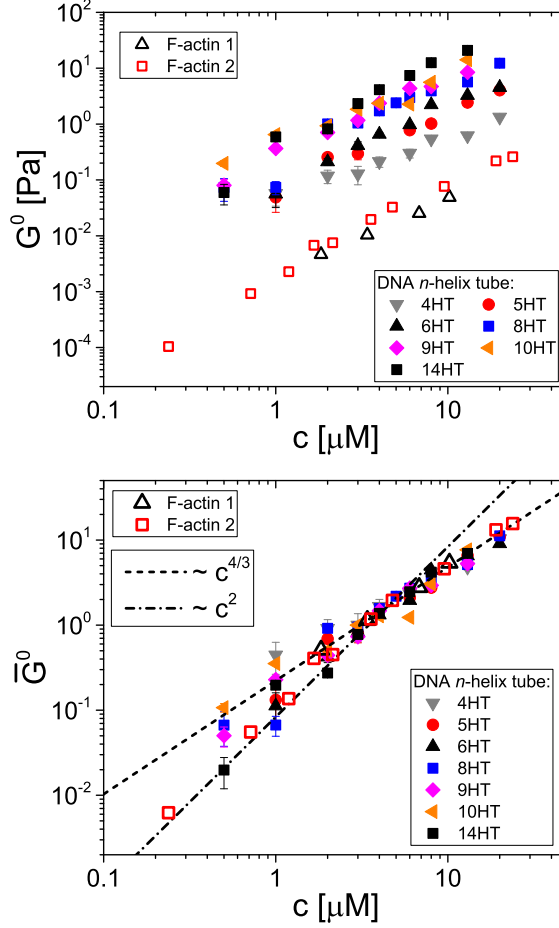
Figure 3: The plateau modulus *vs.* concentration for different biological models of semiflexible polymers. (Top) Data reproduced from References.[16,17] (Bottom) Same data as above, but for each system the modulus has been rescaled ($\bar{G}^0$) by an arbitrary factor to allow data superimposition at high concentrations (i.e., for $c \gtrsim 3\mu M$). Lines are guides for the power laws.

3.2 Persistence length-dependence of the plateau modulus

In order to better discriminate between different scaling laws governing the relationship between the solution's plateau modulus and the polymer's persistence length, the original data of both G^0 and L_p (taken from References[16–18] and here reproduced in Figure 4(Top)) have been normalised by the first value of their own data set and presented in Figure 4(Bottom). From this, it is clear that experimental results fall between two power laws: (i) $G^0 \propto L_p$ and (ii) $G^0 \propto L_p^5$, leaving no doubts on the possible existence of an inverse proportionality of the plateau modulus to the persistence length; at odds with all the current theoretical

predictions leading to either the systems of Equations 10 and 11.

By contrast, Equation 13 is the only theoretical prediction that provides a persistence length-dependence of the plateau modulus with a positive exponent. It was derived by Mackintosh *et al.*[12] from a more general expression valid for entangled solutions of semiflexible polymers:

$$G^0 \propto \xi_m^{-2} L_p^2 L_e^{-3}, \tag{14}$$

based on the assumption that at high concentration (where $\xi_m \propto c^{-1/2}$ and $\xi_m << L_p$) the entanglement length could be expressed as $L_e \propto \kappa^{1/5} c^{-2/5}$, like in the system of Equations 10. Interestingly, it is possible to speculate that if instead one would substitute in Equation 14 the expression of the entanglement length $L_e \propto c^{-1/3} L_p^{1/3}$ derived by Morse for the effective medium approximation, one would obtain:

$$G^0 \propto c^2 L_p, \tag{15}$$

which provides a direct proportionality of the plateau modulus to the polymer's persistence length, in agreement with the experimental evidences presented by Schuldt *et al.*,[17] but with a stronger concentration-dependence of the plateau modulus than the one predicted by the existing models developed for such concentration regime (i.e., $G \propto c^2$ vs. $G \propto c^{1.4}$). However, in my *opinion*, it simply cannot be considered mere coincidence that both the systems F-actin 2 and 14HT show good adherence to a power law of $G^0 \propto c^2$ over an extended range of concentrations, i.e. $0.2 \lesssim c \lesssim 10 \mu M$ (see Figure 3(Bottom)), and that system 14HT, within the experimental error, practically fulfils Equation 15.

Hence, the need of new models that better account the contribution of the concentration-dependent characteristic length scales (e.g., L_e) of the polymer network to its mechanical properties. In this regard, it is worth mentioning that microscopy studies[62–64] on the tube width fluctuations in F-actin solutions have revealed the inappropriateness of theoretical approaches to employ a homogeneity (or mean-field) assumption, treating the tube radius as
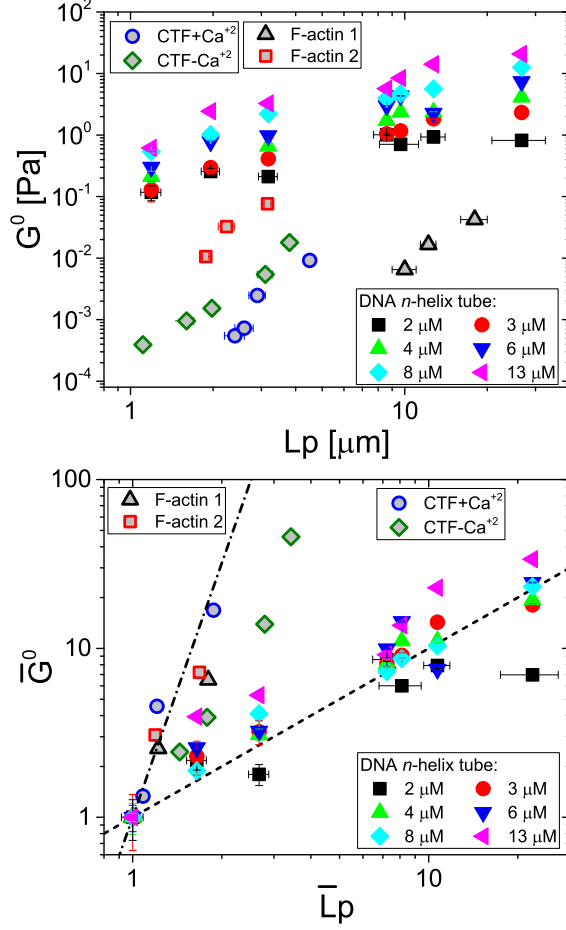
Figure 4: The plateau modulus *vs.* persistence length for different biological models of semiflexible polymers. (Top) Data reproduced from References.[16–18] (Bottom) Same data as above, but for each system the original data of both G^0 and L_p have been normalised by the first value of their own data set, respectively. Lines are guides for the power laws: $G^0 \propto L_p$ and $G^0 \propto L_p^5$, respectively.

a constant; while experiments[13,65–68] and simulations[69] indicate substantial heterogeneities.

The common outcome of these studies indicates that both the concentration-dependent distributions of the tube radius $P(R_e)$ and of the entanglement length $P(L_e)$ are non-symmetric Gaussian functions. Therefore, it is possible to further speculate that the adoption of these skewed leptokurtic distributions may represent an alternative pathway for the development of theoretical models capable to fully describe the viscoelastic properties of semiflexible polymer solutions.

4 Conclusions

After many years, competing theories aimed at describing the dynamics of semiflexible polymer solutions in the *tightly-entangled* concentration regime have converged to similar scaling laws of the plateau modulus as function of polymer's concentration and persistence length, i.e.: $G^0 \propto c^\alpha L_p^\beta$, with both α and β *yet undetermined* on either the values $7/5$ or $4/3$ and $-1/5$ or $-1/3$, respectively.

In this article, I corroborate a strong experimental evidence reported by Schuldt *et al.*[17] asserting that *none* of the above scaling laws are actually able to fully describe the dynamics of semiflexible polymer solutions in the *tightly-entangled* regime. This is achieved by drawing together new[17] and previously published[16,18] independent measurements of both the solutions' plateau modulus and the polymers' persistence length for *ten* different biological models of semiflexible polymers. The collected data provide a strong evidence that β is actually a positive number ranging between 1 and 5 *circa*, depending on *yet unknown* factors; but certainly different from the predicted negative values. On the other hand, α remains undetermined on whether it is $7/5$ or $4/3$ in the *tightly-entangled* concentration regime, and it spans between ~ 1.4 and ~ 2 in the *loosely-entangled* regime.

To conclude, the ensemble of independent experimental data presented in this paper must serve as a springboard for theoreticians to revise current models and to develop new comprehensive theories able to predict the dynamics of entangled semiflexible polymer solutions. These are indeed of crucial importance to many biological processes, including tumor cell motility, migration and invasion.

Acknowledgement

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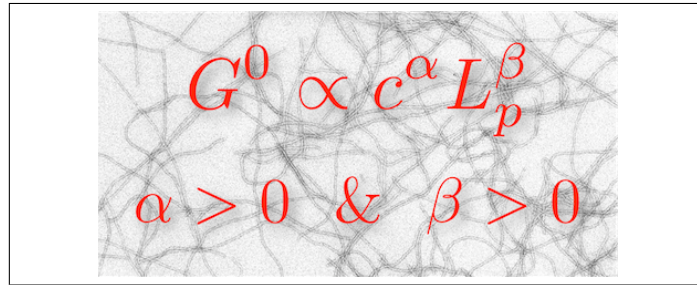
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Graphical TOC Entry

$$G^0 \propto c^\alpha L_p^\beta$$

$$\alpha > 0 \quad \& \quad \beta > 0$$