Polymer 49 (2008) 3353–3361

Contents lists available at ScienceDirect

Polymer

journal homepage: www.elsevier.com/locate/polymer

Feature Article

Force spectroscopy of polymers: Studying on intramolecular and intermolecular interactions in single molecular level

Xi Zhang*, Chuanjun Liu, Zhiqiang Wang

Key Laboratory of Organic Optoelectronics and Molecular Engineering, Department of Chemistry, Tsinghua University, Beijing 100084, PR China

ARTICLE INFO

Article history:
Received 14 February 2008
Received in revised form 23 April 2008
Accepted 30 April 2008
Available online 3 May 2008

Keywords:
Single molecule force spectroscopy
Single polymer chain
Atomic force microscopy

ABSTRACT

Single molecule force spectroscopy (SMFS) is a powerful means in studying the intramolecular and intermolecular interactions in polymer and supramolecular systems, thus opening new horizons for nanomechanic chemistry. This feature article concerns mainly how to employ AFM-based SMFS to obtain some of the important information which is not available by conventional characterizing methods, including single chain mechanics, the interfacial conformation and adhesive energy of polymers, interaction between macromolecules and small molecules, and the direct measurement of intermolecular forces. In addition, some of the recent advances in using the SMFS to bottom–up assembly of functional nanostructure are also discussed.

1. Introduction

Nanoscience and nanotechnology have been driven, to large extent, by key enabling approaches and platforms, including characterization techniques such as atomic force microscopy (AFM) [1,2], optical tweezers, force clamps, magnetic beads, glass microneedles, biomembrane force probe single molecule fluorescence, laser scanning confocal microscopy, and near field optics. These new techniques allow us to image and manipulate single polymers, as well as to study their dynamics at the single molecule level, hence promoting significantly the progress of traditional polymer science and opening new horizons for the future [3–7].

Although AFM has been widely used to image the surface structures of micro-/nano-scale, it is also a force sensor with high sensitivity [5,6]. The AFM based single molecule force spectroscopy (SMFS) can be employed to measure the minute force in molecular scale as well as to record the exceptionally small distance simultaneously. The curves of force signals versus extension can be obtained, thus providing valuable information about the intramolecular and intermolecular interactions of polymer systems in single chain level [3–5].

Besides AFM-based SMFS, there are also some other nanotechniques, such as optical tweezers, glass microneedles, biomembrane force probe, surface force apparatus, which can be used to measure minute force. These methods can provide force signals with different timescale and sensitivity [4]. Among them, AFM-based SMFS becomes popularized in the study of single polymer chains, because it is relatively easy to be operated. By forming a molecular bridge between AFM tip and substrate, the AFM-based SMFS can be used to study the intramolecular and intermolecular interactions not only in polymer systems but also in biomacromolecules and supramolecular systems.

This feature article attempts to provide an overview about what can we learn from the force spectroscopy of polymers on the basis of our ten year endeavor in this line of researches. It will discuss how to employ AFM-based SMFS to obtain some of the important information which is not available by conventional characterization methods, including mechanical properties of single polymer chain, the interfacial conformation and adhesive energy of polymers, interaction between macromolecules and small molecules, and the direct measurement of intermolecular forces. For experimental details, by one can refer to some review articles which have described the basic principle of AFM-based SMFS in detail [4,5].

2. Single chain mechanics

The mechanical property of polymer materials is a classical topic in polymer science. However, it is very difficult to study at a single molecule level. The understanding of mechanical response of individual polymer chains may deepen our understanding of the relationship between the primary chemical composition and the final macroscopic properties of polymer materials. In addition, on the basis of single chain mechanics, a great deal of information about the force-induced conformational transition can be obtained, which is not available by other conventional methods. These data about conformational transition can reflect the intramolecular

---

* Corresponding author. Tel.: +86 10 62796283; fax: +86 10 62771149.
E-mail address: xi@mail.tsinghua.edu.cn (X. Zhang).

0032-3861/$ – see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.polymer.2008.04.056

interaction and reveal their dynamic molecular structures in the polymer systems.

2.1. Single chain mechanics of synthetic polymers

To study the single chain mechanics, a single polymer bridge needs to be established between the AFM tip and solid substrate. To make SMFS experiments feasible, we have deposited the water-soluble polymers onto glass substrate to form a very thin film and immersed it into a liquid cell with aqueous solution. Then, an AFM cantilever is lowered to the substrate by the movement of a piezo tube, whereupon some molecules can be adsorbed onto the AFM tip, forming a single polymer bridge between the tip and the substrate. During the separation of the cantilever and the substrate, the polymer chain is stretched, which results in the deflection of the cantilever. A deflection–extension curve can be recorded and converted into a force–extension curve.

Taking poly(dimethylacrylamide) (PDMA) and poly(diethylacrylamide) (PDEA) as examples, we have employed SMFS to investigate the influence of substitute groups on the polymer chain elasticity [8]. These two polymers share same backbone structure. The only difference lies in the side groups: PDMA is substituted by two methyl groups and PDEA by two ethyl groups. To compare the elasticity of PDMA and PDEA in water, the force–extension curves are normalized according to their contour length, as shown in Fig. 1. The force–extension curves show clearly that there is no difference in the low force region in the figure; however, the single PDEA chain is stiffer than a PDMA in the high force region. The modified freely jointed chain (M-FJC) model fitting makes the difference more obvious. The M-FJC model, which is based on the extended Langevin function shown in Eq. (1), treats a macromolecule as a chain of statistically independent segments [3,4]. The segments, which are freely jointed together, can be deformed under stress.

$$X(F) = \left[\coth\left(\frac{Fl_{\mathrm{k}}}{k_{\mathrm{B}}T}\right) - \left(\frac{k_{\mathrm{B}}T}{Fl_{\mathrm{k}}}\right)\right]\left(L_{\mathrm{contour}} + \frac{nF}{K_{\mathrm{segment}}}\right) \tag{1}$$

Herein, F is the external force applied upon an individual polymer chain being stretched, X represents the extension of the polymer chain (end-to-end distance; in SMFS experiments, X is a section thereof, span between the substrate and the AFM tip.), L_{contour} is the length of the extended polymer chain under full extension, n is the number of segments being stretched, k_{B} is the Boltzmann constant, and T is the temperature in Kelvin. The deformation of segments is characterized by the segment elasticity, K_{segment}. Kuhn length (l_{k}) is the length of the statistically independent segment. The Kuhn length and the segment elasticity represent the elasticity of an individual polymer chain. Fitting parameters allow a quantitative comparison. For PDMA, the Kuhn length is about 1.5 nm, and the segment elasticity is around 16,000 pN/nm; for PDEA, the Kuhn length is about 1.6 nm, and the segment elasticity is around 17,000 pN/nm. Both the Kuhn length and segment elasticity of PDEA are larger than those of PDMA, which means that a single PDEA chain is stiffer than that of PDMA.

Two organometallic polymers bearing ferrocene groups in the backbone, poly(ferrocenyldimethylsilane) and poly(ferrocenylmethylphenylsilane), are used to reveal their single chain mechanical properties in the normal and oxidized forms [9]. The two polymers have shown similar elasticity in normal forms, though bearing different side groups. However, after they are oxidized by anhydrous $FeCl_3$, the different elasticity due to the steric effect can be amplified with the increase of their stiffness, allowing for differentiating the effect of side groups on single chain elasticity in oxidized and natural forms.

Beyond the investigation on the traditional polymers, SMFS can also be used to reveal the elastic property of supramolecular polymers. Vancso et al. studied the elastic property of the supramolecular polymer, which is formed through self-complementary recognition quadruple hydrogen-bonded 2-ureido-4[1*H*]-pyrimidinone (UPy) [10,11]. To probe the single chain mechanical properties of UPy-based supramolecular polymer, SMFS measurements are carried out with tips and substrates that are both modified with the short chain UPy disulfide in the presence of bis-UPys in hexadecane. Bis-UPys form supramolecular polymers with an effective degree of polymerization of approximately 40 in solution, which makes it possible to probe the mechanical properties of supramolecular polymers like a conventional polymer chain. These results indicate that supramolecular polymers can indeed be investigated on the single molecule level, and at the same time, open the pathway to elucidate the properties of these dynamically aggregated polymers as a function of various external parameters on the molecular scale.

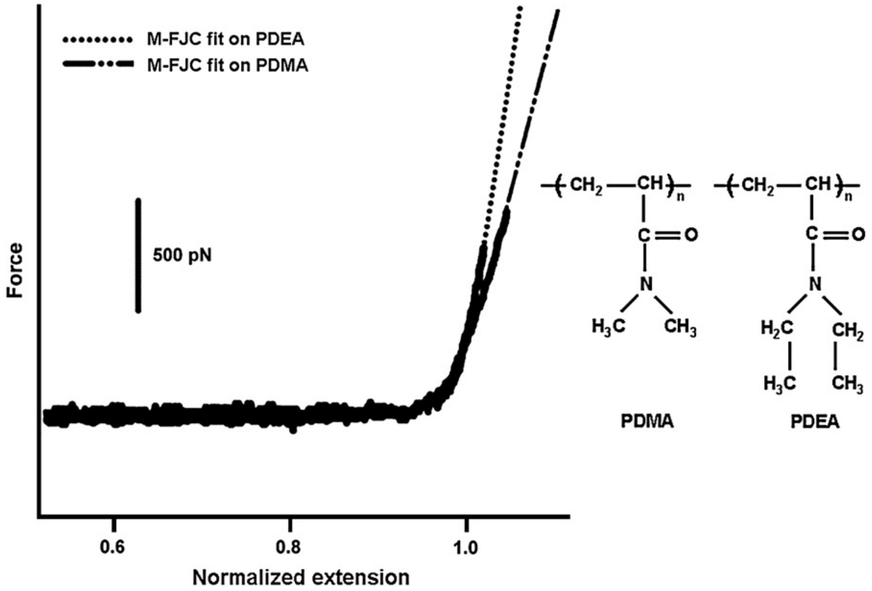


Fig. 1. Primary structures of PDMA and PDEA, and a comparison of the normalized curve of PDMA and PDEA in water. The two dotted lines are the extended FJC fitting on the experimental curves [8].

2.2. Force-induced conformational transitions of polysaccharides

The conformational transition of polysaccharides is very rich, but the information of the force-induced conformational transition can be only available with the advancement of the SMFS. One of the typical examples is to reveal the nanomechanical fingerprint of a α-(1,4)-linked glycan. Carboxymethyl amylose (CM-amylose) and carboxymethyl cellulose (CM-cellulose) are the two isomers of glycan. Their primary structures are almost the same except for the linkage of the pyranose ring. CM-amylose is linked by α-D-(1,4)-glucosidic bonds, while CM-cellulose is β-(1,4)-linked. Li et al. and Marszalek et al. have independently found that the small difference in the primary structure induces an obvious difference in their chain elongation properties [12,13]. For CM-amylose, a marked shoulder-like plateau on the force curves is observed at about 300 pN. The plateau shows a 0.08 nm elongation of each glucose residue and the estimated energy to induce this conformational transition is about 7.3 kT per glucose residue. For CM-cellulose, the force curves of the single chain elongation only show a sharp increase in force with the extension, and no plateau is present, as shown in Fig. 2. The reasons for the effect of different linkages on the single chain mechanics are because α-(1,4)-linked residues can easily adopt a chair–boat transition to achieve an extended conformation during the elongation; while each successive β-(1,4)-linked glucose residue can easily flip 180° to an extended conformation under external force. Hence the force-induced conformational transition is a unique property of CM-amylose.

As predicted, the force-induced conformational transition of the pyranose ring can be inhibited if an oxygen bridge is introduced over the sugar ring. We have comparatively studied the conformational transition using a series of carrageenans bearing oxygen bridges or not, in order to provide direct evidences to confirm this speculation [14]. The primary structures of λ-, κ- and ι-carrageenan are identical in one part of the repeating unit, the 1,3-linked β-D-pyranose ring; however, there is an oxygen bridge over the 1,4-linked α-D-galactopyranose ring in the other part of the repeating unit of κ- and ι-carrageenan. The typical shoulder-like plateau around 300 ± 50 pN appears in the force curves of λ-carrageenan. This shoulder-like plateau indicates that the force-induced conformational transition can still take place during the elongation of the λ-carrageenan single chain without oxygen bridges. But for κ- and ι-carrageenan, the shoulder-like plateaus disappear in the force curves, indicating that the conformational transition of the galactopyranose rings is inhibited efficiently due to the additional barrier of the oxygen bridges.

Marszalek et al. have examined the possibility of amylose to form inter-residue hydrogen bonds under various solvent conditions using SMFS [15]. When amylose forms strong inter-residue hydrogen bonds, which involve the interaction between O2 and H3′ atoms, it can significantly rigidify the polymer. Solvents with a low dielectric constant, such as DMSO, are favorable for the formation of such hydrogen bonds. To be reflected in the force–extension curve is the disappearance of the plateau feature. However, these inter-residue hydrogen bonds can be effectively eliminated in water; instead, amylose forms hydrogen bonding with water.

Besides the effect of solvent with different dielectric constants, we have investigated the change of force spectroscopy of single amylose chain in a crowded environment, which is provided by micelles. The plateau feature, which is characteristic of the force spectroscopy of amylose in water, disappears in the media of high concentration of cetyltrimethylammonium bromide or sodium dodecyl sulfate, as shown in Fig. 3, [16]. The Fourier Transform Infrared (FT-IR) spectroscopy and ^{1}H NMR data suggest that the addition of surfactants provides a more hydrophobic environment for amylose chain, thus facilitating the formation of inter-residue hydrogen bonding. The formation of hydrogen binding between O2 and H3′ can result in the elimination of the plateau feature in the force–extension curve of amylose, as discussed above. On the other way around, the change of the force spectroscopy of amylose may be used to indicate how crowded the solution environment is.

3. Interaction between polymers and small molecules

Since the SMFS experiment is carried out at a solid/liquid interface, it allows for studying the interaction between small molecules and polymers by simply changing the buffers. The comparison of the single chain elasticity before and after the addition of small molecules can indicate whether there exists interaction between the polymer and small molecules or not and how strong the interaction is.

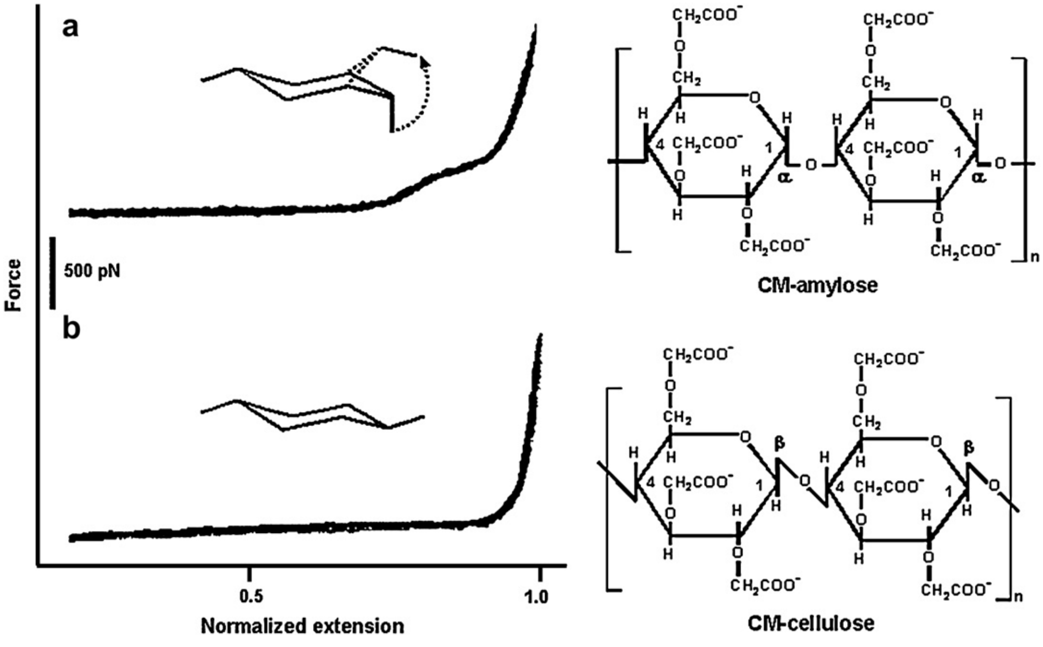


Fig. 2. Primary structures of CM-amylose and CM-cellulose and the comparison of normalized force curves of CM-amylose and CM-cellulose [13].

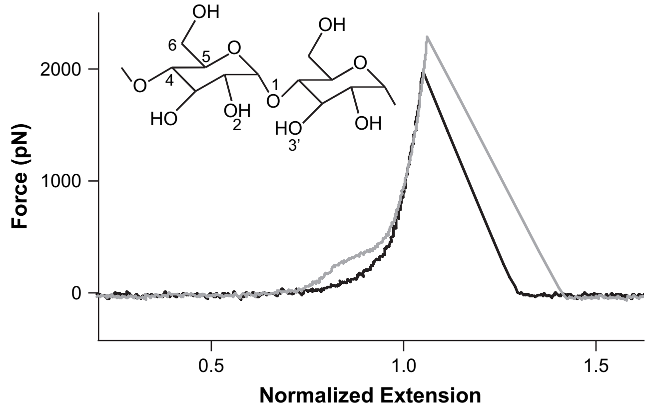


Fig. 3. Comparison of normalized force–extension curves of amylose in water (gray trace) and in 0.14 mol/L CTAB (black trace). The inset shows a building block of amylose [16].

There exists hydrogen bonding governed elasticity in many water-soluble polymers, such as poly(ethylene-glycol) (PEG) [17], and poly(vinyl alcohol) (PVA) [18,19]. The single chain elongation of PEG is like the behavior of an ideal entropic string in hexadecane, and can be described well with M-FJC model. However, in aqueous solution an evident deviation in the middle force region of the force curves is observed, indicating the deformation of a suprastructure within the polymer. By integrating the area between the force–extension curve and the model fitting curve, the binding free energy is calculated to be about 3.0 ± 0.3 kT per repeating unit [17]. Further analysis of the force curves based on the Markovian two-level systems, agreeing well with the *ab initio* calculations, identifies the non-planar suprastructural water bridges between the PEG chain and the water molecules.

To clarify the interaction between poly(*N*-vinyl-2-pyrrolidone) (PVPr) and iodine, we have studied the single chain nanomechanical properties of PVPr under different solution conditions [20]. The force–extension curve of PVPr in water is markedly deviated from that obtained in ethanol or tetrahydrofuran, suggesting different interactions between PVPr and the solvents. Moreover, we have comparatively studied the force signals of PVPr–I_2 and PVPr in aqueous solution of KI or KI_3, and found that only KI_3 influences the elastic property of PVPr dramatically. These experimental results indicate that there exists a specific interaction only between PVPr and KI_3, which is also supported by FT-IR data. By integration of the deviated area between the force curve and the M-FJC fitting curve, the energy needed to destroy the interactions between PVPr and KI_3 is estimated to be 3.6 kT per repeating unit.

4. Polymers at interface: interfacial conformation and adhesive energy

The polymer chain can adopt different interfacial conformations when adsorbed onto solid substrates. The interfacial conformation is supposed in the form of a ‘train’, ‘loop’, or ‘tail’, depending on the interaction between the polymer and substrate. However, it is very difficult to provide experimental evidences as to what kind of interfacial conformation exists and how strong the interaction is at single chain level. What present below is how to employ a few model polymer systems to reveal the polymer interfacial conformation and the adhesive energy using SMFS technique.

To make a convincing conclusion on force profile and interfacial conformation, carefully designed polymer systems are needed. Among them, a multi-segmented copolymer is a good choice. The structure and composition of such a copolymer are schematically shown in Fig. 4, in which short hydrophobic polystyrene (PS) segments are uniformly inserted into a linear poly(*N*-isopropylacrylamide) (PNIPAM) chain backbone [21,22]. As the segment copolymer chains adsorb onto the PS substrate in water, it is reasonable to expect that the adsorption of insoluble short PS segments onto the PS substrate results in many PNIPAM loops. The typical force curves of the segment copolymer on PS substrate exhibit a saw-tooth pattern, as shown in Fig. 4. The analysis of the distance between each two adjacent peaks in the force curves gives an average distance of about 114 nm, as shown in Fig. 5. This value is very similar to the average length of the ‘repeating unit’, i.e. one long PNIPAM segment plus one short PS segment, in the copolymer chain. These results suggest that the copolymer chain forms loops with a similar size on the PS substrate, indeed. In other words, the saw-tooth pattern corresponds to the detachment of multiple loops from the substrate. The most probable desorption force at a given stretching velocity of 4600 nm/s, which is obtained from the histogram of desorption force, is about 41 pN [22]. Such a distribution varies with the stretching velocity. The linear dependence of the most probable desorption force from the logarithm of the stretching velocity experimentally reveals that the adsorption and desorption of the PS segments on the PS substrate are a dynamic process.

Provided that a polymer chain forms a train-like conformation at an interface, the force should be similar for detaching each

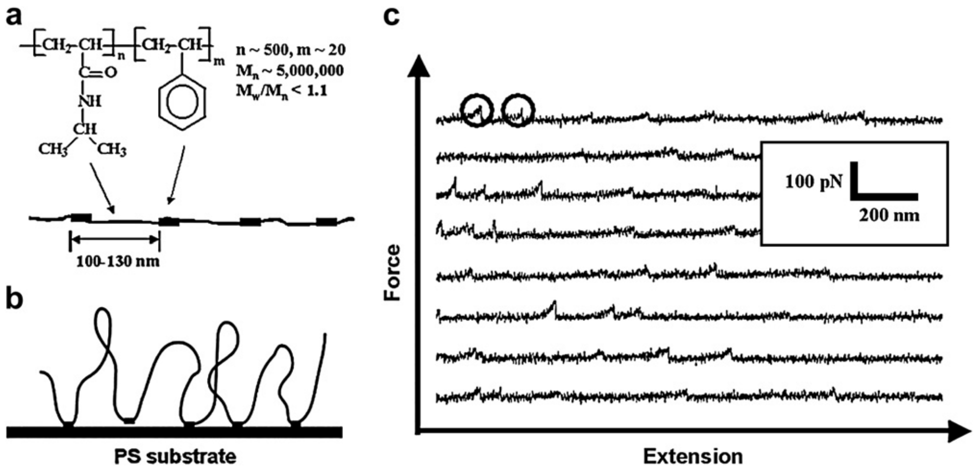


Fig. 4. Interfacial adsorption of a multi-segmented copolymer. (a) The linear segment PNIPAM-*seg*-PS; (b) a possible interfacial conformation of a PNIPAM-*seg*-PS chain on a hydrophobic PS substrate; (c) measured force curves of PNIPAM-*seg*-PS chains adsorbed on a hydrophobic polystyrene substrate in water [22].

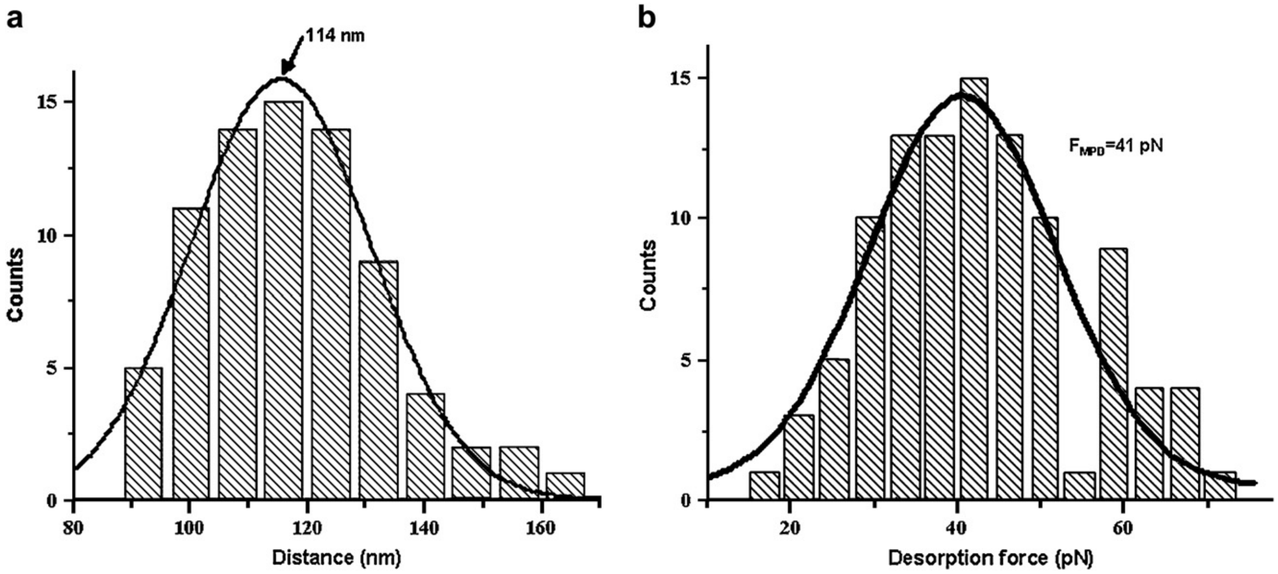


Fig. 5. (a) Statistics of the distance between two adjacent peaks in the measured force curves. (b) Distribution of the measured desorption force for linear PNIPAM-*seg*-PS chains adsorbed on hydrophobic polystyrene substrate, where the stretching velocity is kept at 4600 nm/s [22].

adsorption point during the polymer chain elongation. As a result, the force curves should show a characteristic plateau. Seitz and his coworkers have used chemisorption to immobilize polyvinylamine polymer chain to the AFM tip and studied its desorption force from a quartz substrate as a function of polymer's charge density and electrolyte concentration [23,24]. Their results indicate that, in addition to electrostatic interaction between polyvinylamine and negatively charged silica substrate, which depends linearly on the Debye screening length and the polymer's line charge density, a nonelectrostatic interaction plays an important role in the desorption process. We have used poly(2-acrylamido-2-methylpropanesulfonic acid) (PAMPS) and its random copolymer containing 18-crown-6 (PAMPS-*co*-crown) to study the nonelectrostatic contribution to adsorption/desorption force [25]. The typical force curves of PAMPS with a plateau are obtained from amino-modified quartz in the buffer of water. The long plateau suggests that the PAMPS chain adopts a train-like conformation at the interface. The dynamic force spectroscopy reveals that the adsorption/desorption of this polymer is an equilibrium process in the experimental timescale. Therefore, the measured desorption force of about 120 pN is equal to its adhesive force.

An interesting finding is that the external salt does not influence the desorption force of PAMPS [25]. One possible explanation is that the introduced spacer between the charged groups and the backbone should effectively enhance the nonelectrostatic contribution to the interfacial interaction, making the interfacial interaction insensitive to the ionic strength. The force curves of PAMPS-*co*-crown obtained from amino-modified substrate are resembled to that of PAMPS. The long plateau indicates that PAMPS-*co*-crown chains also assume a train-like conformation at the interface. Similar to PAMPS, the desorption force of PAMPS-*co*-crown is independent of the concentration of electrolyte and the stretching velocity. The result indicates that the 20% content of crown ether side groups in the copolymer chain does not influence the desorption force. In other words, the interaction between PAMPS-*co*-crown chain and the substrate should be mainly dominated by the hydrophobic interaction besides electrostatic interaction.

Based on the above discussions, we can give a tentative correlation between the force pattern and adsorption conformation of polymer chains on the solid support. A force profile containing a single peak corresponds to the detaching of a big loop or tail structure, a saw-tooth pattern corresponds to the detachment of small loop structures in series, and a force curve containing a long plateau should come from the detachment of a train-like structure [5].

Besides the investigation of the interfacial conformation and adhesive energy of polymers at solid/liquid interface, the AFM-based force spectroscopy can be also used to measure the friction coefficient of single polymers adsorbed on surfaces. Gaub et al. have shown that although a force of several tens of piconewtons is required to desorb DNA from a mica surface, the friction force for lateral movement on the surface is extremely low [26]. It has been shown that adsorbed polymers with comparable desorption forces may exhibit drastically different in plane mobility.

5. Direct measurements of intermolecular forces

Intermolecular forces control the self-assembly and direct assembly of molecular building blocks into organized supramolecular structures. The formation of molecular assemblies relies on different intermolecular forces and their cooperative effect, including electrostatic interaction, hydrogen bonding, π–π interaction, van der Waals forces, and hydrophobic effect. Although the nature for each individual interaction is clear, the situation in molecular assemblies becomes complicated. The reasons are the combinatory and cooperative effect in intermolecular interactions, which may result in directivity and selectivity in molecular assemblies. It should be noted that the combined interaction of different intermolecular forces can be strong enough to form stable molecular assemblies, and in some cases the total binding strength can compete with chemical bonds. The molecular recognition refers to the specific interaction between host and guest molecules, and it is crucial for the formation of highly ordered molecular assemblies [27].

In order to realize the force measurement of single molecular rupture, one of the interacting components needs to covalently attach to AFM tip through a flexible PEG spacer and the other components to the substrate surface. The tip is then brought into contact with the substrate so that the formation of specific interaction between the immobilized tip and the surface-immobilized component is established. Pulling the interacting molecules apart can lead to the rupture of the noncovalently specific interaction. The introduction of PEG as a spacer provides a means for differentiating the force signals based on the extension length as well as for avoiding the disturbance of the nonspecific interaction between AFM tip and the substrate [28,29].

5.1. π–π interaction between pyrene and graphite

The π–π interaction is ubiquitous and important in diverse phenomena, including in stabilizing DNA and protein structures, packing aromatic molecules in crystals, binding guest moieties to host systems, and other supramolecular assemblies. It has been suggested that π–π interactions consist of the electrostatic, hydrophobic, and van der Waals forces. Although the π–π interaction has attracted a great deal of interests from both the theoretical and the experimental frontiers, it is still a big challenge to directly measure the π–π interaction at a single molecular level using conventional methods.

Pyrene derivatives can adsorb onto the surface of carbon nanotubes and graphite through π–π interactions to functionalize these inorganic building blocks with organic surface moieties. The interaction of the aromatic system of pyrene with carbon nanotube is so effective that certain pyrene derivatives tethered onto the nanotube surface have been used as molecular linkages for immobilizing proteins or fabrication of layer-by-layer multilayer films. We have used pyrene and graphite as a model system to investigate the π–π interaction. The pyrene molecule was attached to AFM tip through a PEG spacer. The π–π interaction between pyrene and graphite is thus indicated to be about 55 pN [30]. It is also found that the rupture force is independent of the force loading rate, indicating that the rupture events occur at an equilibrium state, and in this case the rupture force is equal to the adhesive interaction.

5.2. Intercalation interactions between dsDNA and acridine

Intercalation occurs by the insertion of planar ligands into neighboring base pairs of dsDNA. The planar ligand that can be inserted into dsDNA is called intercalator. The intercalation is

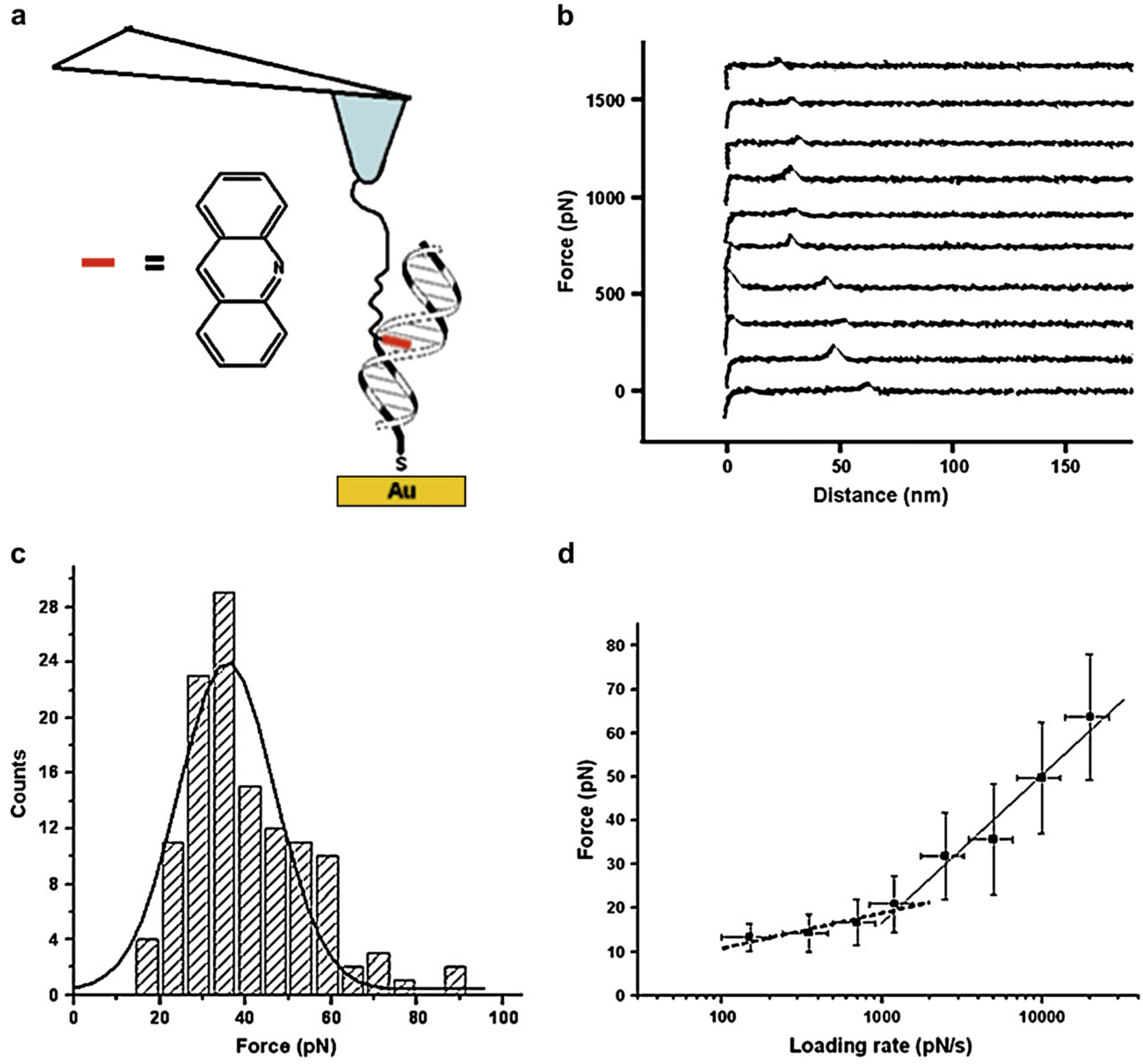


Fig. 6. (a) Schematic setup for measuring the intercalation interactions between acridine and dsDNA using SMFS. (b) Force curves measured between AFM tip modified with acridine and Au substrate functionalized with dsDNA in PBS (phosphate buffered saline, pH 7.4) at room temperature (~25 °C). (c) Histogram of rupture force of the interaction between acridine and dsDNA at a loading rate of 5.0 (±1.5) nN/s. The most probable rupture force is 36 pN. (d) Plot of most probable rupture force between acridine and dsDNA as a function of corresponding loading rate in a logarithmical scale [31].

believed to result from collective interactions of different forces, such as electrostatic interactions, π-stacking, hydrogen bonding, van der Waals force, and hydrophobic interactions, depending on the structure of intercalators. This line of research is of great interest to researchers in different fields, including molecular recognition, chemotherapy, and nanomedicine.

Acridine and its derivatives are typical intercalators for dsDNA. To study the intercalation between acridine and dsDNA, we have immobilized dsDNA bearing thiol group at 5′-terminus onto gold substrate through a thiol–Au bond, and attached acridine on AFM tip through a flexible PEG [31]. The most probable rupture force between dsDNA and acridine is ∼36 pN by Gaussian fitting, as shown in Fig. 6. With dynamic force spectroscopy, details about the kinetics of the binding and information concerning the length scale of the interaction can be extracted. For example, there are two slopes for the relationship between the most probable rupture forces and force loading rate. The combination of SMFS experimental data with the theoretical model [32] indicates the presence of two energy barriers along with the unbinding trajectory of acridine–dsDNA. One of the energy barriers with long interactive distance may be related to long range interaction, e.g. hydrophobic interaction; while the energy barrier with short interactive distance should be corresponding to short range interaction, e.g. π–π interaction. Therefore, the SMFS can provide not only the intermolecular interactions directly but also physical insight about the nature of the interactions.

5.3. *Interaction between poly(benzyl ether) dendrons*

The study of the interaction between surface-bound dendrons of different generations can provide a model system for understanding the surface adhesion of molecules with multiple branches. For these purpose, two generations of poly(benzyl ether) dendrons were synthesized, and one dendron was immobilized on AFM tip through a PEG spacer, while the other dendron was anchored on gold substrate as a self-assembled monolayer (SAM) [33]. The stretching of the single PEG chain, which is well established [17], can be used to indicate whether a single dendron is manipulated. The rupture force between first generation dendrons on AFM tip and the gold substrate is about 235 pN at a force loading rate of 40 nN/s. Since poly(benzyl ether) has no charges or permanent dipole, the interaction between two dendrons cannot be charge–charge or dipole–dipole interaction. Therefore van der Waals interaction should contribute to the interaction between two dendrons. In addition, considering the phenyl groups interacting to each other when the position is suitable, there should have π–π interaction between two dendrons as well. At the same loading rate, the rupture force between second and first generation dendrons is increased to 315 pN. This disproportion is reasonable if considering the topological structure of the dendrons. On the one hand, the interaction between two dendrons should increase with the increase of the generation of dendrons. On the other hand, the dendron modified on the AFM tip cannot guarantee a full contact with the surface anchored dendron on the substrate because of the topological structure, especially for the second-generation dendron. These two factors may work together, resulting in the enhanced but not simply proportional interaction for the dendrons increase from first generation to second generation. Furthermore, there is a loading rate dependence of the rupture force between two dendrons. The rupture force increases with the increase of loading rate. The linear dependence of rupture force on the logarithm of the loading rate indicates that the rupture event is a dynamic process.

6. Single chain polymer motor

SMFS allows for the realization of macromolecular motors with AFM cantilever if the polymer is stimulus responsive. Gaub et al. have utilized a stimuli-responsive polymer of synthetic polypeptide with multiple photoactive azobenzene groups incorporated into the backbone as an example of the optomechanical energy conversion at single molecule level [34,35]. The contour length of the polymer can be selectively lengthened or shortened by switching between the *trans*- and *cis*-azo configurations with irradiation of 420 and 365 nm lights, respectively. Because this mechanical work at the molecular level results from a macroscopic optical excitation ($E_{opt} \sim 10$ mJ), the real efficiency of the prototype optomechanical motor is roughly estimated only on the order of 10^{-18}.

Other responsive polymer is poly(ferroncenylsilane) (PFS), which can be extended upon oxidation and retracted upon reduction. Therefore PFS can be used as a good model system for the realization of an electrochemically powered molecular motor as proposed first by Vancso et al. [36,37]. Recently, we have worked together to realize closed mechanoelectrochemical cycles of single PFS chains using electrochemical SMFS [38]. The experiments are based on the previously established SAM platform obtained by the insertion and subsequent stable attachment of ethylene sulfide end-capped PFS in SAMs of 11-mercapto-1-undecanol on gold

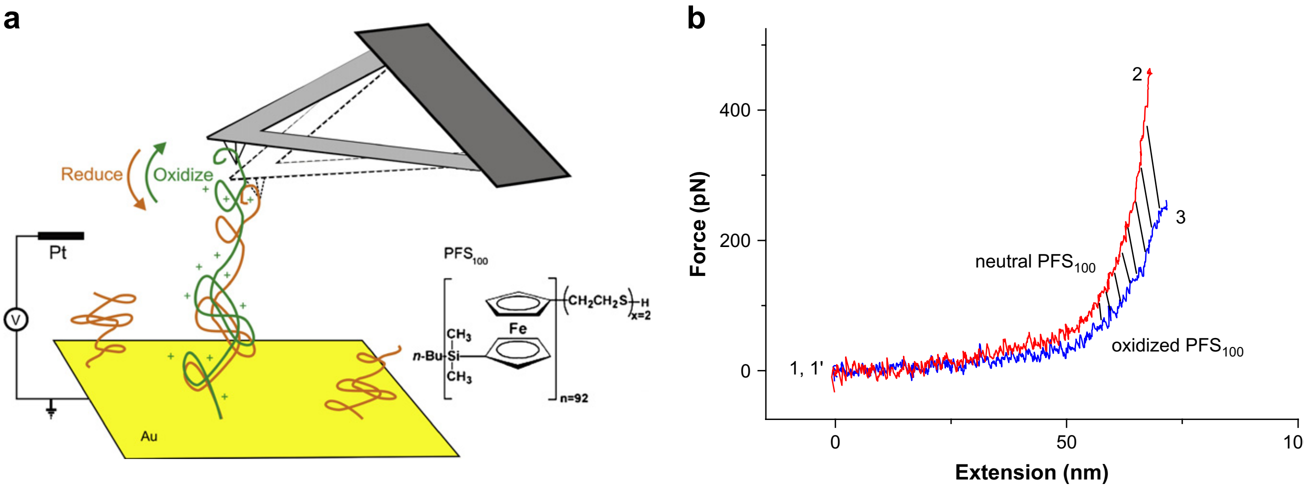


Fig. 7. (a) Stretching of single stimulus-responsive ethylene sulfide end-capped PFS (PFS100) chains by electrochemical AFM-based SMFS. (b) Force–extension curves of one cycle. The enclosed areas of the cycles correspond to the mechanical work input or output of the single polymer chain mechanoelectrochemical cycle [38].

electrodes. A chain lengthening of oxidized with respect to neutral chains is observed, which is attributed to the electrostatic repulsion between the oxidized ferrocene centers along the chain, as shown in Fig. 7. The PFS chain can be lengthened upon oxidation, whereas contracted upon reduction. To repeat such a process of lengthening and contracting by electrochemical method just functions as a single chain polymer motor. Electromechanical cycles are completed in both directions as closed loops and showed comparable efficiencies. The integration of the close force loops gives the work done, and this work is divided by the energy input of electrochemical method, thus giving the electromechanical efficiency. Notably, the single-chain efficiency is found to increase with increasing stretching ratio, and experimentally, a maximum efficiency of 26% is obtained.

7. Outlook

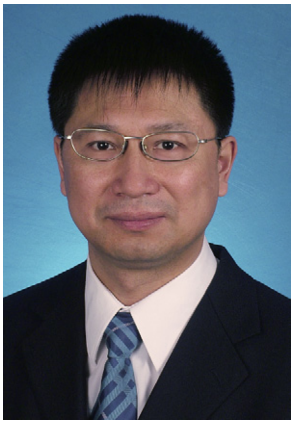
There is no doubt that SMFS is a powerful means for studying the intramolecular and intermolecular interactions in single molecule level, thus providing a lot of information which is not available by conventional methods. The importance and recent advances can be also reflected in a special issue of Langmuir, which appears just the beginning of this year for the highlight of research on molecular and surface forces [39]. Although some of the data about single chain mechanics are well documented, it is rather difficult to establish the relationship between macroscopic and microscopic worlds. Considering the importance of intermolecular interactions that drives for the molecular and supramolecular assemblies, SMFS can play an important role in studying these different interactions and revealing their nature. To fulfill this aim in the long run, the interplay between SMFS experimental study and theoretical simulation needs to be enhanced. One of the recent advances that are worth highlighting is the use of SMFS for manipulating single polymer chains, leading to the assembly of functional nanostructure and nanodevices. Besides mentioned above the single chain polymer motor, very recently Gaub et al. have combined the precision of AFM with the selectivity of DNA hybridization, developing a new method for bottom–up assembly of biomolecular structures [40].

Acknowledgment

The authors thank the Natural Science Foundation of China (20474035) and National Basic Research Program (2007CB808000) for financial support.

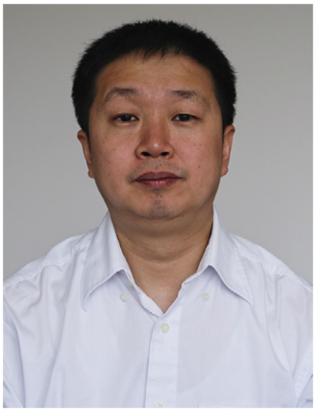
References

[1] Binnig G, Quate CF, Gerber C. Atomic force microscope. Phys Rev Lett 1986;56:930–3.
[2] Weisenhorn AL, Hansma PK, Albrecht TR, Quate CF. Forces in atomic force microscopy in air and water. Appl Phys Lett 1989;54:2651–3.
[3] Janshoff A, Neitzert M, Oberdörfer Y, Fuchs H. Force spectroscopy of molecular systems – single molecule spectroscopy of polymers and biomolecules. Angew Chem Int Ed 2000;39:3212–37.
[4] Hugel T, Seitz M. The study of molecular interactions by AFM force spectroscopy. Macromol Rapid Commun 2001;22:989–1016.
[5] Zhang WK, Zhang X. Single molecule mechanochemistry of macromolecules. Prog Polym Sci 2003;28:1271–95.
[6] Liu CJ, Shi WQ, Cui SX, Wang ZQ, Zhang X. Force spectroscopy of polymers: beyond single chain mechanics. Curr Opin Solid State Mater Sci 2005;9:140–8.
[7] Zhang X, Vancso GJ. Editorial: special issue on single chain polymers. Polymer 2006;47:2482.
[8] Wang C, Shi WQ, Zhang WK, Zhang X, Katsumoto Y, Ozaki Y. Force spectroscopy study on poly(acrylamide) derivatives: effects of substitutes and buffers on single-chain elasticity. Nano Lett 2002;2:1169–72.
[9] Shi WQ, Cui SX, Wang C, Wang LY, Zhang X, Wang XJ, et al. Single-chain elasticity of poly(ferrocenyldimethylsilane) and poly(ferrocenylmethylphenylsilane). Macromolecules 2004;37:1839–42.
[10] Zou S, Schonherr H, Vancso GJ. Stretching and rupturing individual supramolecular polymer chains by AFM. Angew Chem Int Ed 2005;44:956–9.
[11] Zou S, Schonherr H, Vancso GJ. Force spectroscopy of quadruple H-bonded dimers by AFM: dynamic bond rupture and molecular time–temperature superposition. J Am Chem Soc 2005;127:11230–1.
[12] Marszalek PE, Oberhauser AF, Pang YP, Fernandez JM. Polysaccharide elasticity governed by chair–boat transitions of the glucopyranose ring. Nature 1998;396:661–4.
[13] Li HB, Rief M, Oesterhelt F, Gaub HE, Zhang X, Shen JC. Single-molecule force spectroscopy on polysaccharides by AFM – nanomechanical fingerprint of α-1,4-linked polysaccharides. Chem Phys Lett 1999;305:197–201.
[14] Xu QB, Zhang WK, Zhang X. Oxygen bridge inhibits conformational transition of 1,4-linked-D-galactose detected by single-molecule atomic force microscopy. Macromolecules 2002;35:871–6.
[15] Zhang QM, Jaroniec J, Lee G, Marszalek PE. Direct detection of inter-residue hydorgen bonds in polysaccharides by single-molecule force spectroscopy. Angew Chem Int Ed 2005;44:2723–7.
[16] Liu CJ, Wang ZQ, Zhang X. Force spectroscopy of single-chain polysaccharides: force-induced conformational transition of amylose disappears under environment of micelle solution. Macromolecules 2006;39:3480–3.
[17] Oesterhelt F, Rief M, Gaub HE. Single molecule force spectroscopy by AFM indicates helical structure of poly(ethylene-glycol) in water. New J Phys 1999;1:6.1–6.11.
[18] Li HB, Zhang WK, Zhang X, Shen JC, Liu BB, Gao CX, et al. Single molecule force spectroscopy on poly(vinyl alcohol) by atomic force microscopy. Macromol Rapid Commun 1998;19:609–11.
[19] Li HB, Zhang WK, Xu WQ, Zhang X. Hydrogen bonding governs the elastic properties of poly(vinyl alcohol) in water: single-molecule force spectroscopic studies of PVA by AFM. Macromolecules 2000;33:465–9.
[20] Liu CJ, Cui SX, Wang ZQ, Zhang X. Single-chain mechanical property of poly(*N*-vinyl-2-pyrrolidone) and interaction with small molecules. J Phys Chem B 2005;109:14807–12.
[21] Zhang GZ, Winnik FM, Wu C. Structure of a collapsed polymer chain with stickers: a single- or multiflower. Phys Rev Lett 2003;90:035506.1–4.
[22] Cui SX, Liu CJ, Zhang WK, Zhang X, Wu C. Desorption force per polystyrene segment in water. Macromolecules 2003;36:3779–82.
[23] Hugel T, Grosholz M, Clausen-Schaumann H, Pfau A, Gaub HE, Seitz M. Elasticity of single polyelectrolyte chains and their desorption from solid supports studied by AFM based single molecule force spectroscopy. Macromolecules 2001;34:1039–47.
[24] Seitz M, Friedsam C, Jostal W, Hugel T, Gaub HE. Probing solid surfaces with single polymers. Chem Phys Chem 2003;4:986–90.
[25] Cui SX, Liu CJ, Wang ZQ, Zhang X, Strandman S, Tenhu H. Single molecule force spectroscopy on polyelectrolytes: effect of spacer on adhesion force and linear charge density on rigidity. Macromolecules 2004;37:946–53.
[26] Kühner F, Erdmann M, Sonnenberg L, Serr A, Morfill J, Gaub HE. Friction of single polymers at surfaces. Langmuir 2006;22:11180–6.
[27] Zhang X, Shen JC. Supramolecular science: a new way to understand the matter world. Chin Sci Bull 2003;48:1517–8.
[28] Kudera M, Eschbaumer C, Gaub HE, Schubert U. Analysis of metallo-supramolecular systems using single-molecule force spectroscopy. Adv Funct Mater 2003;13:615–20.
[29] Kersey FR, Yount WC, Craig SL. Single-molecule force spectroscopy of bimolecular reactions: system homology in the mechanical activation of ligand substitution reactions. J Am Chem Soc 2006;128:3886–7.
[30] Zhang YH, Liu CJ, Shi WQ, Wang ZQ, Dai LM, Zhang X. Direct measurements of the interaction between pyrene and graphite in aqueous media by single molecule force spectroscopy: understanding the π–π interactions. Langmuir 2007;23:7911–5.
[31] Liu CJ, Jiang ZH, Zhang YH, Wang ZQ, Zhang X, Feng FD, et al. Intercalation interactions between dsDNA and acridine studied by single molecule force spectroscopy. Langmuir 2003;23:9140–2.
[32] Evans E. Probing the relation between force–lifetime and chemistry in single molecular bonds. Annu Rev Biophys Biomol Struct 2001;30:105–28.
[33] Shi WQ, Zhang YH, Liu CJ, Wang ZQ, Zhang X. Interaction between dendrons directly studied by single-molecule force spectroscopy. Langmuir 2008;24:1318–23.
[34] Hugel T, Holland NB, Cattani A, Moroder L, Seitz M, Gaub HE. Single-molecule optomechanical cycle. Science 2002;296:1103–6.
[35] Holland NB, Hugel T, Neuert G, Cattani-Scholz A, Renner C, Oesterhelt D, et al. Single molecule force spectroscopy of azobenzene polymers: switching elasticity of single photochromic macromolecules. Macromolecules 2003;36:2015–23.
[36] Zou S, Ma YJ, Hempenius MA, Schönherr H, Vancso GJ. Grafting of single, stimuli-responsive poly(ferrocenylsilane) polymer chains to gold surfaces. Langmuir 2004;20:6278–87.
[37] Zou S, Korczagin I, Hempenius MA, Schönherr H, Vancso GJ. Single molecule force spectroscopy of smart poly(ferrocenylsilane) macromolecules: towards highly controlled redox-driven single chain motors. Polymer 2006;47:2483–92.
[38] Shi WQ, Giannotti M, Zhang X, Hempenius M, Schönherr H, Vancso GJ. Closed mechanoelectrochemical cycles of individual single-chain macromolecular motors by AFM. Angew Chem Int Ed 2007;46:8400–4.
[39] Schwartz DK, Whitten DG. Molecular and surface force special issue. Langmuir 2008;24:1109.
[40] Kufer SK, Puchner EM, Gumpp H, Lidel T, Gaub HE. Single-molecule cut-and-paste surface assembly. Science 2008;319:594–6.



Xi Zhang is a Professor of the Department of Chemistry at Tsinghua University. His main scientific interests are in the areas of supramolecular assembly and polymer thin films, and single molecule force spectroscopy of polymers. He received his MS and Ph.D. degrees in polymer chemistry and physics at Jilin University under the supervision of Prof. Jiacong Shen. During his PhD study, he spent more than one year at the University of Mainz, Germany, as a joint-training Ph.D. student under the supervision of Prof. Helmut Ringsdorf. He joined the Key Laboratory of Supramolecular Structure and Materials, Jilin University, in 1992, as a lecturer and then was promoted to be full professor. He moved to Tsinghua University in late 2003. He serves as members of the editorial advisory boards of Langmuir, European Polymer Journal, and Chemical Communications.

Dr. Chuanjun Liu received his Ph.D. in polymer chemistry and physics at Jilin University in 2007 under supervision of Prof. Xi Zhang. Now he is doing his post-doc research at MPI-Polymer Research in Mainz with Prof. Hans-Jürgen Butt. His research interest is single-molecule polymers.



Prof. Zhiqiang Wang received his Ph.D. in polymer chemistry and physics at Jilin University in 1997 under supervision of Prof. Jiacong Shen. Since 2003, he has been a professor of Department of Chemistry, Tsinghua University. His research interest is interfacial self-assembly and pattern formation.