Adsorption of polyacrylic acid on self-assembled monolayers investigated by single-molecule force spectroscopy

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New Journal of Physics 6 (2004) 9
Received 15 October 2003
Published 30 January 2004
Online at http://www.njp.org/ (DOI: 10.1088/1367-2630/6/1/009)

Abstract. Single-molecule force spectroscopy was used to study the adsorption and desorption processes of individual polyacrylic acid (PAA) chains from self-assembled monolayers (SAMs) with different terminal groups (CH₃, OH, COOH). PAA was grafted to the tip of an AFM cantilever and all measurements were recorded with the same tip. For the OH- and the COOH-SAM, the desorption force was found to be independent of the salt concentration for NaCl, as well as for CaSO₄. On CH₃-SAM, a repulsive electrostatic interaction was found with NaCl in solution whereas a slightly attractive electrostatic interaction occurred with CaSO₄ in solution. On the OH- and the COOH-terminated SAM, the non-electrostatic contributions to the desorption forces are of comparable magnitude (∼50–65 pN) whereas the non-electrostatic contribution for the CH₃-terminated SAM is significantly higher (∼80–90 pN). Additional experimental findings concerning the length, the total number of adsorption events and the number of multiple polymer strands adsorbed in parallel, support the assumption that this difference in adsorption force is due to a different quality of the solvent for the polyacrylic acid chains at the SAM–water interface. Comparative surface pressure measurements at the air–water interface revealed a surface energy for polyacrylic acid of 30 mJ m⁻² which would correspond to a force contribution of ∼10 pN to the desorption force. This is in good agreement with the experimental results on hydrophobic SAMs. For further characterization of the surfaces the pH was varied from 3 to 11 at constant salt concentration (for a NaCl concentration of

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100 mM and a CaSO₄ concentration of 5 mM, respectively). In the presence of NaCl, the desorption force was found to be independent of the pH for all three substrates. For CaSO₄, the desorption force was pH-independent for the OH- and the CH₃-SAM. In the case of the COOH-SAM, a jump from 60 to 90 pN was recorded at pH 8, which we attribute to the formation of salt bridges between the polyacrylic acid chain and the COOH-SAM at higher pH.

1. Introduction

Intermolecular interactions impact a variety of phenomena in material science, such as adhesion or friction [1, 2]. They also play a leading role in the fields of biology and life sciences, e.g. membrane assembly [3]–[8] or protein folding [9]–[12]. Particularly in biology, weak non-covalent interactions, such as electrostatic, hydrogen bonding or hydrophobic/hydrophilic interactions, control essential processes [3]. The ability to detect and interpret such intermolecular interactions under different environmental conditions provides the unique opportunity to gain deeper insight in the underlying mechanisms and derive a more detailed understanding of such processes at the molecular level.

Single-molecule force spectroscopy has emerged as a powerful technique, which allows the measurement of intra- and intermolecular forces with unparalleled precision and sensitivity [13]–[18]. The measured binding forces range from the piconewton regime, e.g. for receptor–ligand systems [19]–[21] to the nanonewton regime that is reached when single covalent bonds are ruptured [22]. Most of these force spectroscopy measurements take place under non-equilibrium conditions and therefore the rupture forces depend on the force loading rates [23]–[29]; the rupturing of the investigated bonds is an irreversible process. Non-equilibrium conditions are typically reached if the loading time is comparable to or shorter than the natural lifetime of the bond. In turn, if the dissociation and re-association of the binding partners occurs at a much shorter time scale, the measurement occurs close to thermal equilibrium. In this case, no loading rate dependence is observed and the investigated process is reversible.
Depending on the chemical nature, the physical parameters and the solvent conditions, the interaction between polymers and surfaces may vary over a broad range, not only in terms of strength, but also in terms of characteristic time scales. Spontaneous desorption processes may occur as fast as microseconds—in the case of small molecules—but may approach infinity for modern adhesion glues. As a consequence, on the typical timescale of a single molecule force spectroscopy experiment with a typical 10 kHz corner frequency of the cantilever, a broad spectrum of reversible and irreversible processes may be investigated. In this study, we focus on quasi-equilibrium desorption measurements of single polyelectrolyte chains from solid supports, which consequently show no loading rate dependence [30]–[32]. Such experiments have recently been introduced as a subject of investigation in single-molecule force spectroscopy. As there are numerous applications in material science, biology, medicine and engineering, the adhesion of polyelectrolytes on surfaces is in the focus of experimental research [33] as well as theoretical modelling [34]–[37].

In previous studies, single-molecule force spectroscopy measurements revealed valuable details of the adsorption process for single polyelectrolyte strands on charged substrates. For example, the loop-size distribution of adsorbed chains at the interface could be determined [30] as well as the desorption forces\(^4\) needed for pulling the strands away from the substrate [30]–[32]. The equilibrium desorption forces for weak polyelectrolytes adsorbed on negatively charged surfaces were found to be linearly dependent on the Debye screening length, the surface number charge density and the line number charge density of the polyelectrolytes [31, 32]. With the stable covalent attachment of the polyelectrolytes to the AFM-tips, it became possible to investigate the adsorption process of the same set of polyelectrolyte molecules on a variety of surfaces and therefore deviations due to different sample preparations could be ruled out [38]. One additional advantage of this technique is the fact that single polyelectrolyte molecules represent very small probes so that roughness or inhomogeneity of the investigated surface just becomes relevant on the scale of the persistence length of the polymer molecule. In summary, polyelectrolytes grafted to an AFM tip were established as an astonishingly precise analytical tool to investigate the properties of generic surfaces.

Following this approach we extended our measurements to the investigation of SAMs with different head groups with the goal to focus on the contributions of non-Coulombic interactions. SAMs represent well-defined and easily accessible systems [39]. The possibility to tailor the head groups of the constituent molecules makes them an excellent subject for the investigation of phenomena affected by competing intermolecular, molecule–substrate and molecule–solvent interactions. We have investigated the adsorption of poly acrylic acid on SAMs with different head groups, with different ions in solution and varied pH. The results were compared to measurements with a negatively charged mineral surface and a positively charged surface functionalized with quaternary amines. The SAMs consisted of CH\(_3\)-terminated, OH-terminated and COOH-terminated alkylchains that represent a hydrophobic, a polar and an acidic surface, respectively. This work represents a systematic approach to study the relationship between polyacrylic acid surface adhesion and substrate chemistry. It provides complementary results to previous work, e.g. AFM-measurements that focus on the interaction between SAMs with different headgroups [40, 41] or investigations on the properties of water at the SAM–water interface [42]–[44].

\(^4\) The term desorption force is used in this paper to make clear that this force is attributed to the desorption process. Alternatively, we could denote this force also as ‘adsorption-force’ which would emphasize the fact that we actually measure the equilibrium adsorption force.
2. Materials and methods

The cantilevers were purchased from Veeco Instruments. Chromium–nickel and gold, which were used for the evaporation process, were purchased from Degussa-Hüls. Mercaptoundecanol, mercaptohexadecanoic acid, decanthiol and polyacrylic acid were purchased from Aldrich, phosphate-buffered saline, N-hydroxysuccimidy, 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide, ethylenediamine, ethylenediaminetetraacetic acid (EDTA), CaSO₄ and NaCl were from Sigma. Calcite (100) crystals were provided by R. U. Barz (Institut für Mineralogie, LMU, München, Germany). The chemical vapour deposition (CVD) chamber used and the AFM were home-built instruments.

2.1. Preparation of the cantilevers

For the covalent attachment of polyacrylic acid, the cantilevers were gold-coated by thermal evaporation and then coated with a mixed 1 : 1 SAM with OH- and COOH- as terminal groups. The carboxy functions of the resulting self-assembled thiol monolayer were then reacted with ethylenediamine and finally functionalized with polyacrylic acid. The final PAAc-loaded AFM cantilevers were kept overnight in 1 mM aqueous EDTA solution containing traces of sodium chloride and stored in a dessicator if not immediately used in an AFM experiment. This procedure is described in more detail elsewhere [32, 38].

2.2. Preparation of the thiol SAM samples

The glass slides were coated by thermal evaporation with 10 nm of chrome-nickel (80 : 20) followed by 40 nm of gold. They were then cleaned in a mixture (v/v/v = 5 : 1 : 1) of deionized water, ammonia (30 vol%), hydrogen peroxide (25 vol%) at 70°C for at least 1 h (caution: the solution causes severe burns!), and rinsed with Milli-Q water. To produce a SAM with OH as the terminal group the glass slides were incubated in a solution of 1 mM 11-mercaptoundecan-1-ol and 100 ml ethanol (puriss.) for 24 h and afterwards rinsed with ethanol. For the preparation of COOH-SAMs, the solution contained 1 mM of ω-mercaptohexadecanoic acid, and in the case of CH₃ 1 mM of decanthiol. The SAMs were afterwards immediately used for the experiments.

2.3. Preparation of the ammonium-functionalized silicon

A 1% solution of N-trimethoxysilylpropyl-N,N,N-trimethylammonium chloride (ABCR GmbH/Gelest, Germany; from commercial stock solution of 50% silane in methanol) in H₂O was stirred for 20 min and filtered through a 0.2 µm PTFE filter. The substrates were immersed into the prehydrolysis solution for 30 min, washed with Milli-Q water and baked at 95°C for 1 h. After washing with a solvent sequence (acetone, Milli-Q water, acetone, THF) the substrates were sonified in acetone, which yielded high-quality silane monolayers. Water contact angle: 72°/69°/47° (advancing/static/receding); ellipsometric film thickness: 0.5 (±0.1) nm. The whole procedure is described in greater details elsewhere [45].

2.4. Force spectroscopy experiments

The force spectroscopy experiments were performed in liquid environment. The tip was approaching the surface with a velocity of 1500 nm s⁻¹ and was retracted with the same speed.
Figure 1. Experimental set-up: the polymers were covalently attached to the AFM-tip. When the cantilever is approaching the surface, the molecules are allowed to adsorb on to the opposing SAM-covered surface. When the cantilever is retracted, the polymers are desorbed from the surface and the desorption force is measured via the deflection of the cantilever. Desorption measurements of polyacrylic acid were performed with three different SAM-surfaces: one CH$_3$-, the second OH- and the third COOH-terminated. All experiments took place in liquid environment with NaCl or CaSO$_4$ in solution. (See also movie.)

immediately after touching the surface (see figure 1, movie). The measurements were performed with NaCl or CaSO$_4$ in water at pH 6 on the three different SAMs (COOH, OH, CH$_3$ as terminal groups). The concentrations of the ions were increased in five steps: 5, 10, 25, 50 and 100 mM for NaCl and 0.05, 0.1, 0.5, 1 and 5 mM for CaSO$_4$. For each salt concentration, a set of 500 force curves were recorded. To eliminate deviations due to different cantilever preparations, all measurements with varied ion concentrations were made with one and the same tip. A summary of the measurements is presented in figure 4. Each time when the SAM-sample was changed, the tip was washed thoroughly and then stored for a couple of hours in a solution containing 1 mM EDTA and traces of NaCl to restore the initial state of the AFM-cantilever.

Furthermore, measurements with varied pH and constant ion concentration were undertaken for NaCl as well as for CaSO$_4$ with all three types of SAMs. The constant NaCl concentration was 100 mM and CaSO$_4$ concentration was 5 mM. The pH was adjusted by the addition of HCl and NaOH, respectively. The pH was increased in steps of 1 and for each pH a set of 500 curves were recorded. The complete set of pH measurements for a special SAM-sample was performed with one and the same cantilever to avoid deviations due to different cantilever preparations.

Measurements on calcite as well as the measurements on aminofunctionalized silicon were performed with a varied NaCl concentration series analogous to the measurements on the SAMs and again with one and the same polyacrylic acid functionalized AFM-tip.

2.5. Surface pressure measurements

The surface tension of a 5 ml concentrated solution (300 mg ml$^{-1}$) of polyacrylic acid was measured with a Wilhelmi-type pressure transducer system of a homebuilt Langmuir trough [46].
Figure 2. Typical force–distance curves for the three substrates. When the cantilever was retracted in all three cases typical desorption plateaux were recorded (red: approach curve, black: retraction curve). If more than one step is observed, multiple molecules with different lengths adsorbed on to the opposing SAM-surfaces which were then desorbed upon retraction one after the other. (a) CH₃-SAM: in the majority of the scans, desorption of only a single polyacrylic acid molecule was observed. (b) OH-SAM: desorption of two or three polyacrylic acid molecules can be observed more frequently. Compared with the CH₃-SAM, the desorption plateaux are generally longer. (c) COOH-SAM: adsorption of multiple molecules is very frequent. Also the desorption of very long molecules can be observed.

When the surface tension reached a steady value, a few drops of concentrated hydrochloric acid was added, which resulted in the precipitation of excess PAA. Finally, the surface tension of the saturated solution was recorded.

3. Results

For all three types of thiol SAMs, measurements with varying salt concentrations at pH 6 were undertaken for NaCl as well as for the bivalent salt CaSO₄. Figure 2 shows typical force curves for the three SAMs. They were all recorded with NaCl in solution but were also representative for the CaSO₄ measurements. The tip with the grafted PAA was approached towards the surface, while the interaction force was recorded. Upon retraction of the tip for distances of several hundreds of nanometres, the interaction force was measured to stay at a constant level until it eventually dropped discontinuously to zero. If within such a force plateau, the tip was approached...
again towards the surface, the interaction force did not change (results not shown). This finding indicates that the experiment occurs close to the thermodynamic equilibrium, which means that the involved polymer segments dissociate and re-associate with the surface on a much faster time scale than the experimental pulling process occurs. The plateau length directly reflects the length of an adsorbed polymer whereas the height of a plateau corresponds to the desorption force that is required to desorb one or multiple polymers from the opposing surface.

On the CH₃-SAM only short desorption plateaux were found and in most cases only one plateau was observed (figure 2(a)). This means that the desorption of two or more strands in parallel, which would be indicated by additional desorption plateaux at higher forces, is a very rare event. The desorption plateaux were found to be longer on the OH-SAM and occasionally two or more steps occurred (figure 2(b)). On the COOH-SAM, very long plateaux and many steps were observed (figure 2(c)), which indicates that multiple polymer strands had adsorbed on to the surface in parallel, including also the longer polymers. These features found in the force-extension curves are also reflected in the histograms of the measured desorption forces. Three of them, all measured with NaCl in solution are shown in figure 3. For the CH₃-SAM, only one peak appears that represents the desorption of a single polyelectrolyte chain of the SAM-surface, whereas for the OH-SAM a second peak and for the COOH-SAM at least a third peak can be identified, which represent the desorption of two and three polyelectrolyte strands in parallel, respectively. The number of desorption events found in a set of 500 force-extension curves was rather small for the CH₃-SAM (∼30%), larger for the OH-SAM (∼100%) and largest for the COOH-SAM (∼200%) (note that more than one polymer may desorb during one scan). This holds for the total number of desorption events as well as for single and multiple peaks in the force histograms as can be seen from figure 4. Lengths of desorption plateaux found during the whole measurement process are plotted against their curve number in figure 4(a). This graph also gives an overview of the sequence of measurements performed with one and the same tip. First, the ion concentration of NaCl was varied for the CH₃-SAM (curve number 0–2500), then the same was performed for the OH-SAM (curve number 2500–5000) and COOH-SAM (curve number 5000–7500). Then the SAMs were probed in the reversed order with a varied concentration of CaSO₄ in solution (COOH-SAM: curve number 7500–10000, OH-SAM: curve number 10000–12500, CH₃-SAM: curve number 12500–15000). As can be seen in the figure, measurements on the CH₃-SAM start with rather short desorption plateaux. Longer plateaux also appeared when the CH₃-SAM was exchanged against the OH-SAM and the longest plateaux were observed on the COOH-SAM for NaCl as well as for CaSO₄. The plateaux became shorter again, when the CaSO₄ measurements were performed on the OH-SAM and, finally, for the CaSO₄ measurements on the CH₃-SAM only short plateaux were found. In figures 4(b) and 4(c) all mean desorption forces extracted from the histograms by a Gaussian fit of the first peak found for the three different SAM-substrates are plotted against the Debye screening length (depends on the inverse square root of the salt concentration) of the different salt solutions. The desorption forces remain constant for the OH- and the COOH-SAM in the case of NaCl as well as for CaSO₄. No significant dependence on the Debye screening length was found. For the CH₃-SAM, the desorption force was found to decrease with the Debye screening length when NaCl was in solution and increase with the Debye screening length for CaSO₄. The desorption forces for the COOH- and the OH-SAM lie in the same range for both ions (∼54 pN for COOH and ∼48 pN for OH). When NaCl was exchanged against CaSO₄, the desorption force slightly decreased to 47 pN in the case of the COOH-SAM and slightly increased to 53 pN in the case of the OH-SAM. For both ions, the desorption force is significantly higher on the CH₃-SAM.
Figure 3. Histograms for the desorption forces (height of the desorption plateaux found in the force-extension curves of figure 2) of polyacrylic acid measured for the three substrates. The first peak in the histograms corresponds to the desorption of single molecules, the following peaks to the desorption of two or more molecules. Each of the histograms was extracted from a set of 500 force-extension curves that were recorded with different NaCl concentrations. The histograms were normalized to the number of measured force curves. (a) CH₃-SAM: only one peak appears that corresponds to the desorption of a single molecule. Compared with the OH- and the COOH-SAM, fewer desorption plateaux were found in the data set and the distribution of the desorption forces is rather broad. (b) OH-SAM: two peaks can be clearly identified, which correspond to the desorption of one and two polymer molecules, respectively. More desorption plateaux were found than in the case of the CH₃-SAM and the peaks are better defined. (c) COOH-SAM: three peaks can be distinguished, which correspond to the desorption of one, two and three polyacrylic acid molecules. The peaks are well-defined and a large number of desorption plateaux is found in the data set.

(∼77 pN for NaCl in solution and ∼80 pN for CaSO₄ in solution) compared with the OH- and the COOH-SAM.

For further characterization of the investigated SAMs and their interaction with polyacrylic acid, the pH was also varied for both salts at a constant salt concentration of 100 mM NaCl and 5 mM CaSO₄, respectively. The results for NaCl are plotted in figure 5. For all three SAMs the desorption force was found to be independent of the pH of the solution. In the case of CaSO₄ (cf figure 6) the same was found on the CH₃ and on the OH-SAM whereas on the COOH-SAM a jump in the desorption force was observed at pH 8.
Figure 4. Overview of all desorption lengths and desorption forces for single polyacrylic acid molecules measured for the three different thiol SAM-samples. In figure 4(a) the lengths of all plateaux found in the whole measurement are plotted against their curve number. In figures 4(b) and 4(c) all desorption forces found for different concentrations of NaCl and CaSO₄ are depicted against the Debye screening length. The straight lines signify the linear smoothing functions. (a) Desorption lengths for the whole data set. The first three blocks represent the measurements with varied NaCl concentrations on the three different SAMs, the last three blocks represent the measurements with varied CaSO₄ concentrations. For NaCl as well as for CaSO₄ the shortest plateaux were found for the CH₃-SAM. For the OH-SAM, longer plateaux appear in both cases, while the longest desorption plateaux for both ions were found for the COOH-SAM. (b) Measurements with NaCl in solution. The non-Coulombic contribution to the desorption force for the OH-SAM (48 pN) is the lowest. The non-Coulombic contribution for the COOH-SAM is slightly higher (54 pN) while for the CH₃-SAM it is remarkably higher (77 pN). In the case of CH₃-SAM, a repulsive electrostatic interaction can be identified. (c) Measurements with CaSO₄ in solution. The non-Coulombic contribution to the desorption force for the OH-SAM is slightly increased (53 pN) compared with the one for NaCl in solution and is slightly higher than the non-Coulombic contribution measured on the COOH-SAM (47 pN) that is decreased compared with the one measured for NaCl in solution. The non-Coulombic contribution for the CH₃-SAM is nearly unchanged (79 pN) and is again remarkably higher than the value measured for the other SAMs. In the case of the CH₃-SAM, an attractive electrostatic interaction can be identified. The desorption force on the other SAMs is salt-independent.
Figure 5. Variation of pH with 100 mM NaCl in solution. The desorption force was pH independent in all three cases. The mean desorption force was (a) 85 pN for CH$_3$-SAM, (b) 55 pN for OH-SAM, and (c) 61 pN for COOH-SAM.

In figure 7 the desorption forces of a single polyacrylic acid chain measured for different NaCl concentrations on calcite and on ammonium-silanized silicon are plotted against the Debye screening length. In both cases, a pronounced dependence on the Debye screening length was found: for calcite the desorption force slightly decreases and for the ammonium-silanized silicon the desorption force increases with increasing Debye screening length.

4. Discussion

Single molecules attached to an AFM cantilever were employed here as local sensors, which translate the minute interactions between polymers and surfaces of given chemical compositions into a macroscopically measurable signal. Obviously, these interactions may span a broad range both in strength and in their characteristic time scale. The measured desorption forces are composed of multiple contributions, which reflect the characteristic properties of the investigated surface. However, upon variation of the experimental conditions, these contributions may be separated. The electrostatic contribution to the desorption force has been derived previously [3, 31] based on the electrostatic potential $\phi(z)$ of the substrate in the Debye–Hückel approximation:

$$\frac{e \cdot \phi(z)}{k_B T} = -4\pi l_B \sigma \kappa^{-1} e^{-\kappa z},$$

where $k_B$ is the Boltzmann constant, $T$ the temperature, $\sigma$ the surface number charge density of the substrate, $\kappa^{-1}$ the Debye screening length and $l_B$ the Bjerrum length that signifies the distance...
Figure 6. Variation of pH with 5 mM CaSO$_4$ in solution. (a), (b) The desorption force was pH independent for CH$_3$- and OH-SAM. The mean desorption force was 90 and 71 pN for CH$_3$- and OH-SAM, respectively. (c) COOH-SAM: the desorption force depended on the pH. For pH 7 to pH 8 there was a jump from 64 to 91 pN. The mean desorption force was 75 pN.

Figure 7. Monitoring of electrostatic interactions between polyacrylic acid and two surfaces carrying surface number charge of different signs. (a) Measurements on calcite that represent a slightly negatively charged surface with NaCl in solution. The mean desorption force depends linearly on the Debye screening length adjusted by the NaCl concentration in the solution. The falling straight line indicates a repulsive interaction: the desorption force increases with increasing salt concentration. (b) Measurements on ammonium-functionalized silicon, which is positively charged, with NaCl in solution. The mean desorption force also depends linearly on the Debye screening length but the straight line is rising which indicates an attractive interaction because the desorption force decreases with increase in salt concentration.
at which the electrostatic interaction between two unit charges, $e$, in a solvent without counter ions equals the thermal energy $k_B T$.

The force that is required to pull the polymer away from the surface over a distance that is equal to the length $a$ of one polymer segment can be determined simply from the transfer of one charged segment with a line number charge density $\tau$ from $z = 0$ to $\infty$:

$$F_{\text{des}}^\text{el} = \frac{[\phi(\infty) - \phi(0)] \tau \cdot a}{a} = (4\pi l_B k_B T) \sigma \kappa^{-1} \tau. \quad (2)$$

In a first approximation, all non-electrostatic contributions to the desorption force (e.g. van der Waals or hydrophobic/hydrophilic contributions) are summed up in the constant additive term $F_0$ so that the overall desorption force is given by

$$F_{\text{des}} = F_0 + (4\pi l_B k_B T) \sigma \kappa^{-1} \tau. \quad (3)$$

As can be seen from equation (3) the desorption force depends linearly on the Debye screening length if electrostatic interaction is involved in the desorption process. This linear dependence can be easily verified in situ by varying the salt concentration in solution.

Two examples which display electrostatic interactions for polyacrylic acid are shown in figure 7. Figure 7(a) shows measurements on calcite, which has a slightly negatively charged surface with NaCl in solution. The desorption forces that were found for the different salt concentrations are plotted against their Debye screening lengths. As can be seen from the graph, the desorption force decreases linearly with the Debye screening length which indicates a repulsive interaction between the polyacrylic acid strands and the calcite surface. Figure 7(b) shows a graph that corresponds to measurements that were performed with varying NaCl concentrations on amino-functionalized silicon, which is positively charged. Also in this case the desorption force depends linearly on the Debye screening length but it increases with the Debye screening length. Therefore, the electrostatic interaction is attractive. The line-fit is much steeper than for calcite, which is attributed to a larger surface number charge density\(^5\) or to a higher degree of dissociation of the COOH-groups on the polyacrylic acid chain \cite{47}.\(^6\)

A linear dependence of the desorption force on the salt concentration was only found in the case CH$_3$-SAM as can be seen in figures 4(b) and 4(c): the desorption force was found to be increasing with the Debye screening length for NaCl and decreasing for CaSO$_4$. The first case corresponds to a repulsive electrostatic contribution which indicates a negative surface number charge of the CH$_3$-SAM. This is well in agreement with measurements performed by Schweiss et al \cite{48} who determined the surface potential of a CH$_3$-terminated SAM to be $\sim -70$ mV by $\zeta$-potential measurements. The negative potential is attributed to adsorbed hydroxyl ions at the SAM surface. The results for CaSO$_4$ in solution indicate an attractive electrostatic interaction. This could be mediated by Ca ions bound to the carboxyl groups of polyacrylic acid. For the OH- and the COOH-SAM, no significant dependence on the Debye screening length was found. This means that the adsorption is dominated by non-Coulombic interactions for these surfaces. Thus, after a comparison of the results for the three different SAMs, our further considerations have to focus on the constant term $F_0$ in equation (4).

\(^5\) If it is assumed that the polymer is fully charged, a surface number charge density of $0.08 \text{ nm}^{-2}$ would result.

\(^6\) Charge regulation effects mediated by the Coulomb interaction between the polymer and the surface can shift the pK$_a$ of polyacrylic acid. (see e.g. \cite{47}). This effect may interfere with the measured Coulombic interaction. One possibility to separate both the effects is to determine either the surface number charge density or the line number charge density of the polymer by $\zeta$-potential measurements (suggested by one of the referees).
A remarkable finding is the pronounced differences in the length distributions of the desorption plateaux on the different SAMs (figures 2 and 4(a)). They reflect the different length distributions of the polymers that were allowed to adsorb from the tip on to the surfaces upon contact. Obviously, the longer species on the tip did not adsorb predominantly on to the CH$_3$-terminated SAM (figures 2(a) and 4(a)). Also, the probability for the transfer was lowest on this surface (figure 3(a)) while the desorption forces where the highest (figures 3, 4(b) and 4(c)).

Using a hand-waving argument, this finding may be explained by differences in the solvent quality of the water at the different SAM interfaces. If the structured water close to the hydrophobic CH$_3$-SAM is assumed to be a poorer solvent for the polyacrylic acid than the water at the hydrophilic OH- and the COOH-SAMs, an additional barrier for the polymers to transfer from the tip onto the surface upon contact would be established. This barrier would kinetically hinder the transfer of the polymer from the tip on to the surface. The shorter polymers would still be forced into contact while the longer ones may escape the additional barrier (see figure 8). Probably, this might explain why fewer and shorter desorption events were observed for the CH$_3$-SAM for the given time span of contact.

Once a molecule has succeeded in adsorbing on to the CH$_3$-SAM, it is nevertheless hindered by the poor solvent from desorbing and traversing the SAM–water interface again. Thus higher desorption forces are to be expected as observed in the experiment. For the OH- and the COOH-SAM, equivalent considerations can be accomplished.

To corroborate this model, surface pressure measurements at the air–water interface were performed to determine the contribution of polyacrylic acid to the free energy of the surface. Assuming that, in a zero-order approximation, the free energy of the surface of the air–water interface is comparable to that of the interface between water and the methyl endgroups of a SAM, the surface tension of a saturated PAA solution was measured relative to that of pure water. The measured value of 30 ± 3 mJ m$^{-2}$ implies that PAA is in fact rather amphiphilic and that
a hydrophobic interaction may contribute a significant share to the adhesion on the SAMs. In a crude estimate, the surface energy of one polymer segment is given by

$$E = \frac{F_{\text{des}} \cdot a}{b \cdot a} = \frac{F_{\text{des}}}{b},$$

where $a$ denotes the length of one polymer segment and $b$ its width. Based on molecular models this width can be estimated to be of the order of 3 Å. Equating this expression to the experimentally determined surface energy, the resulting desorption force can be estimated to be $F_{\text{des}} \approx 10\, \text{pN}$. This corresponds very well with the experimentally determined difference between the desorption forces on the hydrophobic CH$_3$-SAM and the hydrophilic OH- or COOH-SAMs. Since Coulomb and van der Waals interactions on all surfaces are expected to be comparable, this difference must be attributed to hydrophobic effects and thus to a difference in the structure of water at the different SAMs.

Whereas strong polyelectrolytes are always fully charged independent of the pH of the solution, the line number charge density of weak polyelectrolytes like polyacrylic acid is pH-dependent. Measurements with constant NaCl concentration and varied pH did not show any measurable pH-dependence for all three thiol SAMs. This means that no significant change concerning the charge density on the polyelectrolytes and the SAM-surfaces takes place. Therefore all three SAM surfaces seem to be unaffected by the change of pH, including the COOH-SAM, which could become deprotonated and thus negatively charged.

With CaSO$_4$ in the solution, no pH dependence was found for the CH$_3$ and the OH-SAM, may be for the same reasons as discussed above. However, for the COOH-SAM, a jump in the desorption force could be observed at pH 8. This may reflect the deprotonation of the COOH-SAM which allows a complexation to bivalent Ca ions in the charged state that could increase the desorption force. The occurrence of salt bridges between the polymer and the COOH-terminated surface mediated by Ca ions is also probable.

5. Conclusions

Single-molecule force spectroscopy has been successfully applied to study the interaction between polyacrylic acid and SAMs with different headgroups. Investigations were undertaken with two different salts in solution, varied salt concentrations and varied pH on a CH$_3$-terminated, a OH-terminated and a COOH-terminated thiol SAM. It was found that the desorption force was independent of the salt concentration for both salts in the case of the OH- and the COOH-SAM. This led to the conclusion that in all these cases non-Coulombic contributions dominated the desorption force. For the CH$_3$-SAM, a repulsive electrostatic contribution was identified with NaCl in solution and an attractive electrostatic contribution with CaSO$_4$ in solution, respectively. Significant differences in further adsorption characteristics like length of the adsorbed molecules or frequency of the adsorption events could be explained if the quality of the solvent at the SAM–water interface was taken into account. Taking up this position, the solvent at the CH$_3$-SAM–water interface was found to be less favourable for the polyacrylic acid compared with the OH- and the COOH-SAM. The observed differences in the adsorption forces between the CH$_3$-SAM and the other two SAMs accorded well to the contribution that could be derived from the surface energy of polyacrylic acid that was determined by additional surface-pressure measurements of polyacrylic acid at the air–water interface. Variation of the pH showed no significant change in the desorption for all three SAMs when NaCl was present in solution. With CaSO$_4$, the same effect
was observed for the OH- and the CH3-SAM. For CaSO4 a jump in the desorption force to higher values was observed at pH 8. This could be due to the chelation of CaSO4 ions at the COOH-surface and salt-bridges between the SAM-surface and the polymer. Further investigations, using e.g. spectroscopic methods of the liquid–substrate interface, are required to gain deeper insight.

Acknowledgments

This work was supported by the Deutsche Forschungsgemeinschaft (DFG) through SFB 486. Fruitful discussions with Matthias Rief and Roland Netz are gratefully acknowledged.

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New Journal of Physics 6 (2004) 9 (http://www.njp.org/)
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