Immobilization of Dendrimers on Si-C Linked Carboxylic Acid-terminated Monolayers on Silicon(111)

Till Böcking[†], Elicia L. S. Wong^{††,§}, Michael James[§], Jolanta A. Watson[¶], Christopher L. Brown[¶], Terry C. Chilcott^{††}, Kevin D. Barrow[‡] and Hans G. L. Coster^{††,*}

[†]School of Physics, University of New South Wales, Sydney, NSW 2052, Australia

^{††}Biophysics and Bioengineering, Department of Chemical Engineering, University of Sydney, Sydney, NSW, 2006, Australia.

[§]Australian Nuclear Science and Technology Organisation, Lucas Heights Research Laboratory, Lucas Heights, NSW 2234, Australia

[¶]Nanoscale Science and Technology Centre, School of Science, Griffith University, Kessels Road, Nathan, QLD, 4111, Australia

[‡]School of Biotechnology and Biomolecular Sciences, University of New South Wales, Sydney, NSW 2052, Australia

* To whom correspondence should be addressed. Email: hcoster@usyd.edu.au

Abstract.

Poly(amidoamine) dendrimers were attached to activated undecanoic acid monolayers, covalently linked to smooth silicon surfaces *via* Si-C bonds. The resulting ultra-thin dendrimer films were characterised by X-ray photoelectron spectroscopy (XPS), X-ray reflectometry (XR) and atomic force microscopy (AFM). XPS results suggested amide bond formation between the dendrimer and the surface carboxylic acid groups. XR yielded thicknesses of 10 Å for the alkyl region of the undecanoic acid monolayer and 12 Å for the dendrimer layer, considerably smaller than the diameter of these spherical macromolecules in solution. This was consistent with AFM images showing collapsed dendrimers on the surface. It was concluded that the deformation arose from a large number of ~~amine~~amine groups on the surface of each dendrimer reacting efficiently with the activated surface, whereby the dendrimers can deform to fill voids while spreading over the activated surface to form a homogeneous macromolecular layer.

Keywords. Dendrimers; Organic monolayers; Silicon; Structural properties

1. Introduction.

Dendritic polymers (dendrimers) [1] are spherical molecules composed of a central core with repetitive branching units and a highly functionalised surface. There has been increasing interest in applying dendrimers to fields such as drug delivery [2], and immobilization of biomolecules, which has application to the development of chemical sensors [3]. Among various dendrimers, poly(amidoamine) (PAMAM) is one of the most frequently studied. It has a dense outer amine shell through a cascade type generation, exhibits great surface stability, which allows enhanced immobilization of biomolecules to PAMAM-modified surfaces. These aspects have been demonstrated through the successful attachment of enzyme (horseradish peroxidase) [4], protein [5] and ~~DNA~~deoxyribonucleic acid (DNA) [6, 7] to the PAMAM modified surfaces. The latter studies demonstrated that DNA microarrays based on dendrimer modified surfaces have an increased density of oligonucleotides when compared to conventional microarrays [6, 7].

A common approach to covalently attaching PAMAM dendrimers to a substrate surface is through self-assembled monolayers (SAMs) based on gold-sulphur chemistry [4, 5] or silane chemistry on glass [6, 7]. The drawback associated with the self-assembled monolayer on gold surfaces is the limited stability of the gold-sulphur bond, which is prone to desorption in strong alkaline and acidic conditions [8]. A more stable system is obtained *via* surface modification of silicon with Si-C linked monolayers, which are extremely robust and can withstand harsh chemical treatment and high temperatures. Silicon-carbon linked monolayers can be obtained *via* hydrosilylation reactions between alkenes and hydride terminated flat silicon surfaces [9-13]. Carboxylic acid-terminated monolayer can be formed conveniently by hydrosilylation of undecenoic acid and this approach has recently been used for the immobilisation of DNA [14]. There is great interest in the chemical modification of silicon

substrates because devices based on modified silicon can be integrated with existing semiconductor microfabrication technology.

In this study we have coupled PAMAM dendrimers to carboxylic acid functionalized Si-C linked monolayers. The structures of the resulting multi-layered surfaces were characterized using X-ray Photoelectron Spectroscopy (XPS), X-ray Reflectivity (XR) and Atomic Force Microscopy (AFM). A better understanding of the structure of covalently attached dendrimer layers will aid in the development of highly functionalized surfaces for the immobilization of biorecognition elements for biosensor and high-throughput screening applications.

2. Experimental Methods.

2.1 Materials. Ethanol, methanol, ethyl acetate and dichloromethane were redistilled. Semiconductor grade chemicals were used for cleaning (30% H₂O₂, 98% H₂SO₄) and etching (40% NH₄F solution) of silicon wafer wafers. Undecenoic acid (98%, Aldrich) was redistilled under reduced pressure and stored dessicated at 4°C. PAMAM dendrimer, generation 5 (G5), with amine surface groups was obtained from Aldrich.

2.2 Preparation of Si-C linked undecanoic acid monolayers. Si(111) wafer pieces (n-type, 1-10 Ωcm or p-type, 1-10 Ωcm) were cleaned using “Piranha” solution (concentrated H₂SO₄:30% H₂O₂, 3:1, v/v) at 90°C for 20-30 min and rinsed thoroughly with MilliQ-water (Millipore)-deionized water (18 Ω cm, Millipore purification system). *Caution: Acidic solutions of concentrated hydrogen peroxides react violently with organic materials and should be handled with extreme care.* The Si(111) surfaces were hydrogen-terminated by immersion into deoxygenated 40% NH₄F solution for 15-20 min. Undecenoic acid was placed into a Schlenk flask and degassed by five freeze-pump-thaw cycles. The freshly hydrogen-

terminated silicon wafer piece was placed in the alkene, and the flask was immersed in an oil-bath at 95-100°C. After 16-18 hours the flask was opened to the atmosphere and the sample was rinsed several times with dichloromethane, ethyl acetate and ethanol, immersed in boiling dichloromethane for 2 minutes and blown dry under a stream of high purity nitrogen.

2.3 Derivatisation of carboxylic acid-terminated surfaces. The terminal carboxylic acid groups were activated with an aqueous solution containing 50 mM (1-ethyl-3-(3-dimethylaminopropyl) carbodiimide (EDC) and 30 mM *N*-hydroxysuccinimide (NHS) for 1 hour, followed by rinsing with MilliQ deionized water and ethanol. Coupling of the PAMAM dendrimer ~~(generation 5)~~ (G5) to the activated surface was carried out in a 0.25 mM solution of the dendrimer in methanol for 4 hours. The sample was rinsed with methanol, MilliQ deionized water and CH₂Cl₂ and blown dry under nitrogen.

2.4 XPS measurements. XP spectra were obtained using an EscaLab 220-IXL spectrometer with a monochromated Al K α source (1486.6 eV), hemispherical analyzer and multichannel detector. The spectra were accumulated at a take-off angle of 90° with a 0.79 mm² spot size at a pressure of less than 10⁻⁸ mbarPa. XP spectra were fitted using XPSPeak 4.1 software with a Shirley background. The full-width-half maximum values of the peaks fitted to the carbon 1s envelope were between 1.2 and 1.4 eV.

2.5 X-ray reflectivity measurements. ~~Samples for X-ray reflectometry (XR) were cleaned in boiling dichloromethane for 2 minutes, blown dry under nitrogen and packaged under argon in glass vials for transport.~~ X-ray reflectivity curves were acquired using a Bruker D8 Advance Diffractometer in reflectometer mode. Cu K α (λ =1.54056 Å) radiation produced from a (40 kV) tube source was focused with a Göbel mirror, collimated with pre- and post-sample slits, and detected using an yttrium aluminium perovskite scintillation detector.

Reflectivity data are presented as a function of momentum transfer $Q_z = (4\pi \sin \theta)/\lambda$, where λ is the wavelength and θ is the angle of incidence of the X-ray beam onto the sample. The Parratt32 software [15] was used to fit model parameters to measured sets of XR data with linear background correction.

2.6 Atomic Force Microscopy (AFM). AFM images were recorded using a JEOL JSPM-4200 microscope with a 25 μm tube scanner. The analyses were carried out under air-ambient conditions (temperature of 23°C and 55% relative humidity). The probe (Veeco) consisted of a force-sensing/imposing ‘V’-shaped lever and an integral pyramidal Si_3N_4 tip, with a nominal spring constant, k_N , and tip radius of curvature of 0.06 Nm^{-1} and 40 nm, respectively. Topographical imaging was carried out at a constant force with a lever-imposed loading in the range 20-60 nN. The scanning rate in the fast-scan direction was ca. 3 Hz, and a typical image was composed of 500×500 pixels.

3. Results and Discussion

The steps for covalent immobilisation of generation 5 (G5) PAMAM~~G5~~ dendrimers with amine surface groups on Si(111) are shown in Figure 1. The hydride terminated silicon wafer was heated in neat undecenoic acid to produce a carboxyl-terminated alkyl monolayer that was then activated with EDC and NHS. This activation chemistry has been used extensively for the immobilization of amines on self-assembled monolayers because the coupling reaction occurs under mild conditions and with high yields [16]. Subsequently PAMAM dendrimers were coupled to the surface *via* reaction of ~~amino-groups~~amine groups on the dendrimer with the surface bound NHS esters. The chemical composition, structure and topography of the resulting organic thin films on silicon were analysed using XPS, X-ray reflectometry and AFM.

3.1 XPS Characterizations of Carboxylic Acid-Terminated Monolayers and Coupling of

PAMAM. Modification of Si(111)-H surfaces with undecenoic acid in a thermal ~~hydrodilylation~~hydrosilylation reaction at 95°C as described by Boukherroub *et al.* [17] was confirmed by XPS. In the survey spectrum the 1s binding energies of carbon and oxygen were detectable in addition to the silicon 2s and 2p peaks (Figure 2a). The absence of a sizable oxide peak between 102-105 eV in the silicon 2p narrow scan (Figure 2a, inset) confirmed the high quality of the surface. The XP narrow scan of the carbon 1s region (Figure 3, inset) was consistent with literature data showing a large peak at 285.0 eV due to the methylene carbons of the alkyl chain and a smaller signal at ~290 eV assigned to the COOH carbon. After activation with EDC and NHS a nitrogen 1s signal was detectable at ~402 eV in the survey scan (Figure 2b). Coupling of ~~amino-terminated~~amine-terminated PAMAM G5 to the monolayer *via* EDC/NHS activation was verified by the ~~appearance of the nitrogen 1s signal~~ and the large increase in the nitrogen, carbon and oxygen signals in the XPS survey spectrum shown in Figure ~~2b~~2c. Only low levels of oxide were detectable on the surface as evident from the silicon 2p narrow scan (Figure 2b and 2c, insets), similar to those observed on the undecanoic acid derivatised sample. This suggested that the coupling reaction did not degrade the quality of the surface significantly.

Figure 3 shows the carbon 1s narrow scan of the dendrimer modified Si-C linked monolayer, which was deconvoluted into ~~four~~five components. The peak at 285.0 eV accounted for most of the C-C linked carbons of the alkyl chains in the monolayer. The peaks at 285.4, 286.4 and 288.5 eV were assigned to the carbon bonded to the amide carbon (C-C(O)N), the C-N bonded carbons and the amide carbon respectively of the bound dendrimers. The observed binding energies were in reasonable agreement with those reported for the corresponding carbons in various Nylon polymers [18] Furthermore, the fit proposed here for the dendrimer

was consistent with the carbon 1s spectrum of a PAMAM G4 layer electrostatically adsorbed onto silicon dioxide reported by Bar et al. [19][20]. A small peak at a binding energy close to 290 eV characteristic of terminal carboxylic groups of Si-C linked undecanoic acid monolayers [17] ~~could not be~~ was also fitted to the carbon 1s signal. ~~This~~ The area of this peak assigned to unreacted carboxylic acid groups was reduced to less than half of the area expected for a COOH-terminated monolayer (whereby the attenuation of photoelectrons in the dendrimer layer was taken into account) suggestings that most terminal groups had been activated and reacted with the dendrimer to form amides. Consequently the resulting C(O)N and C-C(O)N carbons of the monolayer contributed to the respective peaks as shown in Figure 3. After subtraction of the monolayer components from these peaks, the area ratio of C-C(O)N:C-N: C(O)N was 1:2.8:1, close to the expected ratio of 1:3:1 for the dendrimer. ~~Based on further quantitative analysis of the XPS data it was estimated that the dendrimer to alkyl chain ratio was ~1:110 — 1:120.~~ The relatively high yield of amide linkages formed between the monolayer ~~on and~~ the dendrimers in our system was similar to the nearly complete reaction observed for chloroformate mediated linking of PAMAM dendrimers to mercaptoundecanoic acid SAMs on gold [3]. ~~However, it still appeared likely that some~~ The residual free carboxylic acid groups ~~were~~ present on the surface presumably as a result of from incomplete activation ~~or and~~ NHS ester hydrolysis during the coupling reaction [21].

Figure 4 shows the nitrogen 1s narrow scan of the dendrimer modified surface. The peak at 400.3 eV was relatively broad (full-width-half-maximum 1.4 eV), presumably due to the slightly different binding energies of the amides, tertiary and primary amines of the dendrimer. The absence of a N1s-nitrogen 1s binding energy at 402.3-4 eV characteristic of the NHS nitrogen (Figure 4, inset) suggested that the NHS moiety was removed from the surface as a result of the aminolysis and ~~possibly~~ competing hydrolysis reactions [22]. The removal of the NHS moiety was further confirmed by the absence of an oxygen peak at 535.4

eV (characteristic to the C-O-N oxygen of surface attached NHS ester groups [22]) in the oxygen 1s narrow scan (not shown).

3.2 Quantitative analysis of XPS data. The carbon 1s spectrum of the PAMAM modified surface was analyzed quantitatively to estimate the number of alkyl chains per dendrimer. First we identified component peaks exclusive to the dendrimer (C-N peak at 286.4 eV) and the underlying alkyl layer (C-C peak at 285.0 eV) respectively and divided the peak areas by the corresponding number of carbons. Then we corrected the peak areas for attenuation within the layered structure as outlined below and finally determined the ratio between the two species from these peak areas.

The intensity of photoelectrons, S_{dend}^C , from the carbons of the dendrimer is attenuated within the layer and the area of the corresponding XPS signal is given by

$$S_{dend}^C = C \sigma_C \rho_{dend}^C \lambda_{dend}^C A (1 - e^{-(d_{dend} / \lambda_{dend}^C \sin \theta)}) \quad (1)$$

where C is an instrument specific factor, σ_C is the sensitivity factor for carbon, ρ_{dend}^C is the density of carbon atoms per unit area in the dendrimer film, λ_{dend}^C is the inelastic mean free path of photoelectrons derived from carbon in the dendrimer film, A is the area probed by the X-ray beam, d_{dend} is the thickness of the dendrimer film and θ is the angle between the analyzer and the surface (90° for the instrument used in this study).

The area without attenuation is equal to

$$S_{dend}^{C,unattenuated} = C \sigma_C \rho_{dend}^C A d_{dend} \quad (2)$$

The measured peak area is then multiplied by the following ratio to account for the attenuation within the dendrimer film:

$$\frac{S_{dend}^{C,unattenuated}}{S_{dend}^C} = \frac{d_{dend}}{\lambda_{dend}^C (1 - e^{-(d_{dend} / \lambda_{dend}^C \sin \theta)})} \quad (3)$$

The intensity of the photoelectrons from the carbons of the monolayer was attenuated by the overlying dendrimer film as well as within the monolayer itself. The XPS peak area is given by

$$S_{ML}^C = C e^{-(d_{dend}/\lambda_{dend}^C \sin \theta)} \sigma_C \rho_{ML}^C \lambda_{ML}^C A (1 - e^{-(d_{ML}/\lambda_{ML}^C \sin \theta)}) \quad (4)$$

where ρ_{ML}^C is the density of carbon atoms per unit area in the monolayer, λ_{ML}^C is the inelastic mean free path of photoelectrons derived from carbon in the monolayer and d_{ML} is the thickness of the monolayer. The corresponding correction factor is then given by the following ratio

$$\frac{S_{ML}^{C,unattenuated}}{S_{ML}^C} = \frac{d_{ML}}{e^{-(d_{dend}/\lambda_{dend}^C \sin \theta)} \lambda_{ML}^C (1 - e^{-(d_{ML}/\lambda_{ML}^C \sin \theta)})} \quad (5)$$

The thicknesses of the monolayer d_{ML} and the dendrimer layer d_{dend} were determined by X-ray reflectivity (see below). We estimated the inelastic mean free path of carbon in the monolayer and dendrimer film to be $\lambda_{ML}^C = 41 \text{ \AA}$ and $\lambda_{dend}^C = 31 \text{ \AA}$ respectively. On the basis of these consideration we estimated that the dendrimer to alkyl chain ratio was $\sim 1:110 - 1:120$.

3.2-3 Characterization by X-ray Reflectometry. X-ray reflectometry is a technique that probes the structure of thin-films and interfaces normal to the surface. Reflectivity data as a function of momentum transfer, Q_z , for the undecanoic acid monolayer, the same layer after activation with EDC and NHS and after reaction with PAMAM G5 are shown in Figure 5. The overall increase in the thickness of the organic film on the silicon surface as a result of the step-wise modification was immediately apparent as the minima of fringes in these reflectivity curves moved to lower Q_z . Structural parameters such as thickness (d), electron density (ρ_{el}) and interfacial roughness (σ) of the individual layers on the silicon surface may be determined by refining a structural model for the reflectivity data. The curves for the undecanoic acid and the NHS ester terminated monolayer could be described adequately with

a single layer whereas a two-layer model was required to fit the XR data observed after immobilization of the dendrimer. The refined structural models based on these fitted reflectivity data are given in Table 1.

On the basis of XR data we estimated a monolayer thickness of 9-11 Å and an electron density of 0.32-0.38 e⁻ Å⁻³ for the undecanoic acid monolayer.[23] The electron density was expected to be slightly higher than that observed for unfunctionalised alkyl monolayers (~0.31 e⁻ Å⁻³) due to the terminal carboxylic acid group. We estimated a molecular coverage of the surface of $(3.1 - 3.7) \times 10^{14}$ molecules cm⁻² which is slightly lower than the predicted maximum coverage of the Si(111) surface with alkyl chains (3.9×10^{14} molecules cm⁻²) [10].

After activation of the monolayer with EDC and NHS the monolayer thickness increased to 13 Å, which was consistent with the presence of the NHS ester moiety on the monolayer surface. The NHS ester group was not resolved from the alkyl base layer as a two-layer model did not improve the fit to the reflectivity data. The electron density of the NHS ester activated monolayer was considerable higher than for methyl terminated alkyl layers, which was expected because of the presence of the electron-rich headgroup.

The XR data of the dendrimer modified surface required a two-layer model to account for the vastly different electron densities between the alkyl and dendrimer layers. The refined electron density of the alkyl sublayer was 0.32 e⁻ Å⁻³, which agreed well with the average electron density of 0.31 e⁻/Å³ for a series of alkyl monolayers on Si(100) and Si(111) [10, 24]. Thus the bottom layer in this model was interpreted as the alkyl chain (C1-C10) of the undecanoic acid layer whereas the electron-rich terminal carbonyl group was considered to be part of the grafted macromolecular layer of dendrimers. The length of the sublayer (10 Å) was within the range expected for a C10 layer. The thickness of the covalently linked layer of PAMAM G5 dendrimer was found to be 12 Å, which was a factor of 4-5 smaller than the

diameter of the dendrimer in solution (54 Å). This result was in agreement with previous observations that dendrimers deposited onto substrates are highly compressed along the surface normal [3, 25-27]. It has been shown that the thickness of PAMAM dendrimers electrostatically adsorbed onto silicon dioxide as determined by X-ray reflectivity was reduced more than twofold [25]. The ellipsometric thickness of PAMAM monolayers covalently linked *via* amide bonds to mercaptoundecanoic acid SAMs on gold was also approximately twofold lower for G0-G4 and approximately threefold lower for G6 and G8 when compared to their hydrodynamic diameter [3]. Dendrimers are thought to change their conformation to maximise the number of links between the terminal groups and the surface [27]. A theoretical study of the adsorption of dendrimers onto surfaces predicts that the degree to which the molecule flattens down and spreads out on the surface is dependent on the strength of the interaction between the surface and the “sticky segments” of the dendrimer, i.e. interactions mainly with terminal functional groups [28].

The roughness for the G5 monolayer determined from the reflectivity data was 5 Å, which was expected from the ovoid shape of the surface adsorbed dendrimer. This value was consistent with the microroughness of 4 Å determined by scanning probe microscopy for a PAMAM monolayer electrostatically adsorbed onto silicon dioxide [25]. The question arises whether complete monolayer coverage could be achieved in our study since the molecules were no longer mobile after the first covalent links with activated surface groups had been formed. It has been found that the surface coverage of dendrimers adsorbed onto gold surfaces measured by mass titration is lower for G8 (76%) than for G4 (88%) [29]. This was thought to be the consequence of stronger adsorption and reduced surface mobility for larger generation dendrimers due to a larger contact area and number of ~~amino-groups~~amine groups available for interaction with the gold surface [29]. Random sequential adsorption models for the irreversible adsorption of hard spheres to surfaces predict that the jamming coverage (when

no further objects can be placed onto the surface) is equal to 55% [30]. However, the surface coverage for dendrimers in this case would be expected to be higher because the flexibility of the dendrimer would enable it to deform to fill voids as it spreads out on the surface. The surface area/molecule occupied by the dendrimer particles determined from the X-ray reflectivity data was approximately $3400 \text{ \AA}^2$ [31]. ~~Therefore On the basis of the structural parameters determined for the dendrimer layer and the underlying alkyl layer we estimated that the ratio between alkyl chains and the dendrimers these species~~ on the surface was approximately ~~130:1 — 140:1 110 — 1:140~~, which was in reasonable agreement with the ratio estimated from XPS data (see above).

3.3-4 Characterization of PAMAM modified surfaces by AFM. AFM using a Si_3N_4 cantilever was used to obtain the surface topographical image of the silicon functionalized surface and to further investigate the coverage of the surface with dendrimers. AFM images of the undecanoic acid modified sample before attachment of PAMAM revealed the topography of the underlying Si(111) surface with its typical flat terraces separated by mono-atomic steps. The PAMAM dendrimers were attached to the NHS activated undecanoic acid monolayer surface in methanolic PAMAM solutions. Regions on the PAMAM modified sample imaged by AFM showed that the surface was covered with a relatively homogeneous layer of dome-shaped features with a distribution of sizes (Figure 6). The AFM image suggested that the layer was densely packed such that the tip mainly did not contact the underlying substrate surface. The higher resolution AFM height image ($500 \text{ nm} \times 500 \text{ nm}$) (Figure 7) shows a topographical image and height profile of two of the larger features. These features protruded ~~up to~~ $\sim 1.5 \text{ nm}$ above the surface and had a diameter of ~~up to~~ $\sim 50 \text{ nm}$ [32]. The diameter of these features was too large for a single dendrimer and thus they were attributed to clusters of a few dendrimers immobilized on the surface. ~~The cross-section analysis suggested that the PAMAM molecules (even in the clusters) lost the spherical structure they have in solution and~~

~~had a hemispherical configuration on the modified silicon surface. This was attributed to the fact that the dendrimer has a large number of amino groups that are available to undergo carbodiimide coupling to the NHS activated surface. Such multiple attachments to the surface would cause the originally spherical dendrimer to collapse on the surface. In conjunction with the average thickness of ~1.2 nm of the dendrimer film determined by XR, the AFM images suggested that most of the PAMAM molecules had lost their spherical shape they have in solution and were collapsed onto the surface to form a relatively compact and densely packed film, whereby a few larger dendrimer clusters protruded from this film. The formation of a largely collapsed dendrimer film was attributed to multiple attachment points between the dendrimer and the underlying monolayer, whereby not all surface groups of a dendrimer would have reacted with the surface.~~ This observation of collapsed dendrimers was also consistent with many previous AFM studies showing that PAMAM molecules had highly flattened configurations when adsorbed to mica [33-35] or gold [36] or covalently attached to anhydride plasma surfaces [26]. The surface morphology with some features of larger sizes resolved by AFM in this study may be mirrored in the relatively large roughness of the top surface of the sample determined from the XR data. It is important to note that X-ray reflectometry determines the average layer thickness across the entire sample whereas AFM typically probes only small regions on the sample.

4. Conclusion.

PAMAM G5 dendrimers were attached to activated carboxylic acid-terminated monolayers, resulting in covalently linked and densely packed layers as evidenced by XPS and XR and AFM. The dendrimers were flattened onto the surface, yielding smooth macromolecular monolayers with a number of isolated clusters of dendrimers. We conclude that the ~~amino-~~ amine groups on the dendrimer surface react efficiently with the activated groups on the monolayer, causing the collapse of the usually spherical molecule. The ratio of alkyl

chains to dendrimers estimated from XPS and XR data were in reasonable agreement and in the range of $\sim 110:1$ - $140:1$.

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- [31] The area per molecule is given by $A = N_e / (d\rho_{el})$, where d is the measured film thickness, and N_e is the number of electrons per molecule in the monolayer.
- [32] We estimated that the width of the cluster is close to 30 nm when tip convolution is taken into account.
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Figure 1. Route for the modification of Si(111) with PAMAM G5.

Figure 2. XP survey spectra of Si(111) (a) modified with an undecanoic acid monolayer and (b) after activation with EDC/NHS and (c) after coupling of PAMAM G5. The insets show the silicon 2p narrow scans of the respective surfaces.

Figure 3. (a) XP narrow scan of the carbon 1s region of Si-C linked undecanoic acid monolayers reacted with PAMAM dendrimers. The residual of the fit is shown below the spectrum. The inset shows the carbon 1s narrow scan of the undecanoic acid modified silicon surface before reaction with the dendrimer. (b) Chemical structure of the underlying monolayer and (c) repetitive unit of PAMAM.

Figure 4. XP narrow scan of the nitrogen 1s region of the Si-C linked undecanoic acid monolayers reacted with PAMAM dendrimers. The inset shows the nitrogen 1s region of the EDC/NHS activated surface before reaction with the dendrimer. The dotted lines mark the positions where the nitrogen of surface bound NHS ester groups occurs.

Figure 5. X-ray reflectivity curves of modified Si(111) surfaces: (a) undecanoic acid monolayer, (b) undecanoic acid monolayer after activation with EDC/NHS, (c) undecanoic acid monolayer after activation with EDC/NHS and coupling of PAMAM G5. The solid lines represent model fits to the reflectivity data. Curves (b) and (c) were offset for clarity.

Figure 6. AFM 3-D topographical image ($1.5\ \mu\text{m} \times 1.5\ \mu\text{m}$) of PAMAM G5 dendrimers reacted onto activated undecanoic acid monolayers on Si(111).

Figure 7. AFM topographical image (500×500 nm) of dendrimer clusters. The inset shows a height profile.

Table 1. Refined structural parameters for molecular films on Si(111) shown in Figure 5.

Table 1.

	Si-(CH ₂) ₁₀ COOH	Si-(CH ₂) ₁₀ COO-NHS	Si-(CH ₂) ₁₀ CONH- PAMAM
PAMAM			
d (Å)			12(1)
ρ_{el} (e ⁻ Å ⁻³)			0.41(2)
σ (Å)			5(1)
Monolayer			
d (Å)	9-11	13(1)	10(1)
ρ_{el} (e ⁻ Å ⁻³)	0.32-0.38	0.43(4)	0.32(1)
σ (Å)	3(1)	3(1)	2(1)
Silicon			
σ (Å)	3(1)	2(1)	3(1)

Figure 1.

Figure 2.

Figure 3.

Figure 4.

Figure 5.

Figure 6.

Figure 7.