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Effect of Water on Silanization of Silica by Trimethoxysilanes

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Water has a big influence on the mechanism of monolayer formation and therefore on the structure of monolayer that can be obtained on a silica surface. Trimethoxysilane in the absence of water can form submonolayers with only one siloxane bond binding functional tails with the surface. We show here that hydrolysis of the remaining methoxy groups on the initial immobilized silane layer by water and followed by another silanization yields enhanced surface density of silanes with improved lateral polymerization and without complications of vertical polymerization. The improved technique allows surface concentration of coumarin dye molecules of 2.7×10^{14} cm^{-2}, almost 1.3 times higher than what is possible without water treatment.

Introduction

The silane coupling agents are widely used now for surface modification. Silane-based self-assembly methods can be applied on such surfaces as silica, glass, and metal oxides, where surface hydroxyls can react with proper chemically active groups, typically methoxy- or chlorosilanes. Such silane-based linkers have taken on special significance in recent years in the field of surface modification.[1,2] It is believed that in many cases the self-assembled monolayers of trifunctional silanes form close-packed and well-aligned molecular films.[3,4]

Well-ordered assemblies and monolayers of silanes are desired and can be used in many applications.[2] Many efforts were made to identify and optimize the factors affecting silanization and monolayer formation, but there are still some unclear peculiarities which are the subject of speculation in many studies. The reaction conditions such as temperature, water content, nature of a silane, and type of substrate have influence on the rate of reaction and the assembly structure.[3−9] Different types of the surface assembly structure can be obtained upon slight change in reaction conditions. Two types of polymerization, horizontal (lateral) and vertical, or their combination, can accompany the attachment.[10] Properties of resulting layers affected by the quality of assembled structures strongly depend on reaction conditions. These properties include orientation of molecules with respect to the surface (tilt angle), thickness and density of resulting films.

Despite the efforts, details of the mechanism of self-assembled monolayer formation remain elusive and controversial. Dependence of the monolayer quality on various particulars in reaction conditions, such as silanizing agent, humidity, temperature, etc., have been reported.[7,11] The least complicated silanizing agents, which have a single functional group (methoxy- or chloro-) form a single Si−O−Si bond with the surface,[12] while bi- and trifunctional silanes bring more complexity to the mechanism, offering a possibility of multiple bonding to the surface as well as lateral polymerization between neighboring silanes.

Two models of high ordered monolayers formation are usually discussed in connection with tri- and difunctional silanes. The first is the continuous growth model in which a liquidlike disordered film is formed initially and then it further improves to a well-ordered monolayer with increasing total coverage.[8] The second model implies formation of close-packed islands from the beginning. Initially the islands are separated by uncovered regions and grow by lateral polymerization until full coverage is reached.[8,9,11] It has been reported[8] that the two mechanisms may vary depending on reaction conditions even for the same reagent. The authors[8] reported that water content in silane solution affects the mechanism of layer formation changing it from continuous growing at low moisture to the island growth at high water content. All these suggest that increasing water content causes silane polymerization to start in solution and later deposit on the surface.

The procedure that we use here to evaluate quality of the silanization is indirect—instead of silanes, we investigate optical absorption of coumarin tethered to amino tails of self-assembled silanes. We reported previously,[13] using the same technique, that the highest density of coumarin molecules obtained on silanized for 3 min silica surfaces can exceed 10^{14} cm^{-2}. This number is noticeably less than the surface concentration of hydroxyl groups on

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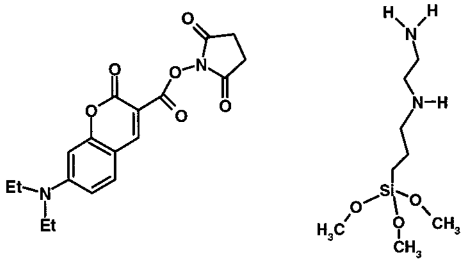
Chart 1. Structures of the Compounds Used in This Study and Their Abbreviations: 7-Diethylaminocoumarin-3-carboxylic Acid Succinimidyl Ester (Coumarin); *N*-[3-(Trimethoxysilyl)propyl]ethylenediamine (AENPS)



Coumarin AENPS

silica, which has been reported to be 5 × 10^{14} molecules per cm^2.[7,14–17] In a number of studies it was shown that the surface concentration of amines achieved in analogous procedures amounted to ca. $n_{NH_2} = 2 \times 10^{14}$ cm^{-2}.[16,18] The authors[18] showed that the surface concentration of silanes grows rapidly in the first few minutes and saturates at a concentration of 2.1 × 10^{14} cm^{-2}.

In this paper we further optimize the silanization procedure by studying effects of the silanization time and water treatment. We show that silanization by trimethoxysilanes in dry conditions cannot produce monolayer films with the highest density due to steric hindrance. Film quality can be improved by a two-step process where two silanizations are separated by a water treatment. It increases surface density to 2.7 × 10^{14} cm^{-2} and allows lateral polymerization between silanes while avoiding vertical polymerization at the same time.

Experimental Part

Materials. Polished 25 × 13 mm^2 quartz slides of 0.3 mm thickness, from Quartz International, were used as substrates. "Coumarin", 7-diethylaminocoumarin-3-carboxylic acid, succinimidyl ester, was purchased from Molecular Probes (D-1412). Aminosilane, *N*-[3-(trimethoxysilyl)propyl]-ethylenediamine (AENPS), was purchased from Aldrich. The structures of these compounds are shown in Chart 1. The solvents acetone and DMSO were of HPLC grade from Aldrich and were used without further purification.

Procedure. (a) Preparation of Substrates. Initial cleaning of slides was done in 1:1 MeOH/HCl solution for 30 min with subsequent rinsing in copious amounts of deionized water. After that, the slides were heated in concentrated H_2SO_4 for 2 h and then rinsed in deionized water and boiled in it immediately prior to silanization. This preparation procedure has proved to be effective; the slides were numerously recycled without noticeable deterioration, as confirmed by reproducible absorption spectra, and the preparation procedure was found to be most effective in removing surface contaminants.[19]

(b) Silanization. Trimethoxysilanes spontaneously react with surface hydroxyl groups so that the self-assembled layer forms.[20] The silanization of slides was performed using a 2% v/v acetone solution of a trimethoxysilane. In all cases, the slides were treated in the solution by complete immersion for a desired time. If washing in water was supposed to be made, slides were immersed in the silane solution for 3-min time intervals with washing in water and acetone in between. At the end, silanized slides were washed in acetone, dried at ca. 100 °C on a hot plate for 5 min, and cooled for approximately 5 min.

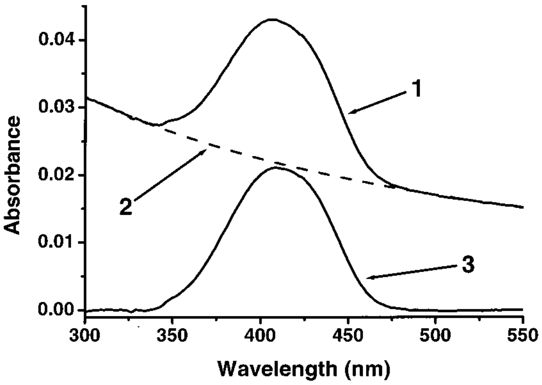


Figure 1. Illustration of the background subtraction procedure in absorption spectra: 1, original spectrum of a slide with immobilized coumarin; 2, the best fit to the baseline of spectrum 1 using function of wavelength λ, $f(\lambda) = c_1/c_2 + \lambda + c_3$, with three variables, c_1, c_2, and c_3, in the range $\lambda > 490$ nm and $\lambda < 330$ nm; 3, the "processed" spectrum is a result of subtracting curve 2 from curve 1.

(c) Staining. The staining of the silanized slides by coumarin-ester was performed only on one side. For that purpose, a drop of 0.6 mM DMSO solution of coumarin was placed between the slide and the surface of a glass beaker. Reproducible results for the amount of immobilized coumarin were achieved when 2 h was provided for completing this step. The stained slides were washed in acetone and dried on a stove at ca. 100 °C. No noticeable deterioration of the surface concentration of coumarin within a month after staining was observed when stored under dark, dry conditions.

(d) Measurements of Absorption Spectra. Absorption spectra were measured using a Perkin-Elmer Lamda 40 UV/VIS spectrometer equipped with a polarizer. A stained slide was placed vertically in a 1 cm quartz cuvette at close to a 45° angle with respect to the incident light. Two linear polarizations of excitation light were used: vertical and horizontal. Typical spectra are shown in Figure 3.

Vertical polarization in the described geometry corresponds to measurement of optical absorption with polarization parallel to the surface (||) and will be named as such from now on. Absorption polarized perpendicular (⊥) to the surface substrate was calculated as the difference between the double horizontal absorption and the vertical absorption spectra for a given stained slide.

$$A_{\parallel} = A_v; \qquad A_{\perp} = 2A_h - A_v \tag{1}$$

Bare slides showed some apparent absorption, as a background, which was each time simulated as a smooth function of wavelength, $f(\lambda) = c_1/(c_2 + \lambda) + c_3$, this background signal was subtracted from each spectrum of a stained slide (Figure 1).

Surface concentration of coumarin, n_s, was calculated from the average absorbance, A, using Lambert–Beer's law and the extinction coefficient of coumarin, $\epsilon = 47\,000$ M^{-1} cm^{-1}:[13]

$$n_s\,(\mathrm{cm}^{-2}) = 6 \times 10^{20}\,\frac{A}{\epsilon} \tag{2}$$

where the average absorbance, A, quantifies the concentration of molecules independent of their orientation using parallel and perpendicular polarized absorptions:

$$A = \frac{2A_{\parallel} + A_{\perp}}{3} \tag{3}$$

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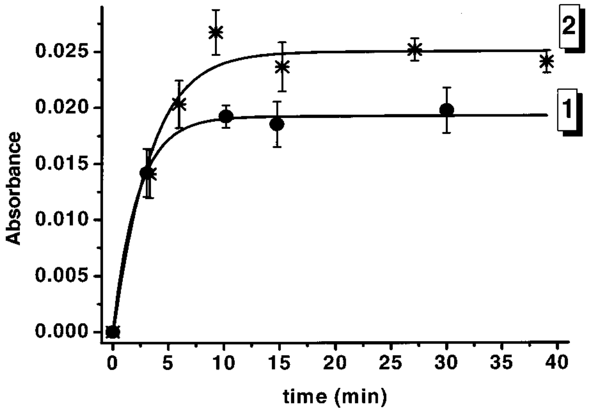


Figure 2. Variation of absorbance for immobilized coumarin as a function of time of treatment by aminosilane solution: (●) dry solution; (∗) the same solution but with washing in water after each 3 min of silanization. Each point is a result of averaging of at least two samples with error bars representing the spread. Lines 1 and 2 are drawn as guides to the eye.

Results and Discussion

Surface silanization with aminosilanes produces amines available for immobilization of coumarin. That surface concentration of amines defines the concentration of immobilized coumarins after staining. The latter step can be kept long enough to allow all accessible amines reacting in full, as was in our experiments. The silanization step, on the other hand, is more susceptible to duration, variation in temperature, moisture, and other less controllable factors. First we consider how duration of silanization in "dry" silane solution affects the surface density. Figure 2 demonstrates that absorption of immobilized coumarin rapidly increases with time of silanization (curve 1). Within 3 min of silanization the absorption reaches 80% of the maximum value. The maximum absorption of 0.016 on one surface corresponds to the surface density of 2.1×10^{14} cm^{-2}. This number is almost identical to what was reported for a monofunctional silane monolayer which can form only one Si−O−Si bond with the surface.[16] This coincidence naturally suggests that trimethoxysilane in the absence of water forms a sparse monolayer, where only a single methoxy group per silane reacted with the surface hydroxyl and the other two methoxy groups remained unreacted. The same conclusion was previously reached based on the linear dichroism measurements in combination with conformational analysis.[13]

There are two consequences of leaving two out of three methoxy groups unreacted with surface hydroxyls: (1) these groups cannot react with other silanes and (2) they partially screen neighboring surface hydroxyls from oncoming silanes as well. If water is present in the solution, hydrolysis of some methoxy groups allows their polymerization in the solution or start polymerization from the surface immobilized silane, the so-called vertical polymerization. We suggest a different approach here and hydrolyze methoxy groups of *only* immobilized coumarins *after* initial silanization and washing. The resulting hydroxyls resolve the two problems numbered above as well as eliminate the unwanted vertical polymerization. The procedure is described by the cartoon in Scheme 1. Trace 2 in Figure 2 demonstrates that the approach works. Alternating silanization and washing in water enhances the maximum surface density of silanes by at least 30%. Moreover, it appears that one treatment in water produces as large an effect as a few consecutive ones and the vertical polymerization, which could in theory start from unreacted methoxy groups after the second silanization, does not take place. We consider the effect of water in Figure 2 to be the low limit because (1) there are probably some traces of water present in the "dry" solution, even though water concentration should be extremely low (typical humidity in Las Cruces is below 15%) and (2) the steric hindrance for coumarins might prevent them from reaching even higher surface concentration, i.e. it might not reflect the surface amine concentration accurately (surface density of a close-packed monolayer of coumarin cannot exceed 3.7×10^{14} cm^{-2}). Nevertheless, the effect of hydrolysis is apparent, reproducible, and may be put to use.

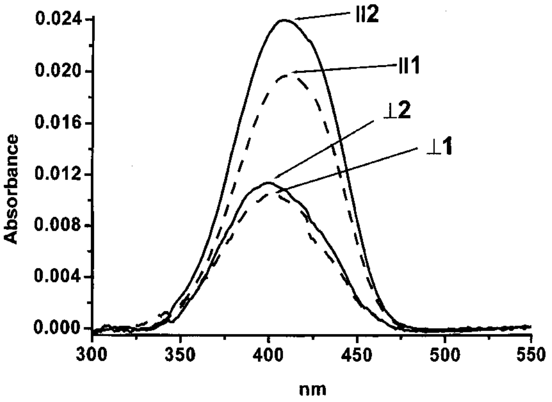
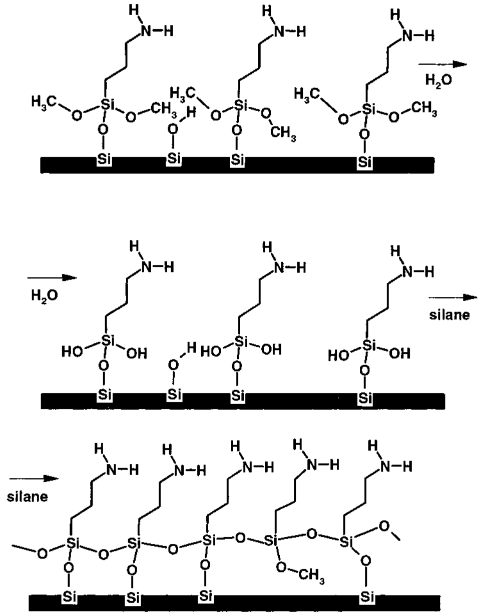


Figure 3. Absorption spectra of slides at different light polarizations with respect to the surface (⊥ perpendicular and || parallel polarizations): 1, for the point at 30 min on curve 1, Figure 2; 2, for the point at 39 min on curve 2, Figure 2.

Scheme 1. Two-Step Silanization with Intermediate Water Treatment



The absorption anisotropy provides information about molecule orientation with respect to the surface through the difference in signal intensity and their mutual interaction through the spectral shift.[13] The latter is related to the density and molecular alignment.[13] The increased density after water treatment does result in an additional hypochromic shift as expected for molecules on the surface, but the effect is small (see Figure 3). Moreover, the anisotropy in intensity does not change at all and corresponds in both cases to a small preference for molecule orientation parallel to the surface. As was previously concluded for these self-assembled monolayers,[13] molecule orientation in such films is close to random on a half sphere and does not change dramatically with increased packing. In a film with randomly oriented molecules dramatic variations in intermolecular interactions (proportional to $1/r^3$) take place for noticeably lower densities. Molecules tethered to the surface by a lengthy tail can extend in all directions limited by the tail length and its flexibility thus “controlling” an area exceeding its size. When the surface density overcomes that for overlapping on average these areas of control between neighboring molecules, the intermolecular interaction becomes not as strong due to molecules random entanglement. The random orientation of molecules, at least in part, is caused by an irregular substrate (fused silica). It would be interesting to investigate whether if ever this random orientation can be transformed into more regular packing by using a monocrystalline substrate.

The alternative explanation to the water treatment effect might imply that, instead of steric hindrance, it could arise from different ways silanes physisorb to the surface. Aminosilanes are known for forming hydrogen bonding with surface hydroxyl using all four active groups: three methoxy groups and amine. It was claimed[21] that, depending on aminosilane concentration, orientation of physisorbed silane can change from primarily a methoxy group attached at low concentration to amine bonded orientation at high concentration. One may argue that the additional treatment with water primarily eliminates improperly oriented aminosilanes, which did not succeed in forming a Si−O−Si bond with the surface. That could be a part of the story, but since dry organic solvents did not produce such an effect, we have confidence in correctness of our interpretation.

Besides just improving the density of immobilized silanes on an oxide surface, there are numerous possible applications for such a new approach in silanization. For example, by using bulkier alkoxy groups, the effect of hydrolysis can be increased beyond the currently realized 30%, thus making it possible to alternate active groups and molecules at the silane tails in the two steps of silanization (before and after water treatment).

Conclusions

An influence of water (and staining time) in silanization of silica by trimethoxysilanes was studied, and a procedure of silica substrate silanization was optimized to get a more dense layer with higher quality. Most of the monolayer forms within the first several minutes and the rest of it then gradually builds up. Maximum coverage of coumarin when time is the only factor having influence is achieved to be $(2.1 \pm 0.2) \times 10^{14}$ cm^{-2}, which is the same for silane with only one functional group. The formation of a *monolayer* completes via covalently attached silanes with only one Si−O−Si bond with the surface and other two methoxy groups remaining intact. During silanization water comes to play in later stages when hydrolysis of intact methoxy groups becomes crucial for further polymerization. Only lateral (horizontal) polymerization occurs in silanization steps after washing in water of silanized substrates. The improved method of silanization and staining of silica substrates allows a surface concentration of dye molecules (coumarin) as high as $(2.7 \pm 0.2) \times 10^{14}$ cm^{-2}.

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