

Fabrication and Visualization of Metal Ion Patterns on Sensing Fluorescent SAMs with AFFM

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INTRODUCTION: Developing miniaturized patterned substrates at μ and sub- μ scale with optical readouts is very important for chemical and biological sensors.[1,2] The combination of optical and scanning probe microscopy and fluorescent self-assembled monolayer technologies will enable the development of sensors of great versatility. In this study, we employed our home-built atomic force fluorescence microscope (AFFM), a combination of atomic force microscope and confocal fluorescent microscope, to write calcium metal ions onto the sensitive fluorescent self-assembled monolayer (SAM) by dip pen nanolithography (DPN), and visualize the modulations of fluorescence intensity resulting from the presence of Ca^{2+} immediately.

METHODS: The sensitive fluorescent SAM, TM1, coated on glass slides was fabricated basically following the protocol published in our group previously except for small modifications [2]. Figure 1 illustrates the procedures of preparing TM1 substrates. The amino terminated SAM were prepared in dry toluene solution of N-[3-(trimethoxysilyl)propyl]ethylenediamine (TPEDA) first. After that, the fluorophore and binding group, 5-(and-6)-carboxytetramethylrhodamine, succinimidyl ester (5(6)-TAMRA, SE) and hexanoyl chloride, were immobilized onto the TPEDA SAM randomly to yield the TM1 SAM.

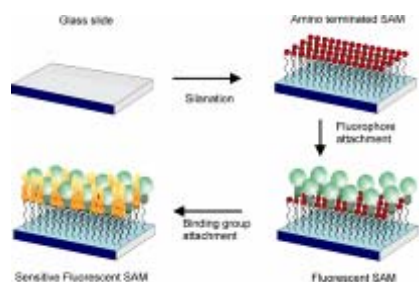


Fig. 1: Schematic representation of the preparation procedures of sensitive fluorescent self-assembled monolayers.

Commercial Si_3N_4 AFM cantilevers with nominal spring constant of 0.05 N/m were used. Right before DPN experiments, cantilevers were rinsed with ethanol and dried under a nitrogen stream gently. The cantilevers were then exposed under UV light for 30 minutes. Subsequently, the cleaned

cantilevers were immersed into the ink solutions (10^{-1} M ethanol solution of the per-chlorate salts of calcium) for 5 minutes and then dried in air. The DPN experiments were all carried out in contact mode. The load applied by the AFM tip to substrate was kept between 10 and 30 nN to avoid damaging of the SAM.

RESULTS: Figures 2 exhibits four horizontal bright lines of 18 μm in long generated by deposition of Ca^{2+} ions onto TM1 SAM with a scan rate of $1.8 \mu\text{m s}^{-1}$ for 2, 4, 6 and 8 minutes individually (from (1) to (4)). The width of each line is approximately 0.6 μm . The line width shown in the image does not correspond to the actual deposited line width because confocal fluorescence microscope has a lateral resolution of the half wavelength value of the excitation beam.

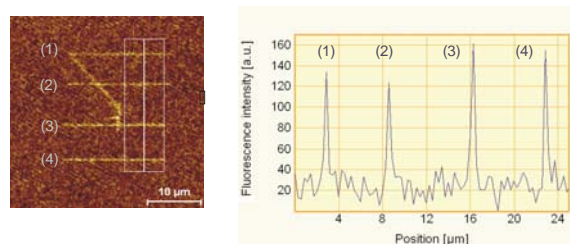


Fig. 2: Fluorescent image of a TM1 SAM acquired from AFFM right after the deposition of Ca^{2+} lines.

DISCUSSION & CONCLUSIONS: We demonstrate that it is possible to transfer Ca^{2+} onto TM1 SAMs observing the enhancement of fluorescent intensity instantaneously using AFFM. This study may allow us to explore in more detail the role of different inks, ink diffusion, and other fundamental aspects of DPN.

REFERENCES: ¹R. Kassies, K. O. Van der Werf, et al (2005), *J. Microscopy* **217**: 109-116 ²L. Basabe-Desmonts, J. Beld, et al (2004), *J. Am. Chem. Soc.* **126**: 7293-7299.

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