Patterning Function and Shape for Application from Microelectronics to Biotechnology

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Introduction

Lithography and other patterning processes are powerful tools catalyzing many developments in science and engineering. The controlled formation of nanometer scale structures in 2 and 3 dimensions is therefore of increasing importance in many applications ranging from biotechnology to nanotechnology. This presentation will discuss new approaches for the construction of small-scale (a few tens of nm) structures using both 1- and 3-photon processes. Several approaches to fine feature lithography including the use of molecular glasses will be described. Such small scale structures can be used in a variety of biological applications including cell function and will be described.

As shown in Figure 1, the remarkable progress in nanofabrication made possible by the microelectronics industry has led to the ability to create small structures overlapping many length scales of biological systems.

A Matter of Scale

![Figure 1. Progress in nanofabrication compared to length scales of biological systems.](image)

Fabrication techniques can be divided into two major categories: "top-down" and "bottom-up" approaches. The former includes well-known electron beam or photolithographic techniques [1] currently used in microelectronics, and the latter focuses on chemical design and includes the use of self-assembling materials for producing surfaces ordered at the nanoscale level. The bottom-up approach allows for a strict control on the physicochemical properties of a surface and represents a versatile method for the production of a variety of surfaces. It also shows promise for sub-100 nm feature fabrication [2].

Typically, pattermable materials are rigid, glass forming organic materials to enable maximum resolution of resulting features. In the growing field of nanobiotechnology, soft materials such as hydrogels have been found to be biocompatible surfaces for applications in drug delivery [3], tissue engineering [4] and specific cellprotein interactions [5]. Elastomers have been proven to be useful materials for fabricating microfluidic channels [6]. Elastomers have been proven to be useful materials for fabricating microfluidic channels [6]. The patterning of such soft materials in two-dimensions has already been achieved by conventional 2D lithography [7-8]. However, to achieve the full potential of these materials, patterning in three-dimensions is necessary as one can add a higher level of functionality. This presentation will discuss fabrication strategies for making small-scale structures with controlled surface functionality with the goal of interactions with biomolecules and living cells.

Experimental

Materials

2-Hydroxy ethyl methacrylate (HEMA) and poly(ethylene) glycol diacrylate (PEGDA) were purified by passage through a MEHQ anion removal column. 7-benzoesulfon-2-yI-9-
the PMMA was then removed from the unexposed regions and titanium followed by gold was evaporated onto the substrate. Using a lift-off method, PMMA/gold was removed leaving gold posts on the silicon. Figure 3 shows SEM images of gold posts (height of 50 nm) on silicon with feature size of 1 μm.

**Two-Photon Lithography.** Figure 5a,b shows optical images of 3 cubic structures (25 μm x 25 μm x 25 μm) formed on a neural prosthetic device (University of Michigan, USA). These hydrogel structures were then loaded with a fluorescently labeled Dextran (molecular weight 10 kDa) (Figure 5c). Dextran serves as a model for the nerve growth factor (NGF) whose molecular weight is around 26 kDa.

**Swelling Studies.** A reciprocal relationship between cross-link density and swelling of gels was observed. As the laser power decreased, a gel of lower cross-link density resulted. This was verified from fluorescent images of 3D microstructures (Figure 6). Approximately 15% swelling was observed after addition of water when waffle shaped objects were produced using laser intensity of 3 mW (compare a and b).

**Conclusions**

Using high-resolution lithography, precisely patterned gold regions were formed on silicon. To these sites DNP-ligand modified alkylthiols were tethered and tested for molecular recognition with anti-DNP antibodies as well as for visualizing stimulated cellular response. Using two-photon lithography, hydrogels were successfully microfabricated on a neural prosthetic device. Different types of hydrogel microstructures were fabricated and tested for their swelling properties. On releasing growth factors from these hydrogels PC12 cells extended their processes towards the device. These in vitro results demonstrate that neuronal-like cells are attracted to the device and indicate that in vivo testing is justified. Both these research areas demonstrate the importance of patterning and formation of the appropriate chemistry and topography of the material systems to elicit the most favorable response from biological systems.

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**References**