A Multi-Electrode Array System for Patterned Neuronal Network

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Abstract

Presently, multi-electrode array (MEA) have been widely used as a non-invasive in vitro recording method to study excitable cells and tissues, such as peripheral neurons, stem cells, or sliced brain tissue. The limitation for a conventional MEA system is the accuracy of the response of an individual-cell-based neuronal network. Here, we have developed a novel MEA system with the capability to culture neuron cells in predefined patterns on top of electrodes which can record the signals of excited neuron cells.

Introduction to Research

The fabrication process is shown on Figure 1. A thin film of Cr/Au/Cr electrodes and their interconnects are patterned on top of a fused silica wafer by metal lift-off with image reversal processing. First, Shipley 1818 photoresist was spin-coated at 3000 rpm to get a 2.2 µm thick film. After soft baking at 90°C for 60 s, a contact aligner was used to expose the photoresist with a dose of 80.4 mJ/cm². This step was followed by baking in ammonia for 90 min in an image reversal oven, where the whole substrate was flood-exposed for 60 s. After developing in MF321 developer for 90 s, oxygen plasma was used to remove the photoresist scum residue. Then, 100 Å of chromium was deposited on the surface followed by 1000 Å of gold and 200 Å chromium by an e-beam evaporator. Finally, the lift off process was performed using a Microposit Remover 1165.

Then, one micron of plasma enhanced chemical vapor deposition (PECVD) oxide is deposited on top at 240°C and the openings in it are etched using dry etching technique. Next, the main distinguishing step with respect to a conventional MEA system was performed: a thin layer of Cr/Au was deposited and patterned on top of the PECVD oxide layer by image reversal processing. The function of this layer is to facilitate the anchoring of the neurons on the pre-defined positions and the guidance of neurites in between the neurons. Prior to culturing neurons on top of these MEAs, proper treatment with SAMs (using a previously described method [1]) has to be performed to make the gold surfaces hydrophobic and unsuitable for neuronal attachment and growth. Finally, an additional 3000 Å of gold was patterned on the contact pads by metal lift-off with image reversal processing for the subsequent wire bonding process.

Figure 1: Fabrication process for the first generation of micro electrode arrays (MEAs).
A single MEA die that has been fixed and wire-bonded on a printed circuit board is shown in Figure 2, and an optical image of the fabricated MEA is shown in Figure 3. The underlying gold electrodes are light gray, whereas the top gold patterns (for neuronal guidance) are pale gray. Dark gray regions are exposed PECVD oxide, which will be treated using hydrophilic SAMs for neuronal anchoring and outgrowth. An SEM image of the 3.1 µm wide opening in PECVD oxide deposited on top of the gold electrode is shown in Figure 4. The circle and interconnect lines are visible as the topographical features, where dark grey is the exposed PECVD oxide and light grey is the thin gold deposited on top of it. Characterization of the MEA device by culturing hippocampal neurons is underway.

Summary

We have developed a novel MEA system with single cell resolution patterning that makes it possible to investigate the activities of individual cells in a patterned neuronal network.

References