The Development of a New Method to Monitor the Activity of Neuronal Networks Using Microelectronic Chips

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Abstract:

It has already been proven that microelectronic chips can monitor neuronal networks in a planar configuration, that is, with only one surface of the cell contacting the electrodes. The focus of this paper is the fabrication and testing of a device that would allow for three-dimensional contact between the neuronal cells and the electrodes, yielding a stronger signal from the cells.

First, as a feasibility test, wells were etched in silicon and glass substrates and cell growth was tested. Once cell growth was deemed sufficient, process details were determined from a rough outline and then the chips were produced.

After several trials, a chip was constructed with gold electrodes and contact plates and a single large well for the cells, which could contain both cells and medium. Wells were constructed using benzocyclobutene, a spin-on dielectric, and were approximately 5 μm deep. The action potentials of the neuronal cells will be monitored with the fabricated chips of microelectrode arrays.

Introduction:

Researchers at the Max Planck Institute for Biochemistry have created microelectronic devices that provide an interface between electrodes and small cultured neuronal networks. These devices have also been used to interface with cultured brain slices. Using transistors on the devices, researchers were able to record the action potentials of multiple cells simultaneously on the small networks, and record changes in potential across the brain slices.

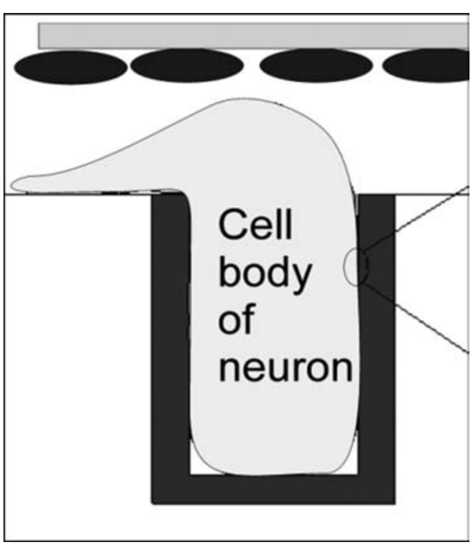

Figure 1: The ideal cell-device interface.

The disadvantage of these chips is that they have a planar interface with the cell surface. This planar interface only puts one surface of the cell membrane in contact with the electrodes.

The goal of our project is to create a microelectronic chip that allows for a three-dimensional interface with the cell. The cell would be contained in a well, as in Figure 1, that would maximize contact between the chip surface and the cell membrane. We expect that this design would increase the electrical signal measured by up to two or three times.

Applications of this research are studies of neuronal network functioning. Eventually, we hope that these devices may be used to study disorders that involve abnormal neuronal network functioning, such as Alzheimer's or epilepsy.

Experimental Procedure:

The initial stage of the project was to test the adhesion of neuronal cells on silicon and glass substrates. Square wells of a depth of 1 μm were etched into the substrates and AtT-20 neuroendocrine cells were cultured on the surface. These cells were examined under an optical microscope and counted.

The next step was to fabricate microelectronic devices capable of registering the action potentials from the cells. Using a rough process outline and a set of masks already created (Figure 2), fabrication techniques were refined. The first step of the process was electrode formation with a liftoff technique, using 500 Å of Ti and 3000 Å of Au. After the electrodes were deposited, a plasma-enhanced chemical vapor deposition process was used to deposit 1 μm of SiO_2, in order to protect the wires connecting the electrodes to the contact pads. The SiO_2 was then patterned and dry etched with a reactive ion etching process to reveal the electrodes while retaining the insulation on the wires.

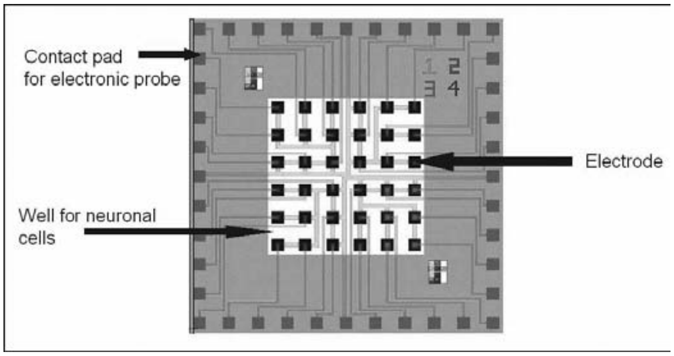

Figure 2: A diagram of the microelectronic device.

After the SiO_2 etch, wells were formed using benzocyclobutene (BCB or Cyclotene), a spin-on, photosensitive polymer. The initial design called for a square well over each electrode, resulting in a six-by-six array of wells 300 μm on a side. However, it was determined that these wells would be suboptimal for testing, as it would be difficult to keep the surface of the chip dry while retaining enough medium to keep the cells alive. Therefore, the masks were redesigned to create a square well 4 millimeters on a side, which allowed neurons and medium to be in contact with all of the electrodes. The BCB was used to create wells 5 μm deep. Cells were then cultured on the surface of the chip and examined under a microscope.

Results and Conclusions:

As this project was just started this summer, data was not obtainable in such a short time frame. However, the process was refined significantly, and specific parameters were obtained for all aspects of the process. It was demonstrated that the chips could be fabricated and that neuronal cells could be grown on them (Figure 3). Testing of the devices should begin shortly.

Future Work:

Fabrication and testing of the chips must continue in order to gain reproducible results from the current design. In addition, chips must be designed that allow for better contact between the electrode and the cell, in order to get closer to our ideal concept of a three-dimensional electrical contact. Once the optimum design has been achieved, comparisons must be made with planar-contact type chips to confirm that the three-dimensional-contact chips strengthen the signal.

Acknowledgments:

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References:

[1] Fromherz, Peter. "Semiconductor chips with ion channels, nerve cells and brain"; Physica E, p. 24-34, 2003.

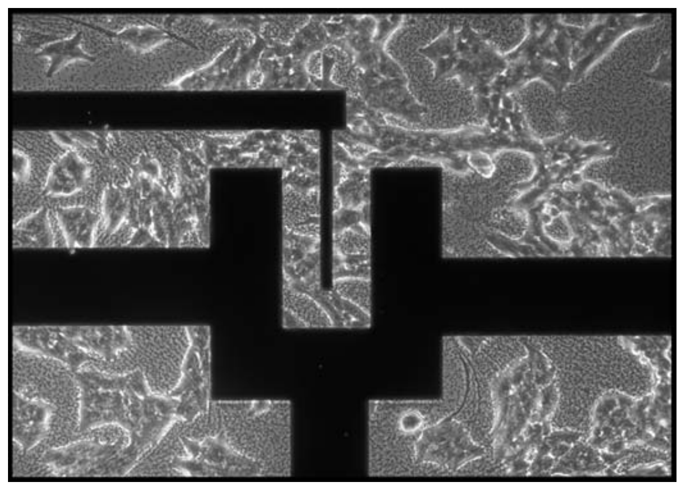

Figure 3: A picture of an electrode from the device, with neuronal cells cultured on the surface (40x).