## INTRACELLULAR RECORDING WITH HIGH ASPECT RATIO MEMS NEURONAL PROBES

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We fabricated and tested micro-machined silicon needles for intra- and extracellular neuronal recording. The fabricated needles have extremely sharp tips (<1  $\mu$ m), they are long (>300  $\mu$ m) and exhibit high mechanical strength. These characteristics are necessary for effective bending and penetration of cell membranes for intracellular recording (Figure 1). To prepare needles for recording, they were metallized and their shank passivated. Wiring and packaging schemes were developed and surgical implanting techniques for *in vivo* intracellular recording were refined. Using these needles, we were able to obtain extremely localized extracellular signals and to perform first intracellular recordings with silicon based micro-probes.

Extensive work in recent years has demonstrated the potential of Micro-Electro-Mechanical Systems (MEMS) devices for neurological applications<sup>1</sup>. MEMS neuronal devices are particularly promising due to their small dimensions and the ease with which multi-site devices can be produced. Previous efforts in MEMS probe fabrication focused on extracellular surface electrodes which are typically rather simple to prepare and implant. However, such extracellular recording methods do not provide critical information about the DC state of a cell nor can they be used to easily identify the behavior of single cells.

To produce needles suited for intracellular recording, we used highly conducting (n-type), 800  $\mu$ m thick silicon wafers. Using a dicing saw, we created arrays of tall pillars<sup>2</sup>. To sharpen the tips we used reactive ion etching (RIE) with SF<sub>6</sub>. This is a robust, self-sharpening process which we optimized in order to obtain high aspect ratio (Figure 2). To produce separated needles, the wafer was bonded with photoresist to another substrate and cuts to separate the parts were made prior to the sharpening process. After the sharpening, the wafer was sputtered with gold and with silicon nitride which was later slightly etched to expose the tip. Finally, we soaked the wafer in acetone and released the needles.

Our process yields a probe geometry that is similar to the pulled glass electrodes commonly used in intracellular recording schemes (Figure 3). To demonstrate the electrode performance, we show results obtained with two biological models: *Manduca sexta* (hawk moth) and *Tritonia diomedea* (a sea slug). We begin with a relatively simple extracellular preparation with a moth. In Figure 4a we show a plot of evoked extracellular potentials in the lobula plate (brain optic lobe) of a moth. The data show excellent single unit (one neuron) evoked potentials. Such results are likely a consequence of the narrow tip diameter of the MEMS probe. Using the brain of *Tritonia* (brain cells >500  $\mu$ m) we tested the needles for intracellular applications. We penetrated into the membrane of cells and performed intracellular recording (Figure 4b). The significant noise level is likely due to insufficient passivation, and a damaged membrane. Future work will focus on improved passivation and insertion techniques.

In summary, we present a technique to produce extremely fine, yet long and strong, silicon needles. Facilitating refined geometry we were able to obtain high quality *in vivo* extracellular recording. Moreover, we demonstrated a first evidence for cell penetration and intracellular recording with silicon needles.

<sup>&</sup>lt;sup>1</sup> G. T. A. Kovacs, "Introduction to the Theory, Design, and Modeling of Thin-Film Microelectrodes for Neural Interfaces", *in Enabling Technologies for Cultured Neural Networks*, D. A. Stenger, and T. M. McKenna, Eds., Academic Press, San Diego, 121-165 (1994).

<sup>&</sup>lt;sup>2</sup> P. K. Campbell, K. E. Jones, R. J. Huber, K. W. Horch, and R. A Norman, "A Silicon Based, Three-Dimensional Neural Interface: Manufacturing Processes for an Intracortical Electrode Array", IEEE Transactions on Biomedical Engineering 38, 758-768 (1991).

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Figure 2. An ESEM image of an array of sharpened silicon needles.



Figure 3. (a) A tip of a sharpened silicon needle. The needle was tilted approximately  $45^{\circ}$  with respect to the plane of the image. (b) A pulled glass capillary commonly used for neuronal intracellular recording.



Figure 4. (a) Evoked extracellular potentials in the lobula plate of *Manduca sexta* (hawk moth) plotted versus time. (b) Evoked intracellular potentials in the brain of *Tritonia diomedea* (a sea slug) plotted versus time (smoothen data). The positioning of the probe was controlled via micro-manipulators and an optical microscope.