Self-closing Micro-Cuff for Neural Recording in the Locust

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Abstract - We have developed a self-wrapping flexible cuff electrode for recording electrical activity on the descending contralateral movement detector (DCMD) neuron of the locust. First, a gold film is sputtered onto a monoaxially oriented polycarbonate membrane. Then, the resulting bilayer film is cut into individual strips parallel to the machine direction of the polycarbonate film. Thermally shrinking the polycarbonate at 155 0 C actuates the curl. This method can be used to fabricate controlled diameters between 50-250 µm; diameter is controlled by thickness of the two layers and the temperature and duration of the heat shrinking process. The cuffs are self-closing, and do not require suturing for attachment to a nerve. The cuffs are carefully unrolled, a gold film deposited, and a plasma-deposited, ion-sensitive resist applied. A stencil mask is then used to define the metallization lines by 50 kV He⁺ ion beam proximity lithography. Ion milling transfers the resist pattern to the metal film. Conformality of the resist and large depth-of-field of the lithography are key to high resolution patterning on these non-flat substrates. The dielectric overcoat is a layer of exposed resist, which simplifies the opening of the contact windows, and provides excellent biocompatibility.

Keywords: Cuff electrode, self-closing, lobula giant movement detector (LGMD) neuron, descending contralateral movement detector (DCMD) neuron, plasma enhanced chemical vapor deposition, ion beam lithography

1. Introduction

Swarming insects have unique capabilities in collision avoidance and escape behaviors [1]; a classical animal model is the grasshopper. The lobula giant movement detector (LGMD) neuron in the visual system of the grasshopper is highly sensitive to looming objects and plays an important role in the generation of escape behaviors [1]-[2]-[3]. The LGMD synapses onto the DCMD, a neuron that relays spikes in a 1:1 manner to thoracic motor centers through the ventral nerve cord. Recordings of the extracellular action potentials of the DCMD are used to monitor the LGMD response to visual stimuli [1]-[2]-[3]. Currently, extracellular recordings on the DCMD are collected by positioning metal hook electrodes around the ventral nerve cord [4]-[5]. The focus of this research is to extend this approach to self-wrapping, multi-electrode cuffs with diameters in the 70-100 μ m range. Generally, extra-neural cuffs for higher vertebrate animals and humans are larger, require suturing for nerve attachment, have limited ability to adjust to nerve diameter, and cannot be safely detached from a nerve.

2. Experimental Methods

A cuff is formed by thermally releasing the strain energy stored in a monoaxially-oriented polycarbonate (PC) layer in a PC/gold bilayer film. The gold layer is relatively stiff and bends during actuation. Figure 1 shows fabrication steps prior to heat shrinking. Cuffs are folded and transferred to the oven while still attached to the metal ring. The separation ensures they do not stick to each other.

We found that heating the cuffs for a fixed period of time does not provide sufficient control of the cuff diameter. Instead, heat treatment is carried out in multiple 15-second exposures between which the cuffs are removed from the oven and inspected under a dissection microscope. Periodically, it is necessary to allow the temperature to stabilize before beginning the next cycle.



Figure 1- Steps of the cuff fabrication process: a) An epoxy-coated washer is attached to a monoaxially-oriented polycarbonate film floating on the surface of a beaker of water and the machine direction marked with north and south annotations [6]. b) Two strips of Kapton tape, 500 µm apart, are attached to the membrane in the transverse direction. c) The ring-mounted membrane is coated with a 250 nm thick gold film. d) The section above the untaped region is removed with surgical scissors under a dissection microscope. e) The bottom section is cut into 3-mm wide strips in the machine direction. After heat shrinking, the untaped *head* becomes the cuff, while the taped *tail* carries the electrode lines that connect to the wiring harness.

3. Experimental Results

Figure 2 shows cuff diameter as a function of cumulative heating time (CHT); each data point corresponds to a single heating cycle. Although the initial cuff diameters may vary significantly, for CHT greater than 40 seconds the distribution narrows considerably. For this example, reliable cuff diameters are obtained between 70 and 250 µm.



Figure 2- Cuff diameter as a function of cumulative heating time.

After the cuff is fabricated, and gently unrolled, the electrodes lines are printed using large depth-of-field ion beam lithography in a negative-tone resist [7]-[8]-[9]. A 110 nm thick gold (Au) film is then deposited by DC-magnetron sputtering. Then an 80 nm thick resist is deposited by plasma enhanced chemical vapor deposition. Metal lines are defined by exposing the resist through a slit mask oriented parallel to the machine direction using a 50 kV He⁺ ion beam. The conductor lines are developed for 40 seconds in amyl acetate solution at room temperature. After the development, the resist pattern is transferred to the gold layer by 500 V Ar⁺ ion milling. Once the electrode lines are defined, a dielectric overcoat layer of exposed resist is applied. The final resist layer is exposed with a 200 μ m wide x 1 mm long wire mask. After exposure, the wire is removed and the membrane developed in amyl acetate to form a contact to the conductor line beneath. Scanning electron micrographs of cuffs at various stages of fabrication are shown in figure 3.



Figure 3- Scanning electron microscopy images of cuff electrodes. a) Top view of an unrolled cuff with a 100 µm wide gold conductor line oriented in the machine direction. The head and tail correspond to figure 1. b) A free-standing, 1 mm wide cuff and interconnect line after trimming excess material. c) Cross-sectional view of a multi-curl cuff, 75 µm in diameter. A single curl cuff can be fabricated by shortening the head region by trimming.

The dielectric overcoat, which is not yet applied to the cuffs described above, has been tested *in-vivo* using an early version of the cuff electrode [10]. Figure 4 is a high-quality recording of the DCMD of a locust. This shows that the pinhole density of the overcoat is very low. In addition, impedance measurements of neural probes that used the same overcoat show that it does not crack due to bending, disinfection, immersion in phosphate buffer solution (PBS) for over a month, or insertion in a cannula [11].



Figure 4- Electrophysiological recording from single-electrode neural Cuff [10].

4. Conclusion

We have demonstrated a cuff, formed by thermally releasing the strain energy stored in a monoaxially-oriented polycarbonate (PC) layer in a PC/gold bilayer film, with controlled diameters of 70-250 μ m, and demonstrated the fabrication of 100 μ m wide interconnect lines. The next steps are to apply a proven dielectric overcoat with vias between the interconnect lines and the hemolymph of the grasshopper. This, and a facile implantation technique, will be discussed at the conference. In addition, measurements of the detachment force and the resilience of the cuff will also be discussed.

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