Published in final edited form as:

Biomed Microdevices. 2012 February ; 14(1): 193–205. doi:10.1007/s10544-011-9597-4.

Microelectrode arrays fabricated using a novel hybrid microfabrication method

Mark W. Merlo,

The Henry Samueli School of Engineering, Department of Biomedical Engineering, University of California, Irvine, CA 92697-2700, USA

Russell L. Snyder,

Epstein Laboratory, Department of Otolaryngology, Head and Neck Surgery, University of California, San Francisco, CA 94143-0526, USA

John C. Middlebrooks, and

School of Medicine, Department of Otolaryngology, University of California, Irvine, CA 92697-5310, USA

Mark Bachman

The Henry Samueli School of Engineering, Department of Electrical Engineering & Computer Science, University of California, Irvine, CA 92697-2700, USA (949) 824-6421
mbachman@uci.edu

Abstract

We present novel hybrid microfabrication methods for microelectrode arrays that combine microwire assembly, microelectromechanical systems (MEMS) manufacturing techniques and precision tool-based micromachining. This combination enables hybrid microfabrication to produce complex geometries and structures, increase material selection, and improve integration. A 32-channel shank microelectrode array was fabricated to highlight the hybrid microfabrication techniques. The electrode shank was 130 μm at its narrowest, had a 127 μm thickness and had iridium oxide electrode sites that were 25 μm in diameter with 150 μm spacing. Techniques used to fabricate this electrode include microassembly of insulated gold wires into a micromold, micromolding the microelectrode shank, post molding machining, sacrificial release of the microelectrode and electrodeposition of iridium oxide onto the microelectrode sites. Electrode site position accuracy was shown to have a standard deviation of less than 4 μm . Acute *in vivo* recordings with the 32-channel shank microelectrode array demonstrated comparable performance to that obtained with commercial microelectrode arrays. This new approach to microelectrode array fabrication will enable new microelectrodes, such as multi-sided arrays, drug eluting electrodes and biodegradable shanks.

Keywords

Hybrid microfabrication; Microelectrode arrays; Mechanical micromachining; Neural recording; Microassembly; MEMS

1 Introduction

Microelectrode arrays (MEA) play a critical role in advancing our understanding of the nervous system by providing a means to record and stimulate neuronal activity (Churchland et al. 2007; Meister et al. 1994). In addition to basic research, electrodes have potential clinical applications including treating diseases (Lozano et al. 1996; Stacey and Litt 2008) and prosthetics (Donoghue et al. 2007; Middlebrooks and Snyder 2010). This large range of

applications has resulted in electrodes with a wide variety of structures and topologies (Cheung 2007; Ward et al. 2009).

Two common electrode types are microwire MEAs (Nicoletis et al. 1997; Nicoletis et al. 2003; Schmidt et al. 1976; Williams et al. 1999) and lithographically defined MEAs (Kipke et al. 2003; Mercanzini et al. 2008; Wise et al. 1970). Microwire MEAs typically consist of multiple insulated wires arranged in parallel in either two-dimensional or three-dimensional configurations. The tips of these insulated wires are exposed, enabling the electrode to stimulate or record neurons within the vicinity of the tips. The diameters of these wires generally range from $\sim 30\ \mu\text{m}$ to $100\ \mu\text{m}$ (Cheung 2007). Microwire MEAs are typically made of multiple components that are assembled together using assembly fixtures and alignment jigs (Nicoletis 2008). These assembly methods offer the ability to produce complex electrode configurations such as, for example, the ability to move each microwire independently (Jackson and Fetis 2007). These assembly methods also allow the microwire MEAs to be constructed from a large range of materials and to have seamless integration with its packaging.

Lithographically defined MEAs are planar devices typically with one or more thin shanks and multiple electrode sites along each shank. This is in contrast with microwire MEAs, which usually have one electrode site per shank. Each site area of a lithographically defined MEA commonly ranges from $\sim 100\ \mu\text{m}^2$ to $1000\ \mu\text{m}^2$ (Wise et al. 2008). Lithographically defined electrodes normally utilize integrated circuit fabrication as well as microelectromechanical systems (MEMS) based fabrication techniques (Kewley 1997; Seymour et al. 2011; Wise et al. 2004). These planar fabrication processes are popular for MEA fabrication because they enable batch fabricating with micrometer-sized features.

Despite their popularity, planar processes have shortcomings. Due to their inherent two-dimensional nature, it is difficult, and many times impossible, to fabricate complex, three-dimensional microelectrodes. Groups have attempted to circumvent these limits to create microelectrodes with complex structures. Some exploit natural phenomena, such as anisotropic etching, but these structures are limited to specific shapes (e.g., Luttge et al. 2007). Others have created three-dimensional microelectrodes by using self-folding methods, but this a complex process that is also limited to very specific structures (Wang et al. 2010).

These planar processes are also limited in the selection of materials that can be used. This is generally because the manufacturing methods rely on specific chemical reactions and therefore substituting materials is not a straightforward process. This also limits the types of devices that can be integrated since many times the components are not compatible with the fabrication processes. Additionally, even though planar MEAs are batch fabricated, almost all require additional individual packaging before they become a functional device, such as the addition of a connector (e.g., Cheung et al. 2007).

Recent efforts have scaled down non-semiconductor manufacturing methods to micron-scale precision. These precision tool-based micromachining methods are now capable of fabricating complex structures on the micrometer scale with a large range of materials (Chae et al. 2006; Dornfeld et al. 2006; Fang et al. 2006; Masuzawa and Tönshoff 1997). This is in stark contrast to planar fabrication methods and makes precision tool-based micromachining an attractive alternative for fabricating complex and diverse MEAs.

Another factor not often discussed is that medical grade MEAs must be fabricated in a good manufacturing practice (GMP) certified facility. Planar MEAs require a cleanroom, of which very few are GMP certified. MEAs fabricated without a cleanroom have a distinct advantage

when transitioning from research to clinical applications since they can be fabricated in many of the existing GMP-certified medical device facilities.

The goal of this work was to develop MEA hybrid microfabrication methods by combining microwire assembly, planar manufacturing techniques and precision tool-based micromachining. Hybrid microfabrication has three potential advantages over other traditional electrode fabrication methods: (1) complex geometries and structures, (2) increased material selection, and (3) improved integration.

Single shank MEAs were fabricated and tested to demonstrate these novel microelectrode fabrication methods. The single shank MEA was selected to demonstrate hybrid microfabrication for several reasons. First, shank MEAs are well established in the literature (Wise et al. 2008) and well understood by the electrode research community. Second, such electrodes are commonly fabricated lithographically. Using new fabrication methods to develop the same electrode topology provides a standard baseline for comparison. Third, shank electrode topology is straightforward and easily understood.

Previous researchers have attempted to fabricate and assemble shank MEAs without lithography (Jellema and Weijnen 1991), but all the fabrication steps involved manual construction techniques and craftsmanship. These techniques are difficult to reproduce and would not easily scale to larger production volumes. Non-lithographic electrodes with multiple electrode sites per shank are available commercially, but these MEAs use topology-specific fabrication methods and are much larger for a given number of electrode sites. In contrast, the work presented here demonstrates versatile hybrid microfabrication methods that utilize precision machines and manufacturing methods found in today's modern manufacturing industry as well as techniques borrowed from MEMS technology. Hybrid microfabrication can produce MEAs with complex geometries, a large range of materials, seamless integration with supporting components, and size and functionality similar to modern MEAs.

In this paper, we present a non-lithographic, 32-channel shank electrode and its hybrid microfabrication methods. The hybrid microfabrication methods will be discussed in detail along with some of the challenges they present. Other electrode topologies constructed using hybrid microfabrication are shown to demonstrate the potential of these fabrication methods. Finally, acute *in vivo* recordings were performed to demonstrate its performance compared to commercial shank electrodes made using planar lithography.

2 Methods

Figure 1(a-c) shows images of the 32-channel shank MEA used in this paper to illustrate the MEA hybrid microfabrication processes.

As shown in Figure 2, the shank MEA was composed of three major elements: (1) an electrode shank, (2) a wire bundle and (3) an interface printed circuit board (PCB). The electrode shank had 32 iridium oxide electrode sites with a 150 μm pitch. Each electrode site was nominally 25.4 μm (0.001 in) in diameter. The most distal electrode site was approximately 150 μm from the tip of the electrode and the entire electrode shank length was approximately 5 mm. The maximum width of the electrode shank was 250 μm at the most proximal site and tapered down to 130 μm in width near the tip of the electrode. The thickness of the electrode shank was 127 μm across its entire length.

The wire bundle was composed of 32 insulated gold wires with a total wire bundle diameter of 200 μm across its entire 50 mm length. The wire bundle terminated at the interface PCB with independent electrical connections to each of the 32 gold wires. The connectors and

PCB configuration permitted mounting to an animal's skull and mating with a ZIF-Clip ZC32 headstage (Tucker Davis Technologies, Alachua, FL).

2.1 Hybrid microfabrication

All MEAs presented herein were fabricated at the Bio-Organic Nanofabrication facility (BiON) in the California Institute for Telecommunications & Information Technology building (Calit2) at University of California, Irvine. The processes required for these hybrid microfabrication methods include precision computer numerical control (CNC) machining, microassembly, electrodeposition, micromolding, and sacrificial release.

Figure 3(a). A micromold was machined from a 381 μm (0.015 in) thick acrylic sheet (SABIC Polymershapes, San Diego, CA). An outline of the micromold and two fixture holes were etched to a partial depth using a CO₂ laser cutter (VersaLaser VL-200, Scottsdale, AZ). The micromold was separated from the acrylic sheet by bending the sheet and applying pressure to the micromold. This was performed in a manner that caused a crack to form through the thickness of the sheet along the entire length of the laser etch. This method prevented burrs that typically form from a laser through cut. The micromold was then fixed to a fixture and attached to the bed of a Haas vertical CNC Mini Mill (Haas Automation, Oxnard, CA).

The shank slot in the micromold was machined using a 127 μm diameter, two-flute, square end mill (McMaster-Carr, P/N 8832A005, Robbinsville, NJ) at 6000 rpm with a vertical and contour feed rate of 25.4 mm/min (1 in/min). The channel was cut in multiple passes in depth increments of 25.4 μm to a total depth of 127 μm . The end mill traveled along a path profile that cut a slot with a width of 130 μm near the tip and 250 μm near the base.

The 32 through holes in the slot were 51 μm in diameter and were peck drilled using a 51 μm (0.002 in) diameter drill bit (McMaster-Carr, P/N 8904A11) at 6000 rpm with a feed rate of 15.24 mm/min (0.6 in/min) and a pecking depth of 25.4 μm (0.001 in). After completing the peck drilling operation, the drill bit was fed once into and out of each hole to help remove any debris that developed in the holes during the drilling cycle.

The machining parameters for the 51 μm drill and 127 μm end mill were determined empirically with focus on achieving smooth walls and minimal burrs. Critical parameters that were varied included feed rate, spindle speed, cutting depth increments, peck depth and number of passes.

It is common practice to operate tools with small diameters at very fast RPMs. This keeps the surface speed of the cutter high enough to allow faster feed rates and therefore less time per operation (Chae et al. 2006). The Haas CNC mill used for this development has a maximum spindle speed of 6000 RPM. Since this RPM was low for small diameter cutters, the maximum RPM was selected for all operations to keep the cutter surface speed to a maximum.

The feed rate was another important factor in micromold quality. Cutting too fast caused the cutting tool to take too much material per rotation. This imposed large cutting forces on the cutter and resulted in tool breakage. We also found that faster cutting speeds resulted in a rougher surface finish. Cutting too slow produced excess heat from the tool scraping the material instead of cutting it. Since these slow cutting speeds prevented the tool from cutting into the material, the tool pushed into the material until enough built up that it could actually be cut (Dhanorker and Ozel 2008). This generally resulted in burrs and plastic deformation of the material near the cutting path.

Alignment holes (1.61 mm diameter) were drilled into the micromold to allow easy alignment with the fixtures used throughout the fabrication process. A 0.8 mm molding reservoir was machined to a depth of 127 μm to aid in the molding process.

To insure proper alignment and reduce position errors, all of the aforementioned CNC machining steps were completed during a single machining setup.

Figure 3(b-c). Assembly of each 50 AWG insulated gold wire (California Fine Wire, Grover Beach, CA) required a two-step process. First, a gold wire was assembled into the micromold (Figure 3(b)) and then an electrical connection was made between the wire and a silver bond pad on the interface PCB (Sunstone Circuits, Mulino, OR) (Figure 3(c)). The insulated gold wire had a nominal total diameter of 30.4 μm (25.4 μm diameter gold wire with 2.5 μm thick polyurethane insulation).

The assembly process started with one end of a 15 cm length gold wire being adhered to double-sided tape and threaded through a via and the wire bundle hole of the interface PCB (not shown). The gold wire was then manually threaded through the proximal most hole in the micromold (Figure 3(b)), pulled in tension and placed on double-side tape to keep the wire in tension. A small drop of epoxy was placed on the PCB and wire (not shown) to relieve any stress that may occur during the assembly process.

Electrical contact between the insulated gold wire and a silver bond pad on the interface PCB was made with a wedge bond (Figure 3(c)). The wedge bond was formed by pressing the insulated gold wire against the bond pad with a wedge bonder at 400 $^{\circ}\text{C}$. Since the temperature of the wedge bonder was greater than the melting temperature of the polyurethane insulation, the insulation was removed locally to allow formation of the wedge bond. After the wedge bond was completed, a small drop of epoxy was placed on the wedge bond to give it added mechanical strength (not shown).

This process was completed under a microscope and was repeated for all 32 electrode site holes with the most proximal free hole being used next in the assembly process. The most distal hole was the final hole to have a wire threaded through it.

Figure 3(d). A micromolding process was used to fix the wires into position and form the shank of the electrode. The molding reservoir of the micromold was filled with a low viscosity epoxy (Epoxy Technology, EPO-TEK 301, Billerica, MA), but kept from entering into the micromold. The micromold was placed under vacuum before the epoxy entered the micromold. Capillary action pulls the epoxy into the micromold, between the individual wires and into the electrode site holes. The micromold was removed from the vacuum and the epoxy was cured in a 40 $^{\circ}\text{C}$ oven for four hours.

Not shown is the process of forming the wire bundle. To form the wire bundle, the micromold was held fixed while the wires were held in tension and the interface PCB was twisted a total of five revolutions. The twisted wire bundle was fixed by running a small drop of EPO-TEK 301 epoxy along its length. The device was placed in a 40 $^{\circ}\text{C}$ oven for four hours to set the epoxy in the wire bundle.

Figure 3(e). A post-molding CNC milling operation was performed to eliminate the residual wire tips ("nubs") remaining in the micromold. The micromold was placed in a fixture upside down and aligned using the alignment holes and pins. A 127 μm square end mill was centered over each wire and plunged into the micromold at 6000 rpm and a feed rate of 50.8 mm/min (2 in/min). The end mill went to a depth that was flush with the bottom of the electrode and dwelled at this depth for 500 ms.

Figure 3(f-g). A sharp tip was added to the electrode shank using a second post-molding CNC milling operation. The micromold was placed right side up and aligned using the alignment holes and pins. A 0.8 mm diameter square end mill cut the contour of the sharp tip in multiple passes at depth increments of 25.4 μm to a total depth greater than 127 μm (i.e., beyond the micromold slot depth). Figure 3(h). The electrode was released from the micromold by dissolving the acrylic micromold in acetone for 30 minutes. After release, the electrode was rinsed in deionized water.

Figure 3(i). The surface conditioning of the electrode sites was completed in a two-step process. The first step increased the surface area of the gold sites and the second step coated the electrode sites with iridium oxide.

The gold site surface area was increased by applying a 1 kHz, 2.8 V peak-to-peak sine wave between a large area platinum wire and each electrode site for approximately two seconds while submerged in a 0.9% NaCl solution at room temperature. The gold dissolved during anodic currents and redeposited on the gold surface during cathodic currents (Hung et al. 2010). Our group independently developed this gold conditioning process, but it is similar in concept and implementation to Huang et al. (2009).

Iridium oxide was electrodeposited onto each gold site using a protocol similar to that described by previous groups (Meyer et al. 2001; Lu et al. 2009). The electrode was submerged into an aqueous solution of 4 mM IrCl_4 , 40 mM oxalic acid, and 340 mM K_2CO_3 having a final pH of 10.5. Using a potentiostat (Digi-Ivy DY2013, Austin, TX), a three-electrode cell was formed between a Ag|AgCl (1 M KCl) reference electrode, a large area platinum counter electrode, and the electrode to be coated as the working electrode.

The electrodeposition protocol consisted of a repeated triangular waveform followed by a repeated rectangular waveform. The triangular waveform ramped the potential between -0.025 V and 0.525 V versus Ag|AgCl at a rate of 50 mV/s for 50 cycles. This was followed by a rectangular waveform that held the potential for 0.5 seconds at both -0.025 V and 0.525 V for a total of 1600 complete cycles. This electrodeposition protocol was coordinated using the potentiostat's control software.

2.2 Electrode site positional accuracy

The position of the electrode sites and micromold holes were measured from scanning electron microscope (SEM) images using ImageJ (NIH, Research Services Branch). An ellipse was manually drawn in ImageJ to outline the perimeter of each micromold hole and electrode site. The distance-to-pixel ratio in ImageJ was calibrated with the scale bar overlay of the SEM images. The coordinates of the ellipse centers were measured using ImageJ's measurement tools. These coordinates were used to calculate the center location and pitch between each feature.

2.3 Wedge bond impedance

An insulated gold wire was wedge bonded onto two silver bond pads and the impedance was measured between the pads. The insulated gold wire used for these measurements was the same wire used in the MEA fabrication (i.e., 25.4 μm conductor diameter insulated with a 2.54 μm thick polyurethane). A wedge bonder at 400 $^{\circ}\text{C}$ was used to make the wedge bonds. The impedance measurements were taken between 18 wedge bond pairs at 1 kHz at 250 mV.

2.4 Electrode site impedance

The impedance of each electrode site was measured with three different surface conditions: (1) as-cut gold, (2) conditioned gold and (3) iridium oxide electrodeposited on gold. The impedance of each electrode site was measured using a PARSTAT 2273 potentiostat (Princeton Applied Research, Oak Ridge, TN) in a standard three-electrode cell with a large-area platinum counter electrode and a saturated calomel reference electrode. All measurements were taken at room temperature in a faraday cage. The electrolyte was phosphate buffer saline (PBS) at approximately 7.3 pH and was composed of 130 mM NaCl, 22 mM NaH₂PO₄, and 81 mM Na₂HPO₄. The impedance was measured with a 10 mV root-mean-square (rms) sinusoidal excitation at 1 kHz (Cogan 2008). The diameter of each electrode site was 25.4 μm , giving each site a geometrical surface area (GSA) of 506 μm^2 (Trasatti and Petrii 1992; Cogan 2008).

The impedance measurements of the as-cut gold sites were taken after releasing the electrode from the micromold but before applying any surface conditioning. Next, the electrode sites were conditioned using the same process outlined in the hybrid microfabrication section. At this point, the electrode site impedances were measured again. Finally, iridium oxide was electrodeposited onto the gold electrode sites using the same process outlined in the hybrid microfabrication section. After the iridium oxide electroplating, the impedance of each electrode site was measured again.

2.5 Cyclic voltammetry

Cyclic voltammograms were taken in deoxygenated PBS at room temperature using a potentiostat (Digi-Ivy DY2013, Austin, TX). The PBS was identical to the solution used for the impedance measurements (Lu et al. 2009). The reference electrode was a Ag|AgCl (1 M KCl) electrode and the counter electrode was a large-area platinum electrode. All electrode sites were connected together as the working electrode of the potentiostat. The cyclic voltammograms were taken between -0.60V and 0.80V versus Ag|AgCl at a scan rate of 50 mV/s (Meyer et al. 2001). The cyclic voltammograms of a 32-channel MEA with four inactive sites (i.e., 28 active sites) were taken before and after iridium oxide electroplating.

2.6 Acute surgery and neural recordings

In vivo tests were conducted in two purpose-bred cats according to a protocol approved by the Institutional Animal Care and Use Committee at the University of California at Irvine. Methods were essentially the same as those described by Middlebrooks and Snyder (2007). Briefly, animals were anesthetized with barbiturate. The skull was opened, and a portion of the occipital cortex was ablated to permit a visually guided approach to the right inferior colliculus. An MEA was advanced through the inferior colliculus using a micropositioner. The orientation was in the coronal plane and $\sim 35^\circ$ from the midsagittal plane. The reference electrode was an uninsulated silver wire inserted in the right temporal muscle. Stimuli were generated and neural waveforms were acquired using System 3 hardware from Tucker-Davis Technology (TDT; Alachua, FL). Stimuli were 40-ms tone bursts presented from a calibrated loudspeaker located 10 cm from the animal's left ear. Neural waveforms were recorded simultaneously through the 32 channels of the MEA using a ZC32 headstage, 2 RA16PA preamps and analog-to-digital convertors, and an RX5 digital signal processor interfaced to a personal computer running custom MATLAB scripts. Waveforms were saved to disk and sorted offline on the basis of waveform shape.

3 Results and discussion

These hybrid microfabrication methods presented are capable of producing a shank MEA similar to those currently presented in the literature (Figure 4(a)) (Cheung 2007; Wise et al.

2008). Due to the versatility and large range of options this hybrid methodology presents, modifications to the presented techniques can produce other MEA topologies. This includes a MEA with electrode sites on three sides of its shank (Figure 4(b-c)) as well as a MEA with an embedded optical fiber (Figure 4(d)).

Our hope is that these hybrid techniques are not treated as a strict step-by-step guide, but instead demonstrate a methodology that can be extended and built upon. Hybrid microfabrication provides a large amount of design freedom and is not limited in many of the ways that limit other microfabrication methods. This increase in design freedom includes material selection, three-dimensional construction, and component integration. A large range of materials can be utilized since the manufacturing techniques are less dependent on material properties and chemistries.

Three-dimensional structures can be constructed since common manufacturing methods are utilized at the microscale. This enables not only complex structures but also enables efficient use of space. For example, typical planar electrodes increase in width linearly with increasing number of channels. This is usually countered by increasing the number of layers, which adds complexity and cost and decreases yield due to the increased number of mask layers. In contrast, the three-dimensional nature of hybrid microfabrication allows full use of the volume of the electrode. This enables the channel wires to be closely bundled within the electrode shank and decreases the dependence between the shank diameter and number of channels.

Hybrid microfabrication also enables dissimilar components to be integrated into a single device since components can be fabricated separately before integration.

3.1 Hybrid microfabrication

The spindle RPM and machine feed rate were critical parameters in achieving clean cut molds with no burrs. Figure 5(a-b) shows a typical micromold machined using the parameters outlined in this paper.

Wire assembly was performed manually under a microscope. Despite the small hole and wire diameter, it was fairly simple to thread the gold wire into the micromold hole. For a 32-channel MEA, this process took approximately four hours to complete. Methods to automate this manufacturing step were not explored in this study, but we envision the use of automated wire handling machines, similar to the wire ball bonders used to assemble integrated circuits, to aid in the wire assembly.

Heated wedge bonding was the preferred method for making electrical connections to the insulated gold wire. Soldering the gold wire proved difficult since, when heated, the gold reacted with the tin in the solder. The intermetallics that were formed between the gold and tin had a lower melting temperature than the soldering temperature (Manko 2001). This led to the gold wire dissolving once the solder wetted the wire.

Reliability of the gold wedge bond was a potential issue. We found that if an electrode site was not functioning, it was generally the result of a poor wedge bond. It was difficult to test the electrical integrity of the wedge bond before the electrode construction was completed since an electrical connection was only made to one end of the gold wire. We observed that the mechanical strength of the wedge bond appeared to be correlated to its electrical integrity. The wedge bond's mechanical strength was tested by brushing a gold wire of the same diameter and approximately 5 mm in length against the wedge bond. The force applied to the wedge bond by this method was calculated to be on the order of tens of micronewtons. If any movement was perceived at the wedge bond, the wire was rebonded until it was

mechanically fixed. This mechanical testing method improved electrical reliability since it gave an indirect method of checking the wedge bond connections prior to completing assembly.

A low viscosity, biocompatible (Loeb et al. 1977) epoxy was selected and molding was performed under vacuum to help prevent the formation of air bubbles. Air bubbles were typically found near the base of the electrode where the wires were compacted together and the epoxy did not flow easily. Other methods, such as injecting epoxy under pressure, could help prevent air bubble formation but were not explored for the purpose of this paper.

In order to fabricate an electrode with flush electrode sites, the wire nubs that were formed in the micromold had to be planarized. We explored two methods to make these electrode sites flush. The first method was the post molding machining operation presented in this paper. This method had the advantage of removing the nubs locally and thus allowed nearby features to remain undisturbed. It also allowed the nubs to be cut to different heights, which can be difficult using other fabrication methods. The second method utilized a precision razor blade to cut the nubs flush to the surface of the electrode. This method gave a surface finish similar to the post molding machining operation (Figure 8(a)), but could not accommodate features with multiple heights. Depending on the electrode configuration, one planarization method might be preferred over the other.

A simple two-dimensional, post molding, sharpening operation was used to form the chisel tip. This tip configuration was the simplest to fabricate and easy to visualize. Due to the three-dimensional capabilities of the machining process, it was also possible to sharpen the tip three-dimensionally. This enabled complex tip structures with multiple curvatures and taper angles.

3.2 Electrode site positional accuracy

Figure 6(a) shows a histogram for the pitch of the machined micromold holes. The pitch measurements for these micromold holes were tightly centered around the as-designed (ideal) pitch of 150 μm with a standard deviation of 2.37 μm . Figure 6(b) shows the histogram for the pitch of the electrode sites of a finished electrode. The electrode sites were also centered around the ideal pitch of 150 μm with a standard deviation of 3.89 μm and had a slightly wider distribution compared to the micromold holes.

Figure 6(c) and Figure 6(d) show histograms for the distance off the centerline for the machined micromold holes and electrode sites, respectively. Both the micromold holes and electrode sites were centered around the centerline. The micromold holes were tightly clustered with a standard deviation of 1.35 μm and the electrodes sites had a slightly wider distribution with a standard deviation of 3.32 μm . All measurements were taken from a single representative micromold and MEA.

Lack of geometry control is often a criticism of electrodes utilizing an element of assembly (Bhandari et al. 2010), but these measurements show that it is possible to achieve a high level of positional accuracy with microassembly methods. The positional accuracy of the machined micromold holes is mostly defined by the accuracy of the machining tools used to machine the micromold. In the case of our CNC mill, the specified positional accuracy is $\pm 5 \mu\text{m}$ with a repeatability of $\pm 3 \mu\text{m}$.

The increase in the positional distribution of the electrode sites was attributed to the difference between the micromold hole diameter ($\sim 50 \mu\text{m}$) and the wire diameter ($\sim 30 \mu\text{m}$). To reduce the effect this diameter difference would have on the positional accuracy, all the

wires were pulled in tension in the same direction. This placed all of the wires on the same side of the micromold holes, increasing the positional accuracy of the electrode sites.

3.3 Wedge bond impedance

The wedge bond pairs had an average impedance of 0.60 ohms with a standard deviation of 0.02 ohms. This wedge bond impedance was insignificant compared to the impedance from the series capacitance of the metal-electrolyte double layer formed at the electrode sites, which can range from low kOhms to MOhms. Wedge bonds were also attempted using the wedge bonder at room temperature, but it was not possible to make an electrical connection under these conditions. This indicates that the temperature of the wedge bonder played a critical role in removing the insulation of the wire to allow the formation of reliable wedge bonds.

3.4 Electrode site impedance

The impedances of the electrode sites with different surface conditions are shown in Figure 7. The average impedances of the as-cut, conditioned and iridium oxide plated electrode sites were respectively 850 kOhms, 424 kOhms, and 167 kOhms. SEM images of the as-cut electrode sites are shown in Figure 8(a). When compared to the conditioned electrode sites shown in Figure 8(b), it is clear that the conditioning process increases the surface area of the electrode sites by producing a complex and porous surface.

At these low voltage and current levels, the impedance is dominated by the series capacitance of the metal-electrolyte double layer (Cheung 2007; Wise et al. 2008). This capacitance is directly proportional to the electrochemical surface area (ESA) of the electrode sites (Trasatti and Petrii 1992; Stieglitz et al. 2000) and is believed to be the main cause in the change of impedance between the as-cut and conditioned sites. Based on these impedance measurements, it was calculated that the conditioned sites ESA is, on average, two times larger than the as-cut ESA. The electrodeposited iridium oxide sites had the lowest impedance, consistent with other results reported in the literature (Meyer et al. 2001). Figure 8(c) shows SEM images of an electrodeposited iridium oxide site. The lower impedance of iridium oxide was also attributed to an increase in ESA, but the porous structure of iridium oxide is not visible at these magnifications (Weiland et al. 2002).

3.5 Cyclic voltammetry

Figure 9 shows the cyclic voltammogram of the iridium oxide electrode with the shaded area representing the area used to calculate the cathodic charge storage capacity (CSC_C). The calculated CSC_C of 64 mC cm^{-2} was larger than typically reported for electrodeposited iridium oxide on a smooth gold surface (Meyer et al., 2001). This larger CSC_C was attributed to the three-dimensional structure of conditioned gold surface and the electrodeposited iridium oxide coating exemplified in Figure 8(b-c) (Cogan 2008). The overall shape of the cyclic voltammogram was typical for electrodeposited iridium oxide (Meyer et al. 2001; Lu et al. 2009) with the distinctive peaks indicating reduction-oxidation reactions of iridium oxide as electrons transfer across the metal-electrolyte interface (Weiland et al. 2002).

The CSC_C was calculated using the area highlighted in Figure 9 and the fact that the potential is directly related to time through the sweep rate (Cogan 2008). The calculated ESA from the impedance measurements for the conditioned gold sites was used to determine surface area for the current density calculations. Using the doubling of ESA between the as-cut and conditioned sites and assuming the as-cut ESA/GSA ratio was equal to unity, each conditioned site has an ESA of approximately $1012 \text{ } \mu\text{m}^2$.

3.6 Acute *in vivo* recordings

We tested the efficacy of the MEA for recording extracellular neural spike activity by recording responses of neurons in the central nucleus of the inferior colliculus (ICC). Stimuli were 40-ms tone bursts varied systematically in frequency and sound level. The ICC is a structure of the midbrain that contains an orderly representation of sound frequency (Rose et al. 1963). The most-sensitive characteristic frequencies (CFs) of neurons increase systematically with neural position from dorso-lateral to ventro-medial – this is referred to as tonotopic organization.

In anesthetized cats, we inserted the MEA approximately parallel to this tonotopic axis, such that the most distal electrode sites (i.e., near the tip of the MEA) encountered neurons with high CFs, up to ~40 kHz, and the most proximal sites encountered neurons with CFs around 0.5 to 1 kHz. In each animal, the MEA penetrated the pia mater of the inferior colliculus easily, with little dimpling of the tissue. Results from one animal shown in Figures 10-12 are representative of those obtained in both animals. Extracellular spike waveforms recorded from one MEA site are shown in Figure 10; 32 traces from the same unit are overlaid. Waveforms from all 32 MEA recording sites are shown across a longer time scale in Figure 11. The traces are numbered 1 to 32, indicating recording from the deepest to most superficial MEA sites. Figures 11(a), (b), and (c) show the responses to tones with frequencies of 2, 8, and 32 kHz, respectively. In each panel, one can see a burst of spikes starting around 15 ms relative to the stimulus onset. The active sites shifted systematically deeper in the ICC from around MEA sites 22 to 15 to 6 as the frequency increased. Signal-to-noise ratios and spike waveforms were similar to those obtained by Middlebrooks and Snyder (2007) using commercial silicon-substrate recording probes (NeuroNexus Technologies, Inc, Ann Arbor, Michigan).

Frequency response areas (FRAs) recorded with the MEA are shown in Figure 12. Each panel shows the FRA recorded at one site; i.e., one depth in the ICC. The panels are arranged from top to bottom and then left to right, with site numbers decreasing from superficial (upper left) to deep (lower right) as in Figure 11. In each panel, horizontal and vertical axes plot stimulus frequency and sound level, and colors indicate mean spike rates. In most panels, one can see a typical V-shaped FRA, which is characteristic of the sharp frequency tuning of neurons in the ICC. The tuning shifts progressively from low frequencies to higher frequencies with increasing ICC depth. The sharpness of frequency tuning and the tonotopic increase of characteristic frequency with increasing depth were similar to those obtained using a commercial silicon-substrate recording probe (Middlebrooks and Snyder 2007).

4 Conclusion

In this paper, we demonstrated a 32-channel shank MEA that was fabricated using a novel hybrid micromachining process that combines precision machining and micro-assembly with MEMS-style fabrication. This approach enables one to design highly integrated microelectrode assemblies with complex 3-D topologies, while utilizing commercially available manufacturing. Our work yielded high quality, functional devices that demonstrated performance that was comparable to commercial devices made using silicon-based manufacturing. The advantage to this manufacturing approach is that it affords a great amount of flexibility in design, including the ability to integrate multiple dissimilar materials without the need for a planar surface. By combining MEMS and lithography with precision machining and micro-assembly, one can produce a wide variety of microelectrode types. For example, MEAs manufactured using this approach may integrate sensors, optical fibers, microfluidics, mechanical components, flexures, drug eluting products, and biodegradable materials.

Acknowledgments

We wish to thank Spencer Chang for his help with the electrode fabrication and CNC operations and Richard Chang for his expertise and assistance in SEM sample preparation. We are grateful to Prof. Reginald Penner and Keith Donavan for providing access to their potentiostat and assisting with the impedance measurements. We appreciate the helpful comments and insightful suggestions provided by the reviewers. Hybrid microfabrication and related devices are patent pending. We gratefully acknowledge support from the Broadcom Foundation and the National Institute on Deafness and Other Communication Disorders contract N263-2009-00024C.

References

- Bhandari R, Sandeep N, Solzbacher F. *Biomed. Microdevices*. 2010; 12(5):797–807. [PubMed: 20480240]
- Chae J, Park S, Freiheit T. *Int. J. Mach. Tools Manuf.* 2006; 46(3-4):313–332.
- Cheung KC. *Biomed. Microdevices*. 2007; 9(6):923–938. [PubMed: 17252207]
- Cheung KC, Renaud P, Tanila H, Djupsund K. *Biosens. Bioelectron.* 2007 doi:10.1016/j.bios.2006.08.035.
- Churchland MM, Yu B, Sahani M, Shenoy KV. *Curr. Opin. Neurobiol.* 2007; 17(5):609–618. [PubMed: 18093826]
- Cogan SF. *Annu. Rev. Biomed. Eng.* 2008; 10:275–309. [PubMed: 18429704]
- Dhanorker A, Ozel T. *Int. J. Mechatron. Manuf. Syst.* 2008 doi:10.1504/IJMS.2008.018273.
- Donoghue JP, Nurmikko A, Black M, Hochberg LR. *J. Physiol.* 2007 doi:10.1113/jphysiol.2006.127209.
- Dornfeld D, Min S, Takeuchi Y. *CIRP Ann. Manuf. Technol.* 2006 doi:10.1016/j.cirp.2006.10.006.
- Fang FZ, Liu K, Kurfess TR, Lim GC. *MEMS/NEMS*. 2006 doi:10.1007/0-387-25786-1_18.
- Huang W, Wang M, Zheng J, Li Z. *J. Phys. Chem. C*. 2009 doi:10.1021/jp8095693.
- Hung A, Goldberg IB, Judy JW. *Biol. Med. Phys., Biomed. Eng.* 2010 doi:10.1007/978-0-387-98120-8_7.
- Jackson A, Fetz EE. *J. Neurophysiol.* 2007; 98(5):3109–3118. [PubMed: 17855584]
- Jellema T, Weijnen JA, Neurosci J. *Methods*. 1991; 40(2-3):203–209.
- Kewley D. *Sens. Actuators A: Phys.* 1997 doi:10.1016/S0924-4247(97)80221-X.
- Kipke DR, Vetter RJ, Williams JC, Hetke JF. *IEEE Trans. Neural Syst. Rehabil. Eng.* 2003; 11(2):151–155. [PubMed: 12899260]
- Lehew, G.; Nicolelis, MAL. *Methods for Neural Ensemble Recordings*. Nicolelis, MAL., editor. CRC Press; Florida: 2008. p. 1-20.
- Loeb G, Walker A, Uematsu S, Konigsmark B. *J. Biomed. Mater. Res.* 1977; 11(2):195–210. [PubMed: 323263]
- Lozano A, Hutchison W, Kiss Z, Tasker R, Davis K, Dostrovsky J. *J. Neurosurg.* 1996 doi:10.3171/jns.1996.84.2.0194.
- Lu Y, Wang T, Cai A, Cao Y, Yang H, Duan YY. *Sens. Actuators B: Chem.* 2009; 137(1):334–339.
- Luttge R, Berenschot EJW, de Boer MJ, Altpeter DM, Vrouwe EX, van den Berg A, Elwenspoek M. *J. MEMS*. 2007 doi:10.1109/JMEMS.2007.899339.
- Manko, HH. *Solders and Soldering: Materials, Design, Production, and Analysis for Reliable Bonding*. 4th edn.. McGraw-Hill; New York: 2001. p. 102
- Masuzawa T, Tönshoff H. *CIRP Ann. Manuf. Technol.* 1997 doi:10.1016/S0143-974X(98)00174-6.
- Meister M, Pine J, Baylor DA, Neurosci J. *Methods*. 1994 doi:10.1016/0165-0270(94)90030-2.
- Mercanzini A, Cheung K, Buhl D, Boers M, Maillard A, Colin P, Bensadoun J, Bertsch A, Renaud P. *Sens. Actuators A: Phys.* 2008 doi:10.1016/j.sna.2007.07.027.
- Meyer RD, Cogan SF, Nguyen TH, Rauh RD. *IEEE Trans. Neural Syst. Rehabil. Eng.* 2001 doi:10.1109/7333.918271.
- Middlebrooks JC, Snyder RL. *J. Assoc. Res. Otolaryngol.* 2007 doi:10.1007/s10162-007-0070-2.
- Middlebrooks JC, Snyder RL. *J. Neurosci.* 2010 doi:10.1523/JNEUROSCI.4949-09.2010.

- Nicolelis M, Dimitrov D, Carmena JM, Crist R, Lehew G, Kralik JD, Wise SP. PNAS. 2003 doi: 10.1073/pnas.1934665100.
- Nicolelis MAL, Ghazanfar AA, Faggin BM, Votaw S, Oliveira LMO. Neuron. 1997 doi:10.1016/S0896-6273(00)80295-0.
- Rose JE, Greenwood DD, Goldberg JM, Hind JE. J. Neurophysiol. 1963; 26(2):294–320.
- Schmidt EM, Bak MJ, McIntosh JS. Exp. Neurol. 1976 doi:10.1016/0014-4886(76)90220-X.
- Seymour JP, Langhals NB, Anderson DJ, Kipke DR. Biomed. Microdevices. 2011 doi:10.1007/s10544-011-9512-z.
- Stacey WC, Litt B. Nat. Clin. Pract. Neurol. 2008 doi:10.1038/ncpneuro0750.
- Stieglitz T, Beutel H, Schuettler M, Meyer JU. Biomed. Microdevices. 2000; 2(4):283–294.
- Trasatti S, Petrii OA. J. Electroanal. Chem. 1992; 327(1-2):353–376.
- Wang M-F, Maleki T, Ziaie B. J. Micromechanics Microengineering. 2010 doi: 10.1088/0960-1317/20/3/035013.
- Ward MP, Rajdev P, Ellison C, Irazoqui PP. Brain Res. 2009 doi:10.1016/j.brainres.2009.05.052.
- Weiland JD, Anderson DJ, Humayun MS. IEEE Trans. Biomed. Eng. 2002 doi:10.1109/TBME.2002.805487.
- Williams JC, Rennaker RL, Kipke DR. Brain Res. Protoc. 1999 doi:10.1016/S1385-299X(99)00034-3.
- Wise KD, Anderson DJ, Hetke JF, Kipke DR, Najafi K. Proc. IEEE. 2004 doi:10.1109/JPROC.2003.820544.
- Wise KD, Angell JB, Starr A. IEEE Trans. Biomed. Eng. 1970 doi:10.1109/TBME.1970.4502738.
- Wise KD, Sodagar AM, Yao Y, Gulari MN, Perlin GE, Najafi K. Proc. IEEE. 2008 doi:10.1109/JPROC.2008.922564.

Fig. 1.

(a) A 32-channel shank MEA manufactured using hybrid microfabrication with attached connector. **(b)** Shank of a 32-channel MEA on a dime. **(c)** Tip of a 32-channel shank MEA with the electrode sites facing the camera

Fig. 2.
Line drawing of a shank MEA with important dimensions and features labeled

Fig. 3.

A schematic outlining the hybrid microfabrication steps of a shank MEA with a chisel tip. **(a)** Machine acrylic micromold on a CNC mill using micro end mills and micro drills. **(b)** Microassemble insulated gold wires into acrylic micromold. **(c)** Wedge bond insulated gold wires onto an interface PCB. **(d)** Inject epoxy into micromold with assembled wires. **(e)** After epoxy cures, flip micromold upside-down and cut nubs flush on a CNC mill (shown) or with a precision razor blade (not shown). **(f)** Define chisel tip by CNC machining cured epoxy and acrylic micromold with an end mill. **(g)** Top view of chisel tip machining. **(h)** Dissolve acrylic micromold in acetone to release electrode. **(i)** Prepare gold sites by conditioning and electrodeposition of iridium oxide. All views are cross-section views with the exception of **(g)** which is a top view. Drawings are not to scale. Dimensions are listed for reference

Fig. 4. (a) The tip of a single-sided 32-channel shank MEA, (b) a 16-channel MEA with electrode sites on three sides, (c) orthogonal view of a three-sided MEA and (d) a MEA with an embedded optical fiber transmitting a green laser. All MEAs were fabricated using hybrid microfabrication

Fig. 5.
(a) A 32 channel micromold for a shank MEA. (b) Close-up of a 32 channel micromold with 30 μm diameter wire assembled in two of the micromold holes

Fig. 6. Histograms of measured pitch for **(a)** micromold holes (29 samples) and **(b)** electrode sites (21 samples) and for distance off of centerline for **(c)** micromold holes (31 samples) and **(d)** electrode sites (22 samples). The dashed line represents the ideal, as designed dimension. Micromold holes were 50 μm in diameter and electrode sites were 25 μm in diameter. Error in measurement technique was approximately 1 μm

Fig. 7. Log scale bar chart comparing the impedances of gold electrode sites in the as-cut, conditioned and electrodeposited iridium oxide (IrOx) states. Diameter of sites were 25.4 μm

Fig. 8. SEM images of **(a)** an as-cut gold electrode site, **(b)** a conditioned gold electrode site and **(c)** iridium oxide electrodeposited onto a conditioned gold site

Fig. 9.

Cyclic voltammogram of a MEA with electrodeposited iridium oxide in PBS at a 50 mV/s scan rate. Calculated ESA was used to determine current density. The shaded area represents a CSC_C of 64 mC cm⁻²

Fig. 10.

Spike waveforms from a single recording site. These waveforms were selected on the basis of a negative-going peak followed 200 to 320 μ s later by a positive-going peak of criterion amplitude. Thirty-two waveforms are superimposed

Fig. 11.

Spike waveforms from 32 MEA recording sites elicited by 40-ms tone bursts at frequencies of (a) 2, (b) 8, and (c) 32 kHz. Recording sites were spaced at 150- μ m intervals and were numbered increasing from the deepest (tip) site to the most superficial. In this example, site 26 was not connected

Fig. 12.

Frequency response areas (FRAs) recorded from 32 MEA sites (same case as in Fig. 11). Each panel represents the FRA recorded at one site (i.e., one depth in the ICC). In each panel, the vertical axis shows the sound level in dB attenuation relative to the maximum level, the horizontal axis shows the tone frequency, and colors represent mean spike counts. Most panels show a typical V-shaped tuning curve. The characteristic frequencies increased from superficial (site 32) to deep (site 2). Site 26 was not connected, and site 1 apparently was tuned to frequencies above 40 kHz, which was the limit of our calibrated audio system. Many of the sites (e.g., 7 to 21) show evidence of high frequency distortion products generated by high-level low-frequency tones.