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Long term *in-vitro* functional stability and recording longevity of fully integrated wireless neural interfaces based on Utah Slant Electrode Array (USEA)

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Abstract

We evaluate the encapsulation and packaging reliability of fully integrated wireless neural interface based on Utah Slant Electrode Array/integrated neural interface-recording version 5 (USEA/INI-R5) system by monitoring the long term *in-vitro* functional stability and recording longevity. The integrated neural interface (INI) encapsulated with 6 μm Parylene-C was immersed in phosphate buffered saline (PBS) for a period of over 276 days (with the monitoring of the functional device still ongoing). The full functionality (wireless radio-frequency power, command and signal transmission) and the ability of the electrodes to record artificial neural signals even after 276 days of PBS soaking with little change (within 14%) in signal/noise amplitude constitutes a major milestone in long term stability, and allows to study and evaluate the encapsulation reliability, functional stability, and its potential usefulness for wireless neural interface for future chronic implants.

1. Introduction

Implantable neural interfaces have attracted much attention to the neurophysiologists and clinicians for use in prosthetics that can help restore sensory function, physiological monitoring and functioning of neuronal activities in patients with neurodegenerative diseases or movement dysfunctions [1-6]. One example of this technology is Utah Electrode Array (UEA)/Utah Slant Electrode Array (USEA) microelectrodes that are designed to record/stimulate many neurons simultaneously [5, 7, 8]. The traditional approach is to connect the devices to external instrumentation via relatively bulky wirebundles and percutaneous connectors. However, the percutaneous connectors are likely to cause infections and limit the use of implant for chronic applications [9]. To eliminate the use of wired connections, significant research efforts have been put recently to develop wireless implantable neural interfaces with wireless transmission of power and data [10-15]. In this context, the University of Utah has been developing fully integrated wireless neural interfaces capable of recording/stimulating neural activity from 100 electrodes. In a fully integrated wireless neural interface, an integrated circuit (IC) that is capable of amplifying, digitizing, and transmitting the neural data out wirelessly, is flip chip bonded to the backside

of UEA/USEA [16, 17]. The power, clock and command signals to the IC are also supplied wirelessly via an inductive link [18].

These implantable wireless integrated neural interfaces (INI) must be intended to function *in-vivo* for years. The failure of chronic neural implants can occur due to both physiological reasons (aggressive immune system response and infection, leading to death of neural cell tissue in the vicinity of the device) as well as failure of the encapsulation, leading short circuits, delamination of films or corrosion of contacts and interconnects, rendering the active and passive electronic components in the device non-functional or destroying them permanently. The packaging and encapsulation becomes crucial as the implantable neural interfaces are submersed in biological fluids containing various organic/inorganic and saline environments that are extremely corrosive and reactive. These challenges have lead to the development and investigation of various encapsulation materials: for example, Urethanes, polyimide, Teflon, Parylenes, silicon nitride (Si_3N_4), amorphous silicon carbide ($a\text{-SiC}$), diamond-like carbon (DLC) and historically known silicones [19-25]. The hermiticity verifications are based on measurements of electrical leakage, resistivity, impedance, transport rates for H_2O and ionic species, dissolution rates from infrared (IR) spectra [22, 24, 25]. With the requirement of reliable hermiticity, selective removal of the electrode tips and the biocompatibility issues makes the selection of encapsulating material much more complicated. A single material that can satisfy biocompatibility, and all the aforementioned criteria is difficult to find. For example, Si_3N_4 has high electrical resistivity but is susceptible to dissolution when subjected to biased soak conditions [24], whereas the polymeric materials have good biocompatibility and insulation but exhibit poor adhesion that require pre-cleaning and adhesion promoter on the surface [24, 25]. Therefore, selecting the packaging and encapsulation material that can protect the electronic circuitry of neural interface from the harsh *in-vivo* environment is a major challenge.

Parylene-C is a biologically inert material capable of being deposited in conformal nanoscale layers and is easily patternable to expose the electrode tips for sensing, making it a viable material for the implant encapsulation [22, 25-28]. The impedance and the leakage measurements on the interdigitated electrodes (IDEs) test structures in phosphate buffered saline (PBS) have shown that encapsulation and insulation properties of Parylene-C are stable for more than one year [22]. However, in a fully integrated wireless device the variation in the geometry, surface properties, and several additional processing steps that are not involved in the IDE's test structures can invalidate the prediction of failure. For example, variations in topography due to surface mounting devices (SMDs), inductive receiver coil, etc. and the variation in surface properties of these components due to involvement of different materials may result in poor adhesion of Parylene-C. The processes during flip chip integration (interconnects between USEA and IC) can create extra interfaces and surface contamination, or the tip deinsulation by plasma can result in minor delamination in the vicinity. All these factors can alter the diffusion rates of mobile ions present in PBS, and the hydrolytic effects in a fully integrated wireless device in comparison to the IDEs and does not necessarily assure an acceptable packaging and encapsulation for a chronic implant.

In the present study, we report the long term *in-vitro* functional stability and recording longevity of fully integrated and encapsulated USEA/INI-R5 (integrated neural interface-recording version 5) systems (designed to record from peripheral nerves), and evaluate the encapsulation and packaging reliability in the wireless neural interfaces. *In-vivo* wireless recording of neuronal data using fully integrated USEA/INI-R5 has been recently demonstrated in an acute experiment [29, 30]. The USEA/INI-R5 is capable of powered and configured wirelessly through 2.765 MHz inductive link, and the neural data can also be transmitted wirelessly via radio-frequency (RF) (900 MHz Industrial, scientific, medical-

ISM band) telemetry link. The functional stability of USEA/INI-R5 device was investigated in PBS (to simulate physiological environment) at room temperature (RT). The USEA/INI-R5 employed biocompatible polymer Parylene-C as an encapsulation layer and was immersed in PBS for a period of over 276 days (the device is still in saline and is being monitored continuously). The INI device, while being soaked was powered and configured wirelessly and the transmitted frequency shift keying (FSK) modulated RF signal was also recorded wirelessly as a function of soak time. Further, the *in-vitro* recording of the artificial neural signals was performed in agarose from time to time by introducing the signals into the agarose, and recording the artificial action potentials wirelessly. Our current *in-vitro* stability (>276 days) of the USEA/INI-R5 provides a measure of the packaging and encapsulation reliability, and the functional stability in a fully integrated wireless neural interface that has not been reported previously in wireless implantable devices.

2. Experimental details

The schematic and the photograph of an integrated wireless neural interface assembly with the INI chip, SMD components and the Au (1% Pd)-wire wound receiver coil are shown in figure 1(a) and (b). A 100-channel wireless neural recording IC designated as INIR-5 (integrated neural interface recording version-5), was fabricated in a commercially available 0.6- μm 2P3M BiCMOS process at X-fab Semiconductors. The details of the system integration including design, fabrication and the chip design are described elsewhere [12, 16]. The INI-R5 IC was flip chip bonded to 10 $\times$ 10 USEA using Au/Sn reflow soldering. SMDs 0402 (smoothing and the tuning capacitors) were bonded to the back of the USEA and connected to the chip via backside metallization. An inductive wireless link provides power, clock and command signals to the chip. For inductive power and command reception, a flat spiral coil 5.5 mm in diameter was manufactured by winding insulated 56 μm diameter Au (1% Pd) wire [31]. The coil was connected to the INI chip by wire bonding using a semi-automatic wire bonder (West Bond Model 545657E, West Bond Inc.) to the bond pads on the backside of the USEA. Silquest A-174® silane (GE silicones) was vaporized onto the integrated device as an adhesion promoter for Parylene. Finally, the biocompatible polymer Parylene-C with a thickness of 6 μm was chemically vapor deposited using a Paratech 3000 labtop deposition system (Paratech Coating, Inc.) [22]. The electrode tips for sensing were deinsulated by exposing the tips (30-50 μm) with an aluminum foil mask followed by a DC pulsed oxygen plasma (Oxford Plasmalab 80 Plus PECVD system, Oxford Instruments). The details of the tip de-insulation process has been described elsewhere [22].

Custom made transmit coil that uses a class E power amplifier to create a 2.765 MHz waveform of up to 80 V_{rms} at a 10 V supply was used to deliver the inductive power. The transmit board is connected to the PC through the National Instrument (NI) board in order to send a command by amplitude modulation of the inductive power. The resulting AC magnetic field was inductively coupled to the receiving coil present in the INI device and was supplied to the INI chip in the device. The transmitted RF signal was detected using two different antenna-receiver systems. A monopole antenna connected to the spectrum analyzer (Agilent E4402B) was used for displaying the RF spectra. The second antenna receiver system was a custom developed wireless receiver board that displays the frequency and signal strength, and decodes the real time information in MATLAB on a PC through the universal serial bus (USB) interface. PBS (1% NaCl) in a covered glass Petri dish with 11 mm depth was used for long term soak testing (PBS depth was maintained at constant level all the time). For generating the artificial neural signals into the agarose, a Grass SD-9 pulse stimulator was used.

3. Results and discussion

3.1. RF functionality in PBS

First, the initial bench-top testing of fully integrated and encapsulated INI device was performed to check the wireless power/command reception and responsiveness to change the telemetry frequency in PBS. Figure 2 shows the RF spectra transmitted wirelessly from the fully integrated USEA/INI-R5 neural interface immersed in PBS, measured using the monopole antenna connected to the spectrum analyzer. Upon reception of the wireless inductive power, the RF signature appeared with peak at 934.8 MHz and signal strength of -89 dBm was measured at 8 cm antenna distance from the device. The chip in the INI device contains an 8-bit register that can take a value between '0' and '255' and sets the command transmitter frequency in the range of 902-928 MHz (ISM band) [12]. The responsiveness of the INI device to the commands for two different telemetry frequencies ('0' and '4') are depicted in figure 2 to show the full functionality of the INI device in PBS. The strengths of the transmitted RF signal for the two telemetry frequencies are close to -87 dBm. The FSK modulated RF signal characteristics analyzed on wireless receiver board are summarized in table 1 (corresponding measurements from spectrum analyzer are also shown for comparison). The wireless receiver measured the RF signal for the telemetry frequency '0' at 911.2 MHz with strength of -57 dBm at 8 cm from the device. The discrepancies between the signal strengths measured by receiver board and the spectrum analyzer can be expected because of two different antenna-receiver systems. However, the important information to be noted here is that the INI device is fully functional i.e. powers up as well as responds to the telemetry frequency commands while being immersed in PBS.

3.2. Long term functional stability with soak time in PBS

In order to investigate the long term functional stability of the INI device in PBS, the FSK modulated RF signal for telemetry frequency '0' that corresponds to 911.9 MHz on a spectrum analyzer was monitored as a function of soak time. Figure 3(a) shows the peak RF signal strength and the respective frequency as a function of soak time that were extracted from the spectra measured on the spectrum analyzer. For clarity, the extracted values are plotted for every after 1 h, although the RF spectra were measured every after 20 min. (as depicted in the inset for few consecutive intervals). Corresponding measurements using the wireless receiver board are shown in 3(b). It can be seen from figure 3(a) and 3(b) that the RF functionality of the fully integrated INI device is maintained in PBS even after 276 days. It should be noted here that during this period of soak time, the device was never taken out of the PBS solution except when agarose testing was preformed. The variation in the RF signal strength and the transmission frequency lies within 15% and 0.3% of the initial values, respectively. Further, the responsiveness of the INI device to the commands for changing the telemetry frequency was checked at $t=150$ days, $t=211$ days, and $t=276$ days. The RF signal strengths and the corresponding frequencies for two distinct telemetry frequencies ('0' and '4') after different soak times are compared in table 1. It was observed that the position of the transmitted RF signal was shifted to lower frequency side for both the power (not shown here) and commands (see table I, also clear from figure 3). Additionally, it appears that there is an oscillating pattern in the RF signal strength which is almost periodic. This oscillating pattern in the signal strength could possibly due to the variations in the temperature as the data was recorded at room temperature. However, the INI device continued to receive power and commands, and transmitted the RF signature even after 276 days of soaking in PBS at RT. This long term full functionality in the wireless RF signal clearly demonstrates that the electronic circuitry has remained unaffected in the PBS solution even after more than 9 months of soaking. Therefore, the Parylene-C coating is providing an effective protection of the INI from ingress of the moisture and other ionic species that are present in the PBS.

3.3. Long term in-vitro recording ability of the INI device

In order to test the long term recording ability of the electrodes in the INI device that was soak tested, *in-vitro* wireless recording was performed in agarose gel in a petri dish for few channels at time $t=0$, 108, 150, 211, and 276 days of PBS soaking. The experimental set-up for *in-vitro* wireless recording of artificial neural signals is shown in figure 4. The agarose impedance measured between the testing probe and the reference Pt electrode separated at 8 cm distance was 5 k Ω at 1 kHz frequency (10 nA current signal). The impedance of the agarose gel for the given distance was kept similar every time while performing the *in-vitro* testing after different soak periods of time. The integrated neural interface was taken out from the PBS solution, and was inserted into the agarose gel in the petri dish (underneath the transmit coil in figure 4). The artificial neural signals were introduced into the agarose gel from a SD-9 Grass stimulator using the Pt electrodes. The input signal from the SD-9 consisted of a biphasic cyclic voltage pulse (2 V_{pp}) with a frequency of 12 pulses/s (0.4 ms duration). During artificial neural signal recordings, the Pt electrodes for injecting the signal into agarose were also separated by 8 cm for maintaining the similar impedance and consistency voltage drop (for μm lengths, the voltage will be dropped to a few hundred mV). During the wireless recording of the artificial neural signals in agar, the telemetry frequency at 912.3 MHz ('ch 0') showed the lowest bit error rate ($<10^{-2}$) on the receiver board, and therefore, was used for the real time decoding in the MATLAB GUI program.

Figure 5 shows the real time wirelessly recorded artificial action potentials from one channel at $t=0$ and after soaking the INI device in PBS for $t=108$ days, $t=150$ days, $t=211$ days, and $t=276$ days. The inset compares the recording of the artificial action potentials (for soak time $t=0$ days and $t=276$ days) at different time scale. The peak-to-peak voltage (V_{pp}) of the artificial action potential waveform that was wirelessly recorded from the INI at $t=0$, soak time $t=108$ days, soak time $t=150$ days, soak time $t=211$ days, and soak time $t=276$ days is 745 ± 32 μV , 723 ± 13 μV , 691 ± 97 μV , 783 ± 50 μV and 796 ± 25 μV , respectively (these values are average of V_{pp} that were recorded for over 60 s in real time). The respective noise levels were 99 ± 15 μV , 101 ± 26 μV , 98 ± 2 μV , 92 ± 8 μV , and 111 ± 4 μV . The variance in the recorded signal/noise amplitude lies within 14% and indicates that the long term soaking did not have much influence on the tip deinsulated areas of the electrodes, and the ability to record the artificial neural signals. Similar long term recoding ability was observed for few other channels/electrodes (not shown here). The mean and the standard deviation of the voltages recorded for 4 different electrodes at soak time $t=276$ days was 722 μV and 81 μV , respectively. These *in-vitro* recordings show that for a given channel the ability to track the artificial action potential is maintained even after 276 days of PBS soak. Further, some of the integrated and encapsulated INI devices implanted in animals in acute experiments could survive the impact of the pneumatic inserter as well as immersion in the real biological environment for several hours [29, 30]. The devices successfully recorded and wirelessly transmitted digitized electrical activity in live anesthetized animals. The *in-vitro* stability together with the acute animal experiments demonstration constitute major milestone, the encapsulation reliability, functional stability, and the recording longevity in a fully integrated wireless neural interface.

4. Conclusion

In summary, the encapsulation and packaging reliability of Parylene-C in fully integrated wireless neural interfaces is evaluated by monitoring the long term *in-vitro* functional stability and recording longevity in USEA/INI-R5 (integrated neural interface-recording version 5) system. The device was fully functional (wireless RF power/command and data transmission) even after 276 days soaking without significant change in the transmitted RF signal strength and the transmission frequency. Additionally, the *in-vitro* recording of the artificial neural signals performed in agarose shows the ability of the channels in the INI

device to track artificial action potentials with signal/noise amplitude within 14% variation for more than 276 days of PBS soaking. These *in-vitro* results provide a measure of the encapsulation reliability, functional stability, and the recording longevity in a fully integrated wireless neural interface. The evaluation of the neural interface in PBS over 9 months demonstrates its stability in a real physiological environment, and the usefulness of wireless neural interface for future chronic implants.

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Figure 1. (a) Schematic, and (b) the photograph of the Utah Slant Electrode Array/integrated neural interface-recording version 5 (USEA/INI-R5) device.

Figure 2.

Frequency shift keying (FSK) modulated radio-frequency (RF) spectra transmitted wirelessly from the fully integrated and encapsulated USEA/INI-R5 in phosphate buffered saline (PBS) upon power up, and command telemetry frequency '0' and telemetry frequency '4'.

Figure 3.

Transmitted wireless RF signal monitored for the telemetry frequency '0' as a function of soak time in PBS (antenna was @ 8 cm from the device). (a) Peak RF signal strengths and the respective frequencies as extracted from the spectra measured using the spectrum analyzer (inset shows few consecutive RF spectra at 20 min. intervals). (b) RF signal strengths and the respective frequencies as monitored on custom-developed wireless receiver board.

Figure 4.
Experimental set-up for wireless recording of artificial neural signals in agarose.

Figure 5.

In-vitro wirelessly recorded artificial action potentials that were injected from Grass SD-9 stimulator into the agarose dish. Recording is shown from one channel at time $t=0$, at soak time $t=108$ days, at soak time $t=150$ days, at soak time $t=211$ days and at soak time $t=276$ days to demonstrate the recording longevity of particular channel in UTAH/INI-R5 device after PBS soaking. Inset shows the recording of the artificial action potentials at $t=0$, and at soak time $t=276$ days at different time scale.

Table 1

FSK modulated wireless radio-frequency (RF) signal characteristics of USEA/INI-R5 device measured through the PBS using a custom made wireless receiver, and the spectrum analyzer, when antenna was placed at 8 cm from the device.

Soak time	Telemetry frequency	RF signal using wireless receiver	RF signal using spectrum analyzer
$t=0$	'0'	911.2 MHz, -57 dBm	911.9 MHz, -87.4 dBm
	'4'	912.5 MHz, -59 dBm	913.5 MHz, -86.9 dBm
$t=150$ days	'0'	910.6 MHz, -57 dBm	910.8 MHz, -80 dBm
	'4'	912.5 MHz, -57 dBm	912.6 MHz, -81 dBm
$t=211$ days	'0'	910.1 MHz, -55 dBm	910.5 MHz, -84 dBm
	'4'	911.9 MHz, -60 dBm	912.0 MHz, -83.3 dBm
$t=276$ days	'0'	910.1 MHz, -59 dBm	910.5 MHz, -84.5 dBm
	'4'	911.6 MHz, -60 dBm	911.8 MHz, -84.9dBm