Chronic multi-modal monitoring of neural activity in rodents and primates

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ABSTRACT

We developed multi-modal systems comprising implantable carbon fiber (CF)-based electrodes to record synchronously chemical (e.g., dopamine) and electrical (e.g., local field potential, LFP) forms of activity in the brain. These systems were equipped with implantable micro-invasive probes and moveable silica-based CF probes capable of recording chronically from fixed locations, or from multiple depths along predetermined trajectories, respectively, spanning 48 spatially distinct sites in the caudate nucleus and the putamen. Electrochemical fast scan cyclic voltammetry (FSCV) was implemented in combination with standard electrophysiology to provide subsecond chemical and electrical recordings. The chronic stability of our micro-invasive probes, as tested previously in rodents and translated for use in nonhuman primates (NHP), was necessary to ensure functional recording from fixed locations in the brain without degradation in probe sensitivity over time. These systems were used to examine the relationship between dopamine and beta-band LFPs, prominent biomarkers of untreated Parkinson’s disease. We recorded dopamine and beta in rhesus monkeys performing oculomotor tasks in which reward valuation and movement control, key functions impaired in Parkinson’s disease, could be quantified. Highly stable measurements of dopamine and LFP neural signals were made over a period of months.

Keywords: multi-modal, dopamine, neurochemical sensor, minimally invasive, brain intracranial implant, chronic, electrochemical recording, fast scan cyclic voltammetry

1. INTRODUCTION

The loss of normal dopaminergic signaling and the amplification of electrical beta-band (13–35 Hz) oscillations are key neurological hallmarks of Parkinson’s disease. Beta-band local field potentials (LFPs) increase excessively and in proportion to the motor disabilities of these patients. L-DOPA replenishes dopamine in the brain, reduces the amplitude of beta LFPs, and helps to restore normal movement control to patients. These observations have led to a theory of an inverse relationship between dopamine (chemical) and beta (electrical) activity. Beta-band LFPs have been used as a proxy for dopamine release, and research is underway to use these electrical signals as biomarkers for guiding treatment. Clinical trials are now in progress to use beta activity measurements for direct control of the therapeutic stimuli produced by invasive deep brain stimulation (DBS) devices for adaptive closed-loop treatment. The utility of beta-band biomarkers in diagnostics and treatment, however, remains disputed and the long-held hypothesis of an inverse relationship between dopamine and beta activity has only been tested very recently. Methods to monitor synchronously both chemical and electrical neural signals are necessary to identify sources of pathological neural processes and behaviors. This type of multi-modal monitoring has been limited by issues such as the degradation over time in recording performance of implanted probes, electrical noise and crosstalk between electrochemical and electrophysiological instrumentation, and the need to introduce a large numbers of invasive implantable probes that would create extensive brain trauma. We recently developed systems to address these issues and allow multi-modal monitoring of chemical and electrical neural activity. Here, we characterize the operations of those systems, as well as their performance over time.

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2. METHODS TO MONITOR MULTI-MODAL NEURAL ACTIVITY

Multi-modal systems were developed to measure dopamine and electrical neural activity synchronously, using fast scan cyclic voltammetry (FSCV) to estimate dopamine release and standard electrophysiology systems for electrical neural activity. These systems were applied in behaving nonhuman primates (NHPs, i.e. rhesus monkeys), to identify the relationship between the dopamine and beta-band oscillatory activity and to understand how their co-active operations mediate mood and motor functions that are critically compromised in Parkinson’s. The multi-modal system incorporates a form-fitting MRI compatible chamber\textsuperscript{18,19} installed on the skull to expose a window of tissue through which probes can be inserted into the brain parenchyma (Fig. 1A and B). Probes (18 or 19 per subject) are inserted into the chamber grid to be lowered to targeted brain sites along trajectories estimated by MRI (Fig. 1C–E).

![Diagram of multi-modal system](image)

Figure 1. Overview of multi-modal system. (A) Illustration of chamber component which incorporates a grid with holes through which probes (red) are inserted to be lowered to brain targets; micro-drives (µ-drives) that are clamped into the grid.
and attach probes to moveable shuttles for subsequent lowering; Mill-Max connectors to which external recording instrumentation can plug into to measure signals from the connected probes; and stainless steel (SS) and Ag/AgCl reference and ground wires. (B) Close-up of the two types of probes (CFM, left, and µIP, right) implanted. (C) MRI showing estimated trajectory through the grid of a probe (probe id: P5) towards a site in the putamen. (D) Photo of chamber system with implanted probes, (E) Map of estimated probe positions towards specific brain areas labeled by color (color label at the bottom) as projected from the chamber-mounted grid. Green dashed line denotes position of the anterior edge of the grid in the illustrated map as well as the photograph in D.

**Implant configuration**

Two types of carbon fiber (CF) probes were implanted and used for multi-modal measurements: rigid silica-threaded microelectrodes (CFMs) that could be driven to lower brain sites by screw operation of the microdrive, and micro-invasive probes (µIPs) that remained fixed in place (Fig. 1B). CFMs have successfully been employed in both rodents and primates, but they typically display lowered sensitivities of dopamine detection over the course of recordings, over weeks and months. The CFMs were sufficiently stiff to be driven to deep (> 15 mm) sites on a weekly to monthly basis, allowing measurements to be made from multiple depths along their implanted trajectories. µIPs are ultra-small (~10 µm diameter) and flexible probes that have been shown to provide long-term function in recording dopamine chemical and electrical neural activity. These were shown to provide lasting dopamine recording functionality in rodents, over the course of over a year, in previous studies. We further demonstrated the ability to record dopamine without degradation in performance in NHPs.

**Addressing crosstalk between FSCV and electrophysiological recording instrumentation**

Concurrent operation of FSCV and electrophysiology required modifications in the implantation hardware as well as offline analysis in order to record both dopamine and electrical local field potential (LFP) neural activity. Separate references, epidural Ag/AgCl and/or stainless steel wires, were used for the FSCV and electrophysiological recording systems to prevent crosstalk and ground-loop noise. A single ground connection was supplied by the FSCV system to be connected directly to the animal through implanted Ag/AgCl wires and/or titanium headposts and intracranial screws. This same connection provides the reference for the FSCV signals. The electrophysiology system also provides a ground terminal and this was only used for reducing environmental electromagnetic noise system by connecting it to the conductive isolation chamber effectively creating a Faraday shield for the animal. Spectral interpolation methods were also developed to remove the electrical artifacts in the LFP recordings generated from the FSCV system. These electrically coupled signals create large artificial spikes in the LFP recordings at a frequency of 10 Hz, the FSCV sampling frequency. Recorded electrical neural signals are converted to the frequency domain for removing this 10 Hz fundamental associated with the applied FSCV waveform as well as its harmonics. With this procedure, it was possible to retain original LFP data with a correlation coefficient of 0.99, as assessed using artificially FSCV-contaminated LFP recordings.

**Multi-modal systems applied to record dopamine and LFPs in primates**

The multi-modal systems were used to record dopamine and LFPs from the striatum of NHPs performing a visually guided reward biased task (Fig. 2). Animals performing this task were head-fixed, and they must make eye movements to targets appearing on the left or right to obtain a small or large amount of liquid food reward. This task allowed quantification of reward size and motor variables (saccadic movements) that are critical aspects of motivated behaviors known to be impaired in patients with Parkinson’s disease. Reward-related dopamine increases were found in the majority of sites in the caudate nucleus (CN), as expected based on the well established function of dopamine in processing positive reward value. On the other hand, most of the sites sampled in the putamen did not show this expected large reward dopamine increases and instead displayed dopamine increases for the small reward. Dopamine responses to reward and movement task variables largely depended on the striatal site (CN versus putamen) as well as location within these subregions. These types of measurements were made over the course of several months to over a year in the NHPs. The µIPs displayed highly consistent measurements of dopamine for the same task conditions repeated over several months (Fig. 3). As found in previous tests in rodents, the µIPs developed for NHP use did not display degradation in performance over time in our subjects. Signals were related to task conditions, differentiating reward size and movement direction, as well as to online indicators of licking, pupil size, heart rate, and reaction times, to attempt to dissect the behavioral functions of both dopamine and beta signals side-by-side.
Figure 2. Recordings of multi-modal, electrical and chemical, neural activity in task performing NHP. Concurrent dopamine (DA) and LFP signals with FSCV color plot (top) showing current changes along DA redox potentials, PCA-computed DA (middle), and the raw LFP waveforms (bottom) with clear modulation of beta-band LFPs during DA changes (close-up inset on top right). Task events are shown above the color plot (key on bottom right).
Figure 3. Chronic longitudinal recording performance of micro-invasive probes implanted in NHPs over the course of several months to over a year. (A) Measured dopamine concentration changes (Δ[DA]) from a µIP as aligned to the target cue (T_on) task event and averaged for big reward (brown) and small reward (green) for fixed contralateral target conditions for a given recording session made on a single day (the number of days the probe has been indwelling in tissue is noted by the post-implant days shown above each plot). (B) Peak Δ[DA] measured from target cue periods for a fixed task condition (big reward, contralateral target) from implanted µIPs as a function of days post-implant. Probe IDs containing the letter ‘c’ indicate measurement sites in CN and ‘p’ indicate measurement sites in putamen and ‘c’ in CN. ‘op10’ was implanted in monkey M2 and the other probes were implanted in monkey M1.

3. CONCLUSION

We demonstrate a minimally invasive implantable platform for synchronous monitoring of both dopamine molecular concentrations and electrical neural activity that provides sustained long-term (> 1 year) performance in nonhuman primates. Chemical signals often precede and regulate the electrical neural signals that are more commonly measured by scientists. A major limitation for our understanding of the co-active chemical and electrical operations mediating our everyday behavior, as well as the debilitating motor and mood deficits observed in Parkinson’s disease, is imposed by the lack of reliable methods to monitor these molecular signals. The multi-modal function of the system that we describe here, capable of recording electrical and chemical neural signals simultaneously, addresses a critical need to expand our view of critical neurochemical processes underlying our ability to make movements or decisions, learn new skills, and perform other important behaviors, as well as how these abilities become compromised in disease. The chronic longitudinal stability of the neural recording functions imparted by these ultra-small implants may have implications in the clinic, where safely implantable sensors could be used for identifying sources of neural dysfunction and dysregulation. Online recording of dopamine may provide a more direct measure of the chemical dysregulation known to subserve Parkinson’s and could be used to steer the therapeutic stimuli delivered by newly developed closed-loop DBS systems, improving overall patient well-being.
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