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Illuminating Seizures: Combined Optical and Electrophysiological Recording Techniques Provide Novel Insights Into Seizure Dynamics

Original Article Citation:: Multimodal in vivo recording using transparent graphene microelectrodes illuminates spatiotemporal seizure dynamics at the microscale.

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Neurological disorders such as epilepsy arise from disrupted brain networks. Our capacity to treat these disorders is limited by our inability to map these networks at sufficient temporal and spatial scales to target interventions. Current best techniques either sample broad areas at low temporal resolution (e.g., calcium imaging) or record from discrete regions at high temporal resolution (e.g., electrophysiology). This limitation hampers our ability to understand and intervene in aberrations of network dynamics. Here, we present a technique to map the onset and spatiotemporal spread of acute epileptic seizures in vivo by simultaneously recording high bandwidth micro electrocorticography and calcium fluorescence using transparent graphene microelectrode arrays. We integrate dynamic data features from both modalities using non-negative matrix factorization to identify sequential spatiotemporal patterns of seizure onset and evolution, revealing how the temporal progression of ictal electrophysiology is linked to the spatial evolution of the recruited seizure core. This integrated analysis of multimodal data reveals otherwise hidden state transitions in the spatial and temporal progression of acute seizures. The techniques demonstrated here may enable future targeted therapeutic interventions and novel spatially embedded models of local circuit dynamics during seizure onset and evolution.

Commentary

Epilepsy affects more than 3.4 million people in the United States,[1] including about 1 million people diagnosed with drug-resistant epilepsy. Ongoing seizures are associated with poor quality of life, cognitive deficits, and a higher risk of death. Despite several treatment options for epilepsy, there remains a dire need for more effective treatments, including precision therapies tailored to individual patients. As some targeted therapeutic approaches depend on precise localization of the ictal-onset zone, such as epilepsy surgery and focal neurostimulation, one obstacle to developing improved therapies is the limited characterization of the spatial and temporal dynamics of seizure onset and evolution. In particular, there is a lack of diagnostic tools that simultaneously have both high spatial and temporal resolution. Over the last hundred years, significant progress has been achieved using electrographic recordings, benefitting from advances in computation and electrode microfabrication. It is now commonplace for diagnostic clinical studies to involve tens to hundreds of intracranial electrode contacts; however, there still remain substantial areas of brain in between electrodes that are largely unsampled and may contain microcircuits involved in seizure generation. The spatial resolution of research-based electrode arrays has also increased by orders of magnitude. Arrays can now be fabricated that contain hundreds to thousands of densely packed electrode contacts spanning less than 10 square millimeters.[2] However, while electrophysiological recordings intrinsically have high temporal resolution on the scale of milliseconds, these arrays still only sample a small volume of tissue, and limit single-unit recordings to a fraction of nearby neurons (up to a couple hundred, out of hundreds of thousands of neurons), thus still suffering from limited spatial resolution. Conversely, optical methods using voltage and ion-sensitive dyes and high-resolution cameras have been developed to visualize physiological activity without the same spatial limitations, allowing investigators to capture spatial dynamics over broad areas in unprecedented detail. With the advent of optogenetics, neuroscience research has also increasingly involved optical stimulation methods, which allow for targeted modulation of specific, spatially targeted neuronal populations. However, optical techniques have inherent limitations in temporal resolution due to underlying chemical processes that drive the optical signals. Therefore, combining both optical and electrical modalities is attractive to researchers who seek to simultaneously achieve both high spatial and temporal resolution.

The present study by Driscoll et al[3] addresses several of the pitfalls involved in multimodal recording, combining both electrophysiological and optical approaches. As conventional electrodes that use opaque metallic conductive materials block optical signals, they first describe the development and characterization of optically transparent electrodes that use a

modified graphene substrate. Pure graphene is composed of carbon atoms tightly bound in a honeycomb lattice in a single monolayer, which is both highly transparent and electroconductive; however, due to its high sheet resistance, electrodes made from this substance have a prohibitively high impedance. In 2010, Bae et al. reported that doping the sheet lattice with nitric acid (HNO_3) reduced the sheet resistance while preserving optical transparency.[4] In the present study, HNO_3-doped stacked graphene layers were used to produce 50 × 50 μm electrode contacts that achieved reasonably low impedances (~1 MΩ) and high optical transparency (~91–98% transmittance). Although this impedance is higher than comparable platinum electrodes,[5] the electrodes demonstrate acceptable signal-to-noise ratios in electrophysiology recordings, while allowing sufficient optical transparency for simultaneous optical imaging. The results from this study are consistent with prior work using transparent graphene electrodes,[6,7] with equivalent or better electrode impedances and transparency.

The authors present optical and electrical multimodal data from an experiment that was purposefully designed to evaluate the performance of their transparent electrodes. Seizures were provoked in mice expressing a genetically encoded calcium indicator (GCaMP6), using 4-aminopyridine (4AP) as a chemoconvulsant, while electrical recording with surface microelectrode arrays and wide-field calcium epifluorescence optical imaging were simultaneously performed. The fast temporal dynamics of seizure onset and high-frequency oscillations were well captured in the electrical, but not the optical, recordings. At the same time, the spatial evolution of seizure spread was best demonstrated in the optical recordings, such as in identifying a slowly traveling "ictal wavefront" that expands from the initial seizure onset zone. While these methods overcome some of the prior technical pitfalls of acquiring simultaneous electrical and optical recordings, an additional gap in the field is a dearth of analytical approaches for integrating electrophysiological and calcium imaging data with marked differences in temporal and spatial parameters. To address this limitation, they developed a novel multimodal analysis method. First, the microelectrode recordings were converted to features of gamma activity and network synchrony, and downsampled to match the calcium imaging data. The combined data were then reduced using nonnegative matrix factorization (NMF), which can simplify the complexity of data and reveal hidden features. Using this approach, the authors identified features of the observed state changes during ictogenesis in both the spatial and temporal domains. After combining the 2 modalities, NMF analysis revealed 6 states during seizures that were accompanied by evolution in epileptiform discharge characteristics and increases in EEG power and calcium epifluorescence which were spatially maximal near the site of 4AP injection.

While this proof-of-concept study represents a notable technical advance, there are significant limitations. Similar to previous work, their electrodes involve cortical surface recording only, limiting spatial data to 2 dimensions. As the genetically driven calcium indicator targeted excitatory pyramidal neurons, the results from this study are relevant primarily to excitatory circuits, but do not provide direct information on inhibitory networks and interneurons; this could be investigated in future studies targeting interneurons. Furthermore, this study only utilized an acute seizure model induced by a chemoconvulsant, which may not be representative of seizure dynamics that occur in chronic spontaneous epilepsy. It will be important to apply these novel methods to a chronic epilepsy model in the future.

In terms of clinical translation and other future directions, the findings in this study demonstrate the potential of transparent electrodes to provide novel insights on neurophysiology with unprecedented spatiotemporal detail. While the current study primarily involves recording of network activity, similar devices have been shown to be compatible with optical stimulation to activate or inhibit specific brain regions.[6] Future work to reduce the impedance of transparent electrodes could further decrease contact size, permit recording single-unit activity, and make electrical stimulation feasible. Using this technology to improve the descriptions of seizure dynamics may provide novel insights on the localization of ictogenesis and optimize the targeting of focal surgical resection or electrical stimulation methods for the clinical treatment of epilepsy. Improving the definition of seizure states may be of particular relevance to closed-loop electrical neurostimulation treatments[8] such as responsive neurostimulation (RNS), as certain states may be more susceptible to disruption than others. Though calcium imaging is unlikely to translate to human studies in the foreseeable future, optical intrinsic signal imaging of brain activity has been used for decades to identify functional areas of human cortex[9] and may be compatible with transparent electrodes. Simultaneous electrical recording and optical imaging will not be translatable to patients until significant technical advances in these technologies overcome barriers to their clinical use in people. Finally, to characterize electrophysiology in high temporal and 3-D spatial resolution, significant advances are needed to develop transparent electrodes capable of depth recordings of structures and networks beneath the cortical surface. Overall, despite the current limitations, there is an impressive variety of potential basic and clinical applications of multimodal recording for understanding and treating epilepsy.

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