Optimizing the Fabrication of Carbon-Fiber Microbiosensors for Simultaneous Detection of Glucose and Dopamine in Live Brain Tissue

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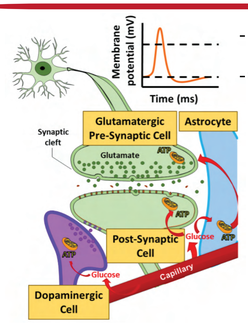
Abstract

Neuronal communication is an energetically demanding process. There is substantial evidence indicating a dysregulation in both brain metabolism and dopamine dynamics in several neurological disorders, including Alzheimer's disease and addiction. However, the precise relationship between glucose and dopamine remains unclear due to a critical lack of analytical tools and techniques capable of studying energetic substrates and dopamine dynamics simultaneously. Carbon-fiber microelectrodes are commonly coupled with fast-scan cyclic voltammetry (FSCV) for the subsecond detection of electroactive neurotransmitters, such as dopamine, in situ. These electrodes can be modified with xidase enzymes to create microbiosensors capable of simultaneously quantifying real-time fluctuations of non-electrochemically active substrates, such as glucose or lactate. Hydrogel entrapment of the enzyme within a chitosan matrix on the carbon-fiber surface provides for stable, sensitive, and selective detection of dopamine and the enzyme substrate using FSCV. **The purpose of this study is to characterize the chitosan hydrogel that entraps oxidase enzyme on the electrode surface, and the effects of the hydrogel's physical nature on acquired electrochemical data.** Hydrogel was deposited using linear sweep voltammetry, and membrane consistency and electrochemical performance were characterized to optimize the deposition potential range and sweep rate. Finally, voltammetric measurements were used to determine how these factors relate to sensor performance. **Overall, these experiments are important because they provide an improved understanding of the hydrogel matrix that is integral to microbiosensor function, thus advancing this much-needed technology for monitoring real-time neurochemical kinetics.**

Background



- Dopamine is a major signaling molecule in rat striatum
- Glucose is thought to be the primary energetic substrate that fuels dopaminergic signaling

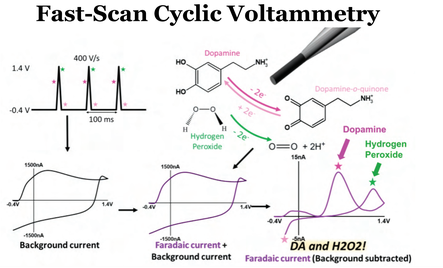
Little is known about the dynamics by which glucose is supplied to meet the metabolic demand associated with neuronal activation

- *How does glucose scale with neuronal activation?*
- *How do the kinetics of glucose availabilty pair with those of dopamine release?*

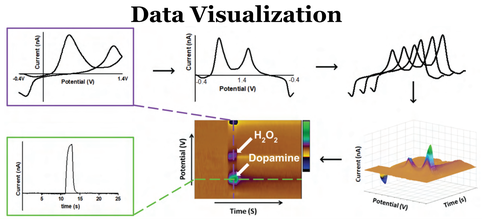
An *in situ* characterization of dynamic changes in the availability of neuroenergetic substrates is needed.

Electrochemical Detection of Dopamine and Hydrogen Peroxide

Fast-Scan Cyclic Voltammetry



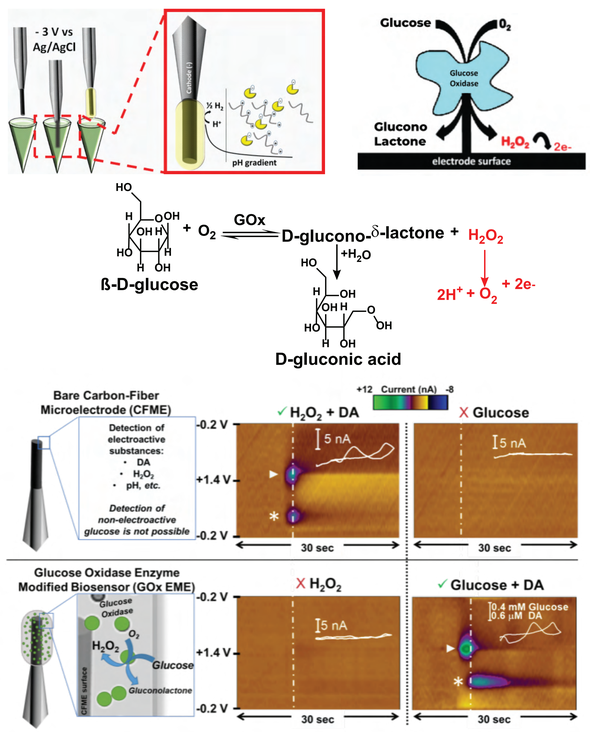
Data Visualization



Enzymatic Microbiosensor Fabrication

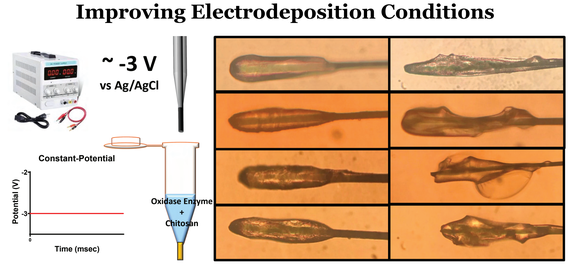
Carbon-fiber microelectrodes were modified with **glucose oxidase (GOx)** to allow for the detection of the non-electroactive molecule of glucose while preserving sensitivity to dopamine.

Hydrogel Entrapment of the Oxidase Enzyme in a Chitosan Matrix



Commercialization of the Technology

Improving Electrodeposition Conditions



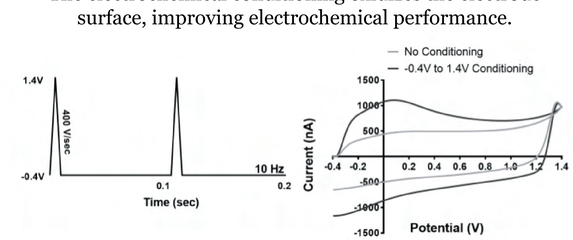
What is Contributing to the Variability?

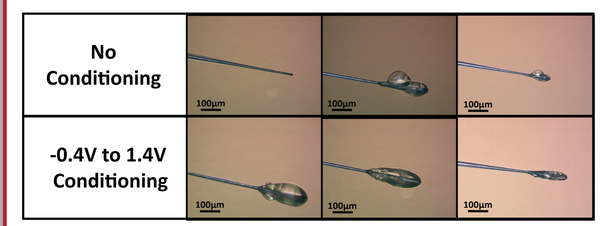
- *Hydrogen evolution at the electrode surface?*
- *The carbon surface has inherent variability?*

Electrochemical Conditioning of the Electrode Improves Membrane Consistency

Conditioning the electrode prior to electrodeposition is critical to ensuring biosensor function

The electrochemical conditioning oxidizes the electrode surface, improving electrochemical performance.

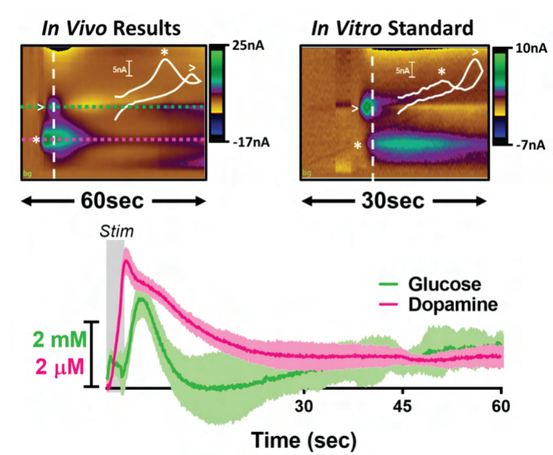




Real-Time Measurements in Rat Striatum

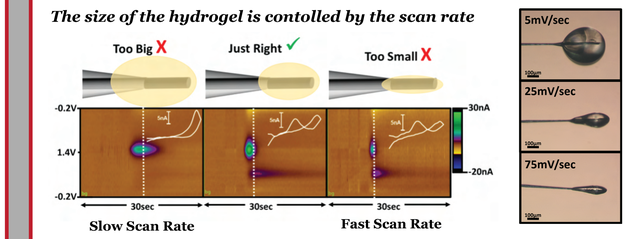
We are collaborating with Pinnacle Technologies Inc. to fabricate these microbiosensors at a larger scale so that the technology can be transformed from a custom laboratory tool to something accessible the the whole research community.

Pinnacle Produced Electrodes Generate Quality In Vivo Data

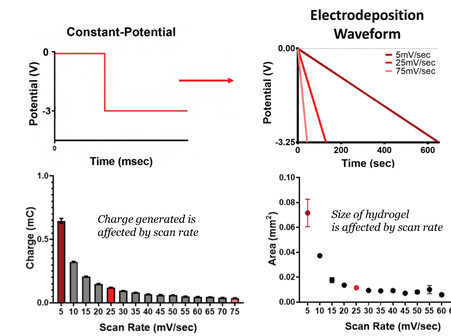


Gradual Application of Electrodeposition Potential Improves Membrane Consistency

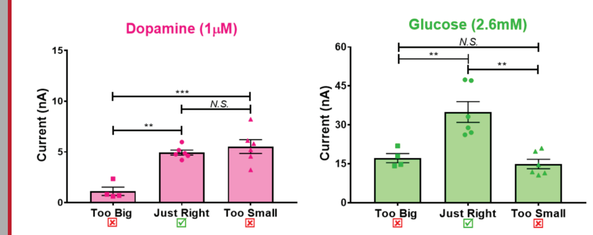
The size of the hydrogel is contolled by the scan rate



A controlled linear sweep improves the quality of the membrane by limiting hydrogen evolution



Sensor performance is dependent upon the size of the hydrogel



Conclusion

In order to understand the impacts of diseases on brain metabolism and to inform potential therapeutic strategies, it is important to have better tools for understanding how these substrates fluctuate relative to one another in real-time.

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