**Effects of Cholesterol Enhancement on the Cell Properties in the Presence of Graphene**

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### Introduction

Motivation: Graphene is a two-dimensional carbon crystal with remarkable mechanical strength and a singular electrical conductivity, a combination that has led to interest in biomedical application. However, this demands a clear understanding of graphene’s interaction with cell surface molecules and its impact on cell function.

**Objectives:**
- To analyze the cholesterol concentration of culture media in the presence of graphene using spectral analysis
- To modify cell properties using graphene and demonstrate a relationship between graphene concentration and membrane resistance
- To highlight the importance of proper procedure when producing cell cultures to prevent cortex contamination

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**Previous Research**

Previous research concluded:
- Graphene does not cause significant changes in neuronal morphology
- An electrophysiological change in frequency, but not amplitude of mEPSCcs suggests that most likely the impact is presynaptic
- Synaptic vesicle tracking combined with cholesterol imaging implicates that graphene increases both membrane cholesterol content and membrane curvature
- As such changes are limited and defined, bare graphene may be used as both a growth substrate and a field effect transistor to detect membrane potential changes

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**Graphene May Increase Membrane Resistance**

- figure showing membrane resistance changes with and without graphene

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**Testing the Graphene and Cholesterol Interaction**

- figure showing spectral fluorometry measurements

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**Impact of Cortex on Hippocampal Networks**

**Figure 1:** A hippocampal neuron sample with minimal cortex contamination. Images were produced following immunocytochemistry and applying a specific wavelength of light to excite fluorophores. B) Hippocampal neuron sample with cortex contamination. Note debris in the soma image. C) Shell analysis intersection masks. The process works by creating concentric circles and counting the number of intersections as a function of radius.

- figure showing shell mask analysis

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**Results and Future Work**

- Cortex has a slight negative impact on hippocampal networks. The debris increases the uncertainty of analysis done via immunocytochemistry as the antibodies will also stain the cortex.
- Preliminary data shows implicates that graphene may increase membrane resistance, although additional verification is necessary
- Further spectral analysis of fluorophore interaction with graphene reveals a time-dependent fluorescence decay that is unique from that of the fluorophore, consistent with an interaction involving energy transfer

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**Future Work** will include additional patch clamping experiments on different cell types to further study the relationship between chronic or acute graphene exposure and membrane resistance

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**References:**