Optical Detected Magnetic Resonance and fluorescent Nanodiamonds: towards molecular resolution neuronal imaging

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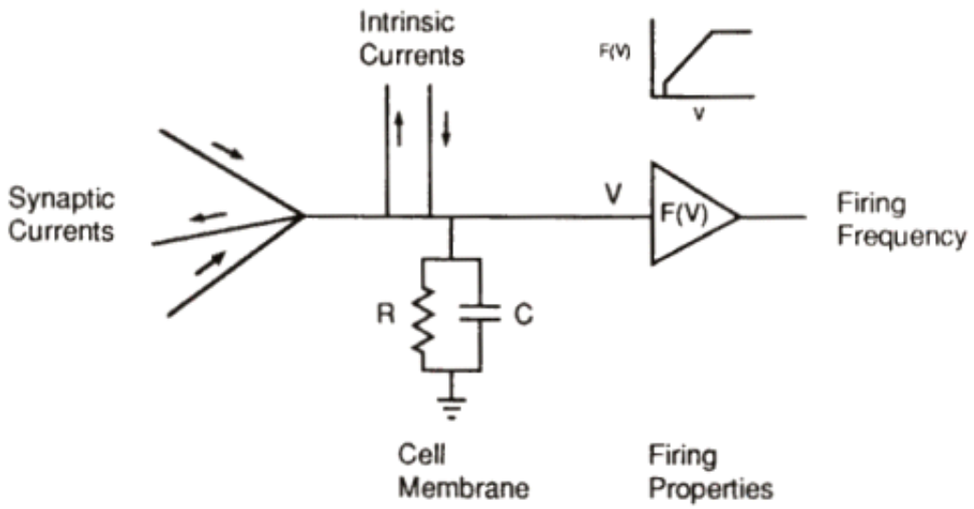
Outline

- Neurons as RC circuits
- Background and motivations
- Why NV-NDs?
- Bare VS functionalized NV-NDs
- Experimental
- Future plans

Neurons: a tale of circuits and Action Potentials



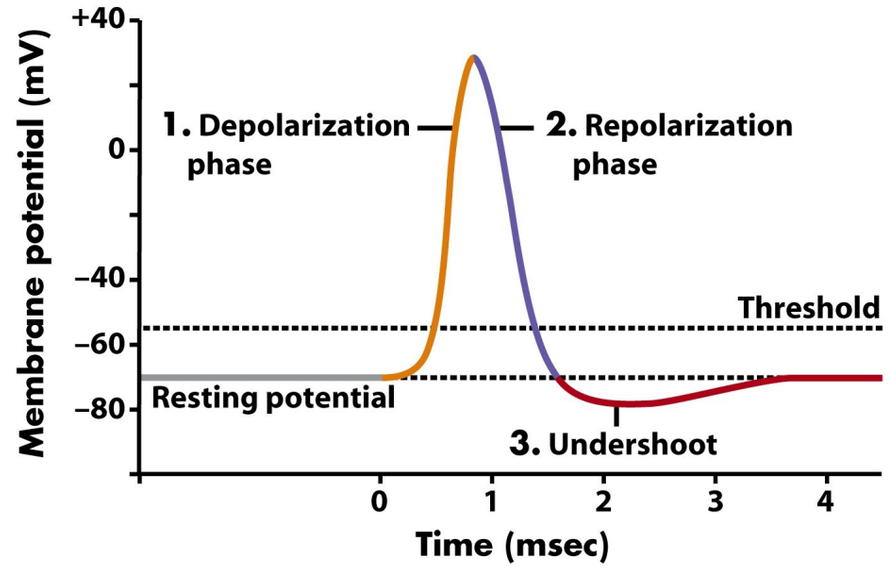
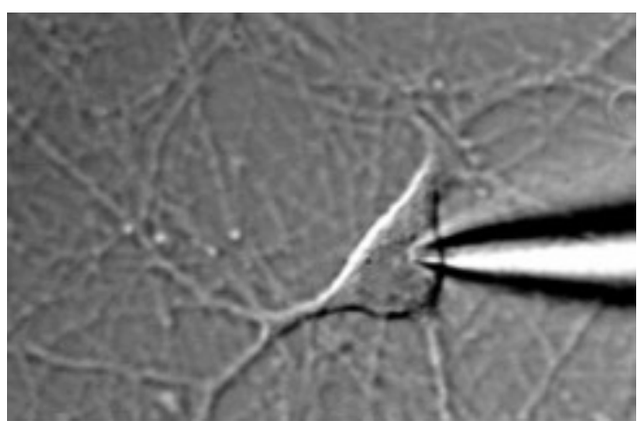
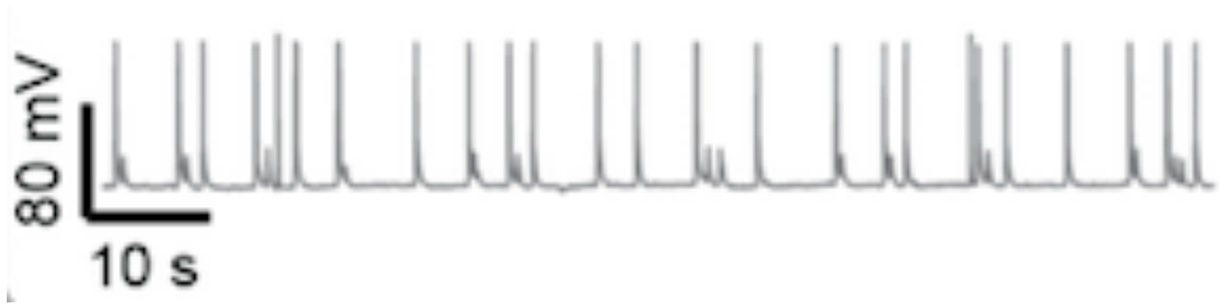


Figure 45-5 Biological Science, 2/e





Background and motivations

Nanoscale tools for monitoring cellular electrical activity

Drawbacks of Voltage/ Calcium-Sensitive Dyes

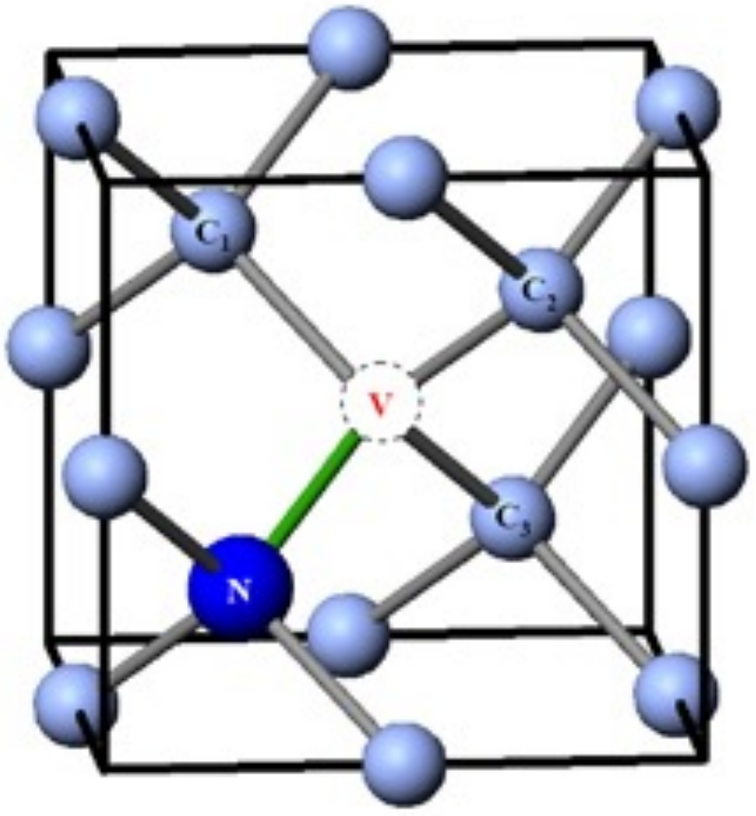
Intrinsic limitations of existing techniques (Electrophysiology, MEG, EEG)

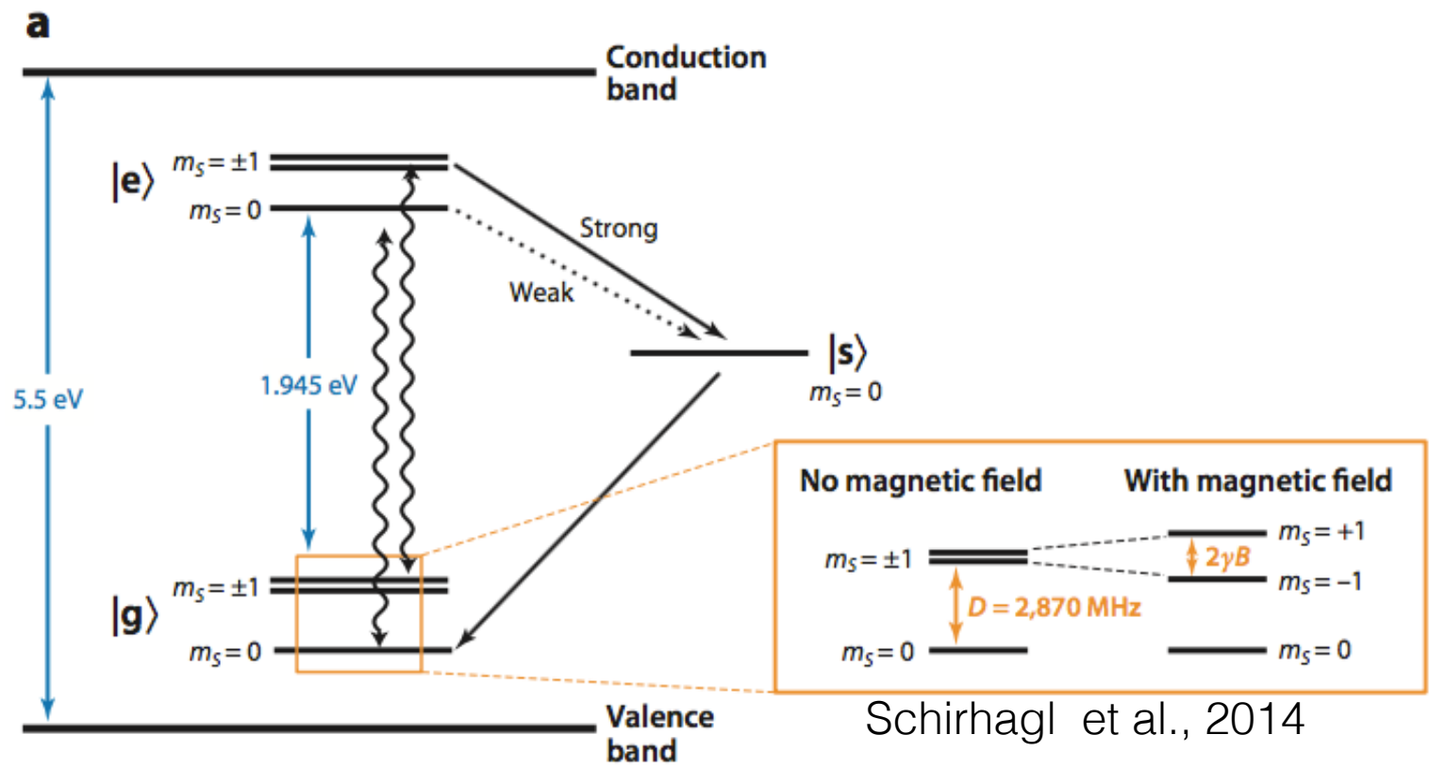
Non-invasive access to subtreshold synaptic events

Direct evaluation of local neural electromagnetic field

Temporal resolution combined with localization of sources of neuronal activity

Why Nitrogen Vacancy-NanoDiamonds?





Schirhagl et al., 2014

Voltage Sensitive Dyes versus NV-NDs:

- **only** electrical potential changes versus electric **and** magnetic fields measurements

High-resolution measurements of membrane potentials

Characterization and functionalization of NV-NDs

Electronic, chemical and morphological properties:

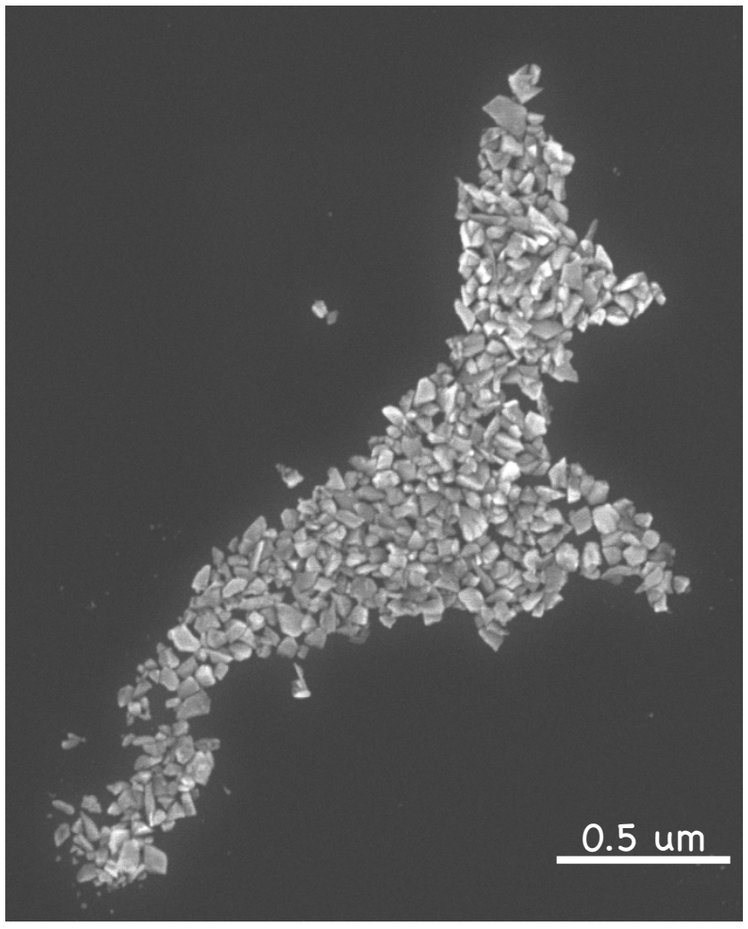
- SEM, TEM, DLS, Z-Potential, XPS, Raman, FTIR

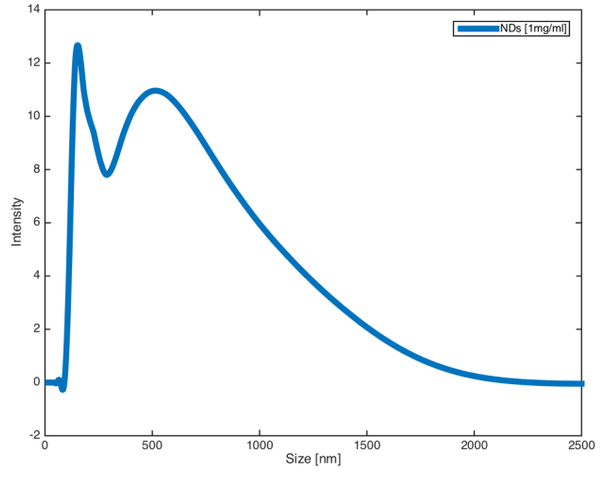
Avoiding clustering:

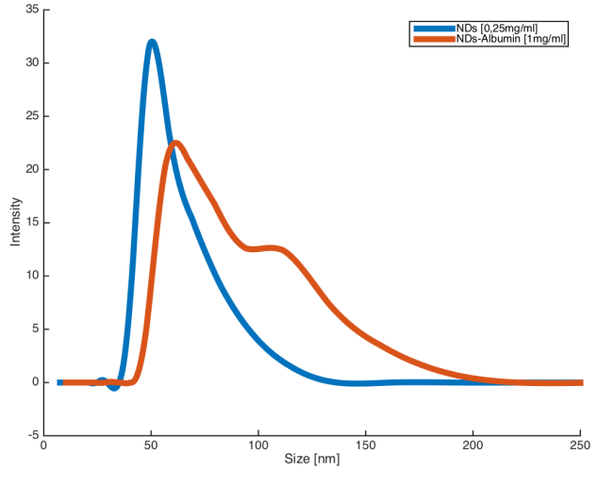
- albumin
- lipid micelle embedding

Promoting internalization:

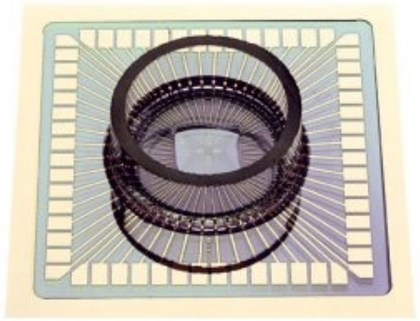
- cell-penetrating peptides

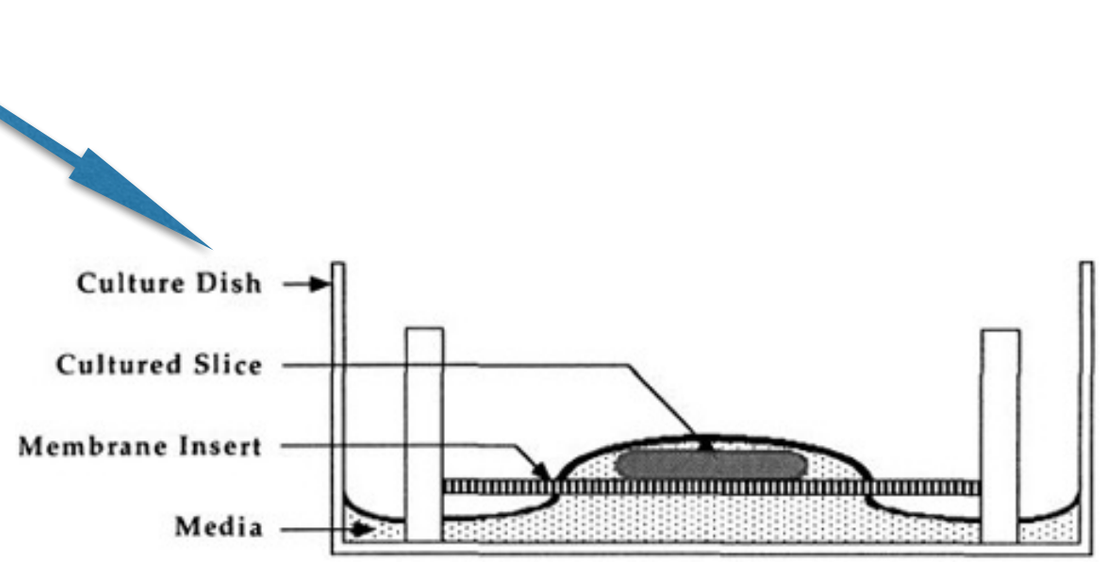


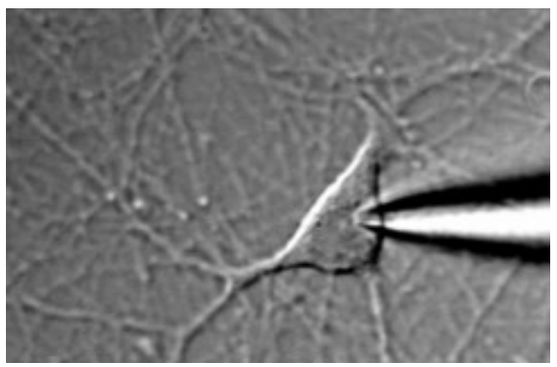




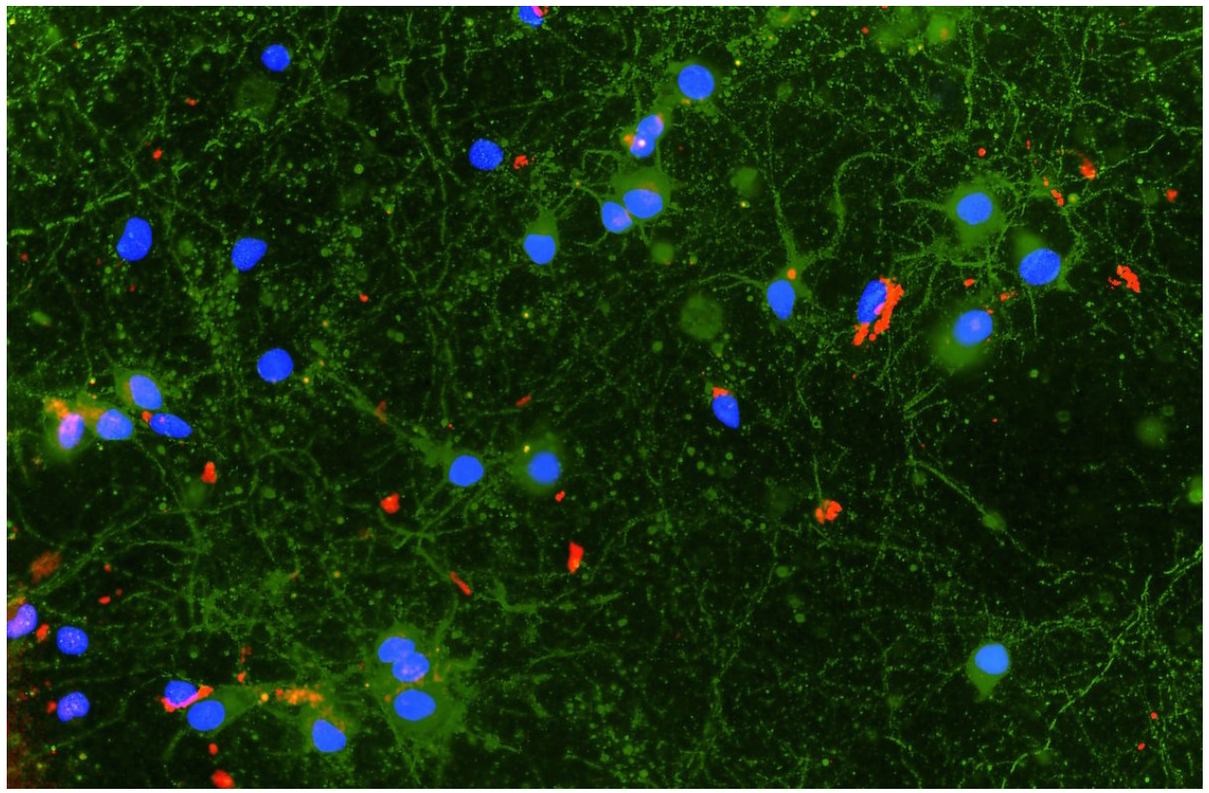
Interfacing NV-NDs with primary neuronal cultures/organotypic brain slices

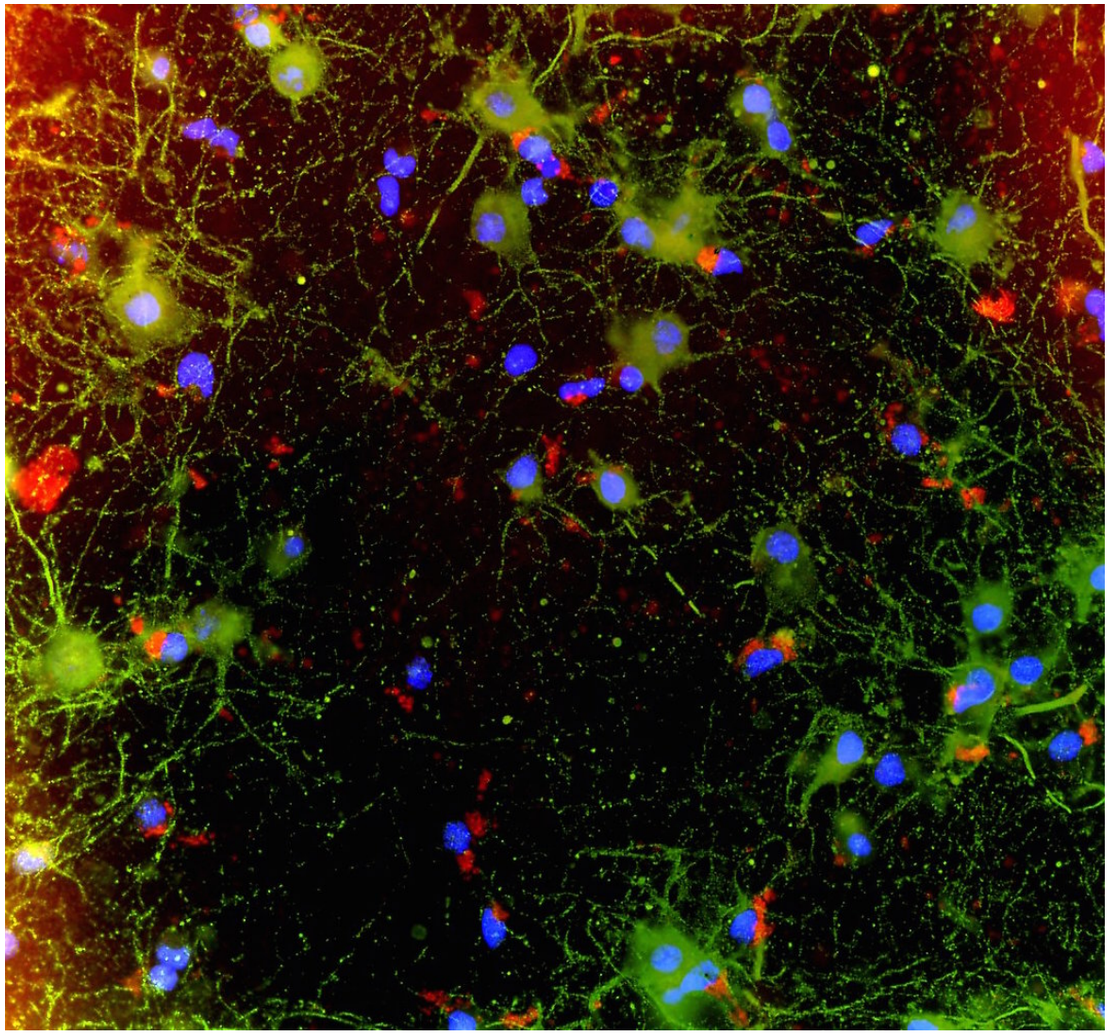






Immunostaining

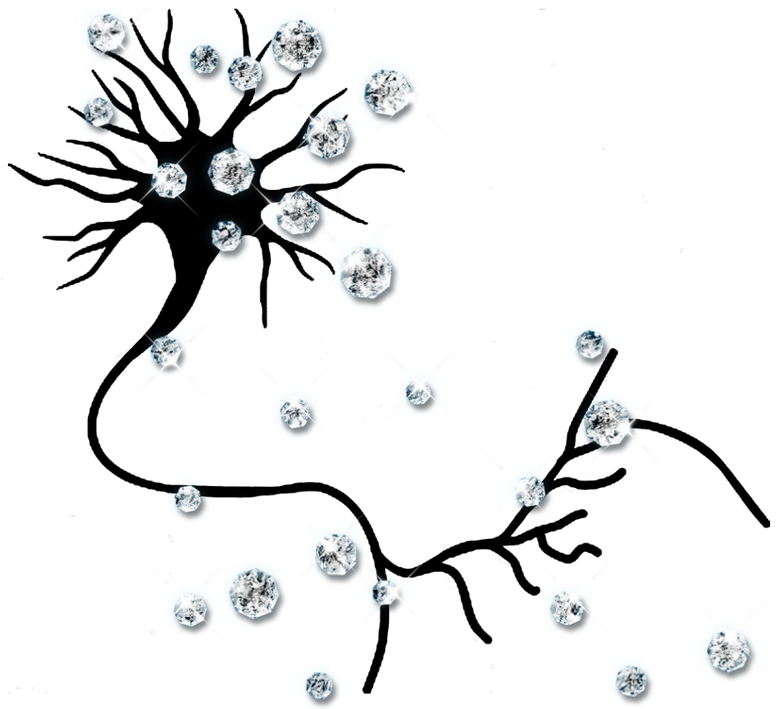


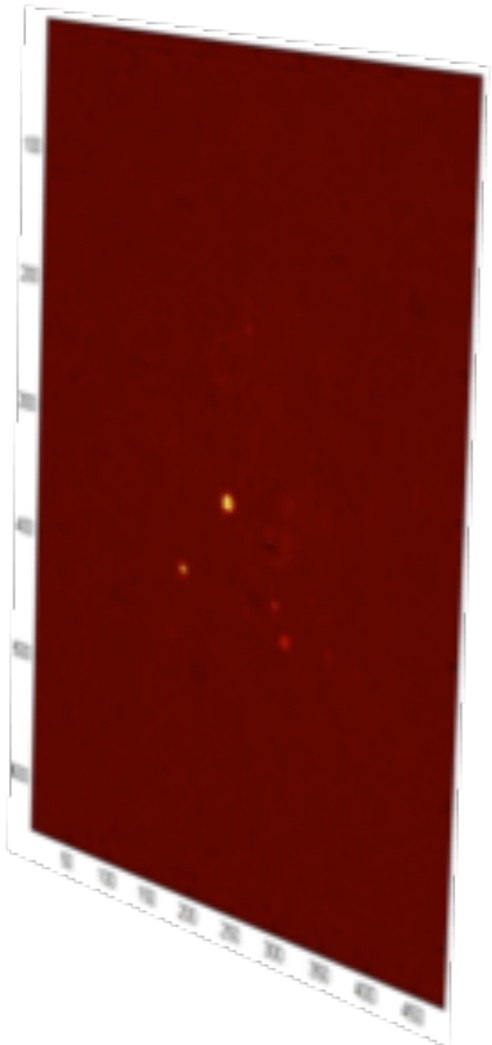


Neurons NV-NDs

Nuclei

Observing differences in NV-NDs photoluminescence in living neurons

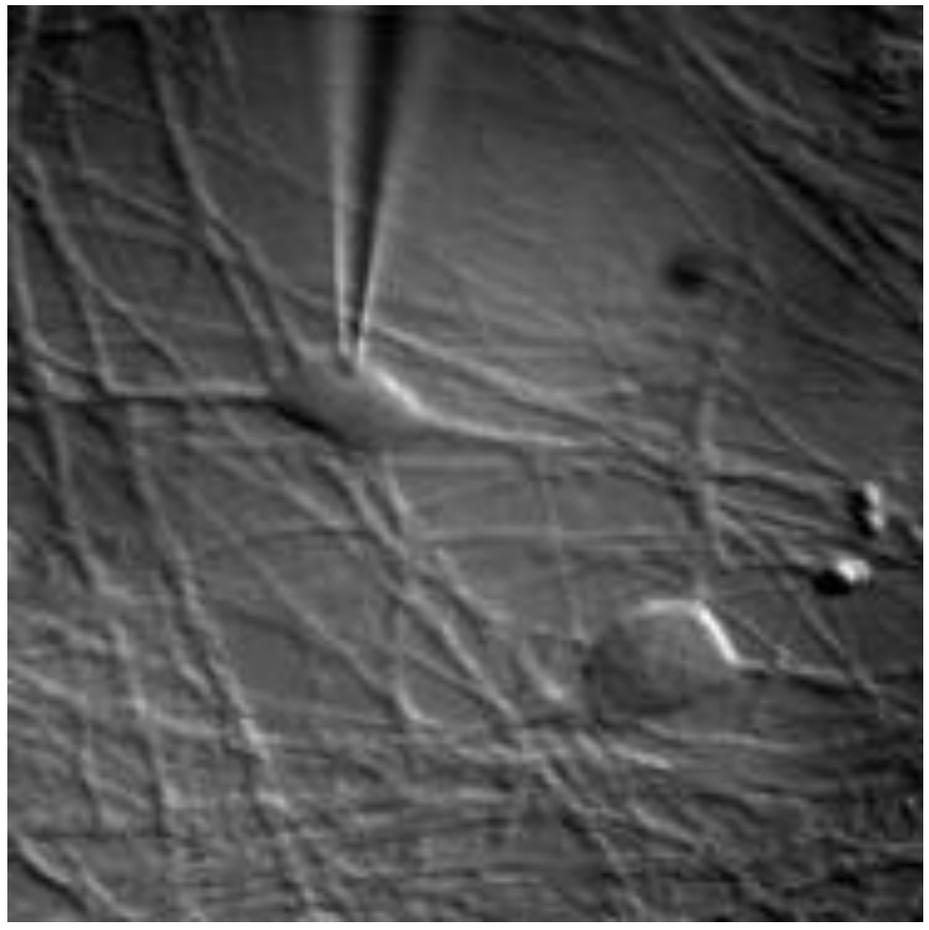




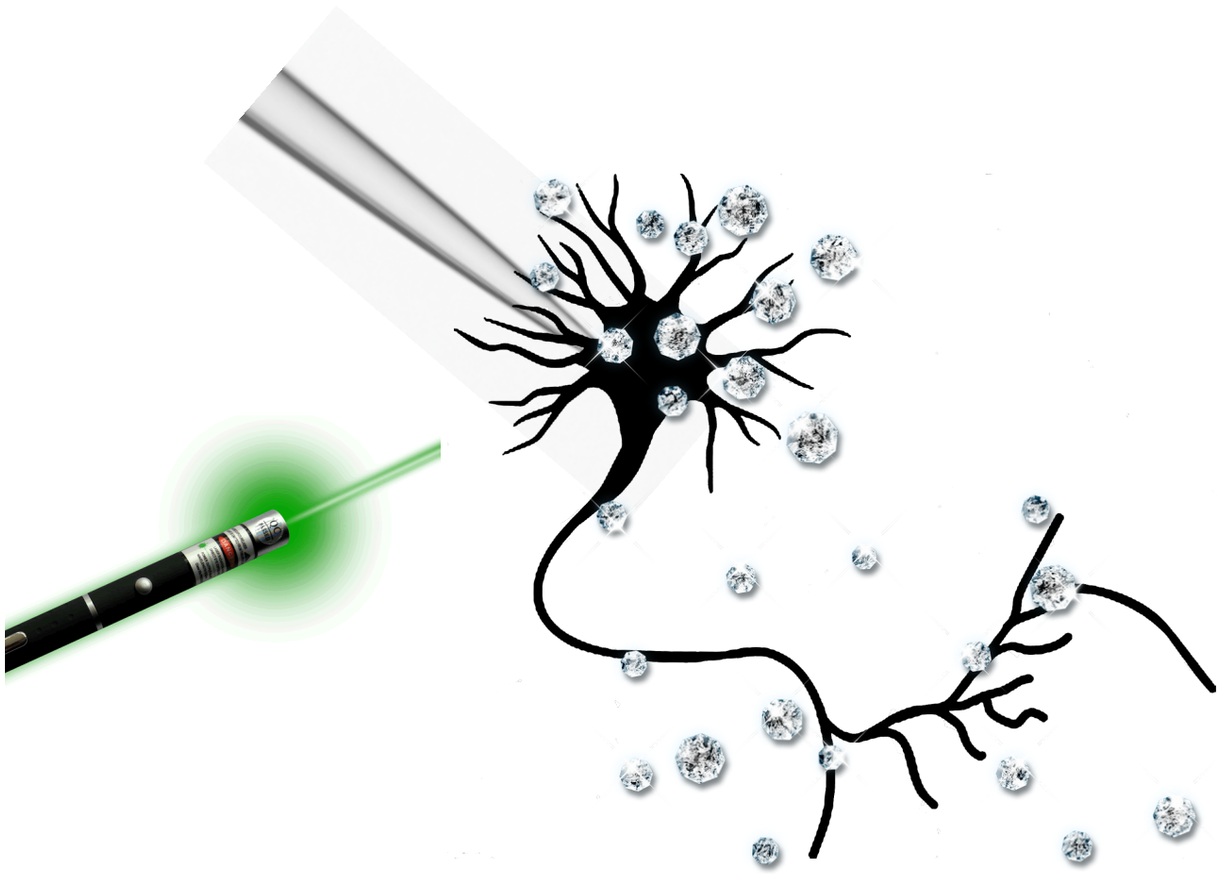
Work in progress

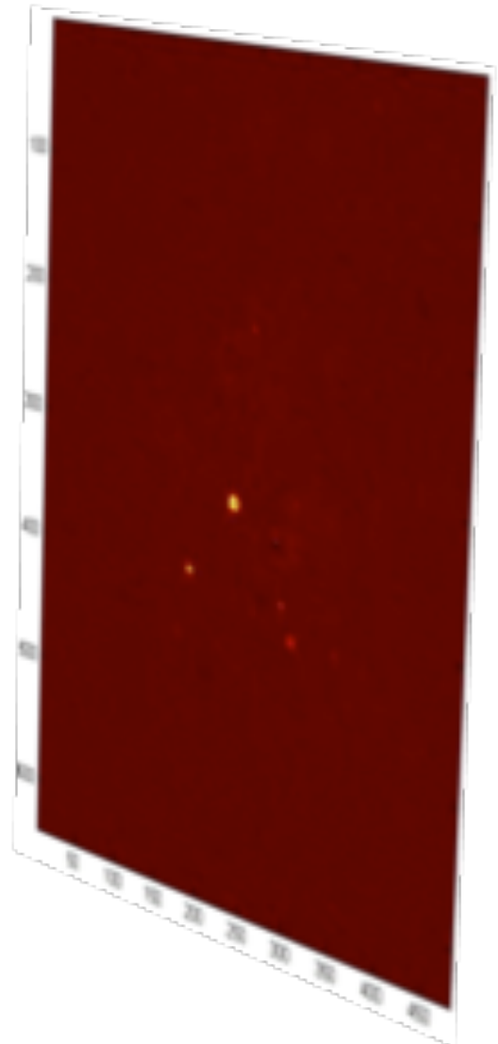
NV-NDs localization and Patch Clamp recordings

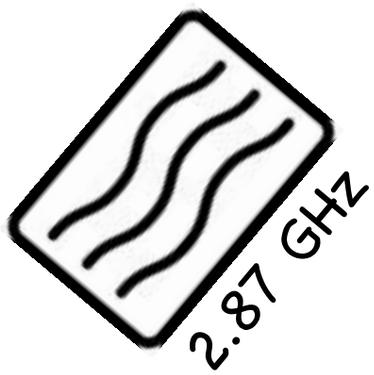
- Internalization:
  - Fluorescence Microscopy, TEM
  - Synchrotron Light: NEXAFS & Cryo-Electron Tomography
- Effects on neuronal electrical activity and morphology



Correlating difference in NV-NDs photoluminescence with neuronal spontaneous activity







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