Microfluidic Patch Clamp Technique for Measuring Membrane Disruption

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Overview

1. Background
2. Motivation
3. Data
4. Work to be done
An Impressively Brief Background

• Luigi Galvani
  – Realization that electrical phenomena are part of animal physiology
  ➔ Electrophysiology
• 1940’s Voltage Clamp measures the ion currents across the membrane of excitable cells
  - Kenneth Stewart Cole
    “pioneer in the application of physical science to biology”
  - George Marmount

• 1970-1980 – Patch Clamp
  - Erwin Neher
  - Bert Sakmann
What is a patch clamp?

• “An extremely fine glass pipette with a very small opening [that] is used to make contact with a tiny area, or patch, of the cell's outer membrane.” A small amount of suction is applied through the pipette and forms a tight seal.

• Fit the pipette with a highly sensitive electrode to record very minute changes in current produced as ions flow through the clamped channel into the pipette.
Nobel Prize!

• 1991 Nobel Prize in Physiology and Medicine
  – “conclusively proving the existence and function of ion channels.”
Hypothesis: Cationic nanoparticles induce the formation and/or growth of nanoscale defects in cell membranes, such as holes, causing cell membrane leakage.

Chen et al. *Journal of Physical Chemistry* 2009
Goal: Measure changes in electrical properties of membrane to infer damage
Importance

• Applications in medicine
  – Drug and gene delivery
  – Viruses
  – Prions
    • Untreatable and fatal!

• Cytotoxicity
  – Nanoparticles
IonFlux

- Traditional Patch Clamp
  - Slow and labor intensive
- Microfluidic Patch Clamp
  - Incredibly quick
  - “microfluidic design to provide automated cell introduction”
  - channel network that connects the wells with a series of tiny channels for cell and compound introduction.
  - pneumatic flow control
What the Literature Says

• There haven’t been any measurements of individual nanoscale hole forming events induced by cationic nanoparticles on living cell membranes

• There hasn’t been characterization of the time course of the events

Chen et al. Journal of Physical Chemistry 2009
Paper Results

• 1. “Exposure of cells to noncytotoxic levels of cationic nanoparticles results in the formation of defects that enhance conductance through the cellular membrane”
• 2. “These defects can recover over time allowing a decrease in transmembrane conductance toward it’s original value.”
Hong, et. Al. *Bioconjugate Chemistry* 2006
Cells...
Setup
Images
Future Work

• Quantify current fluctuations
• How to treat data
• PAMAM Dendrimers
  – Nanoscale drug delivery
Works Cited

• http://www.bem.fi/book/04/04.htm
• Wikipedia
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Questions?