SickKids®

UNIVERSITY OF TORONTO

Using Stem Cells to Predict Drug Effect

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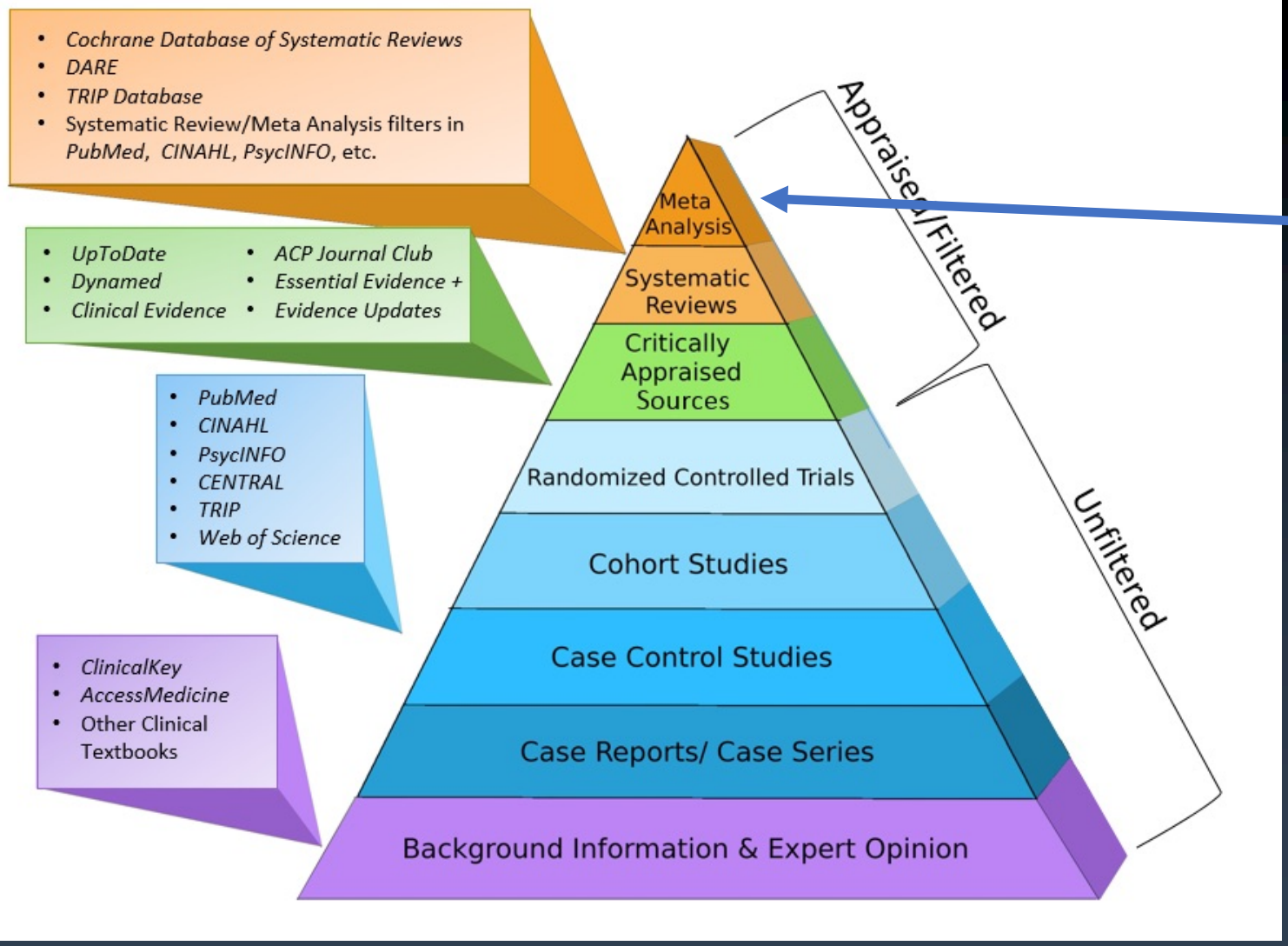
Disclosures:

- Current and past funding/drug discovery agreements with:
  - Merck
  - GSK
  - Novartis
  - MaRS Innovation
  - Blueline Bioscience
  - Versant Ventures
  - Acea Biopharma
  - Ontario Center of Excellence
  - IRiCOR

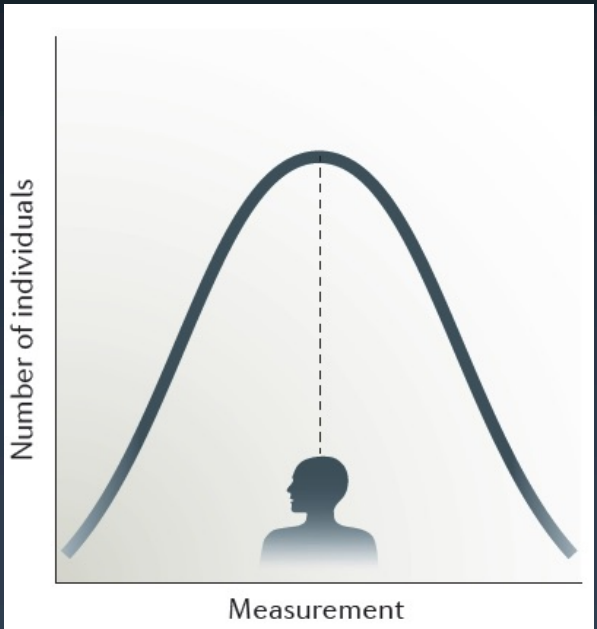
Intelligent Clinical Decision Making

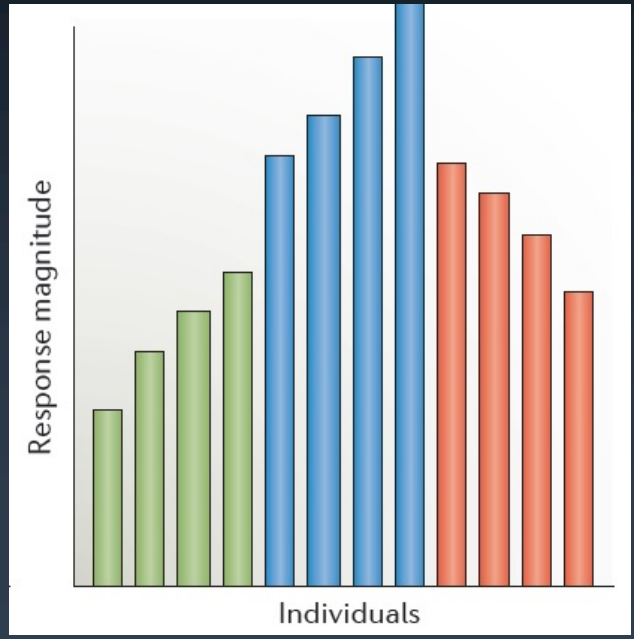
- An oncology patient needs chemotherapy:
  - What regimens risk cardiac disease?
- A post-surgical patient needs pain treatment:
  - Are they at a high risk for opiate side effects?
  - Are they likely to develop chronic pain?
- An ICU patient has poor cardiac function:
  - Will this patient respond to milrinone, and at what dose?



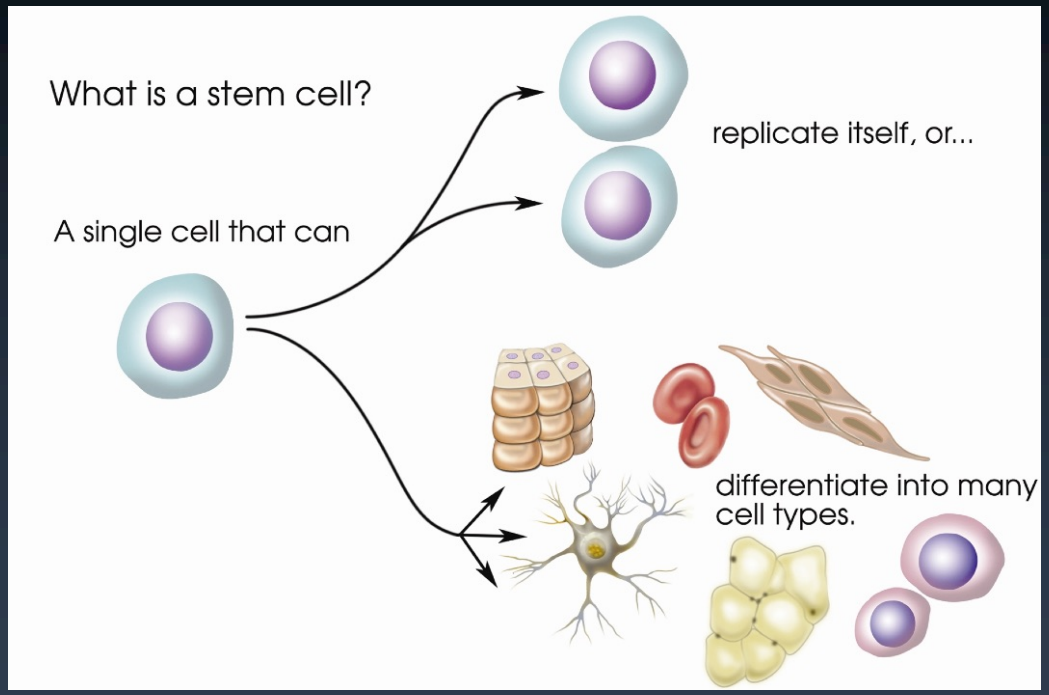
Is this your patient?

Patient Phenotyping



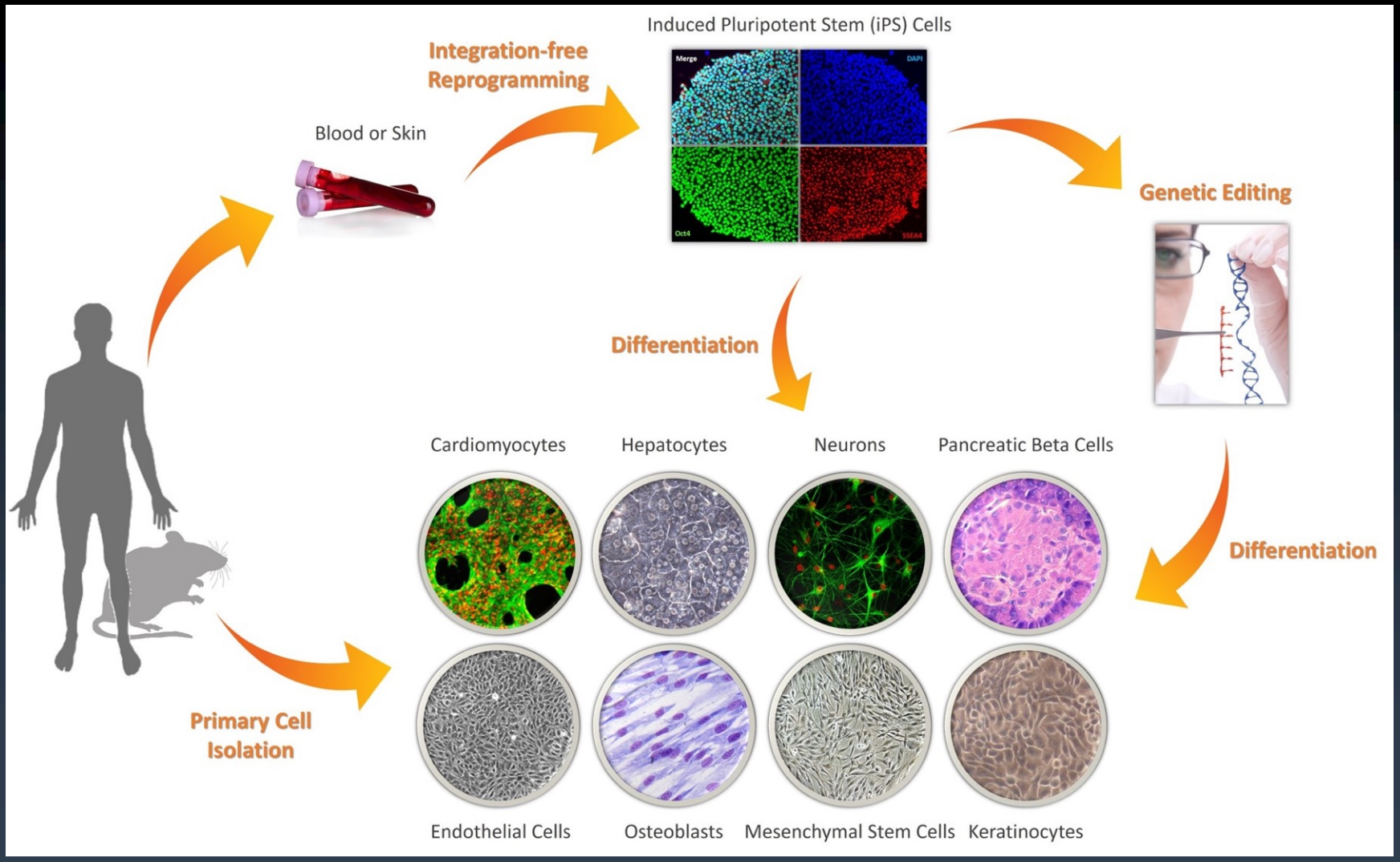


Stem Cells and Stem Cell Tissue



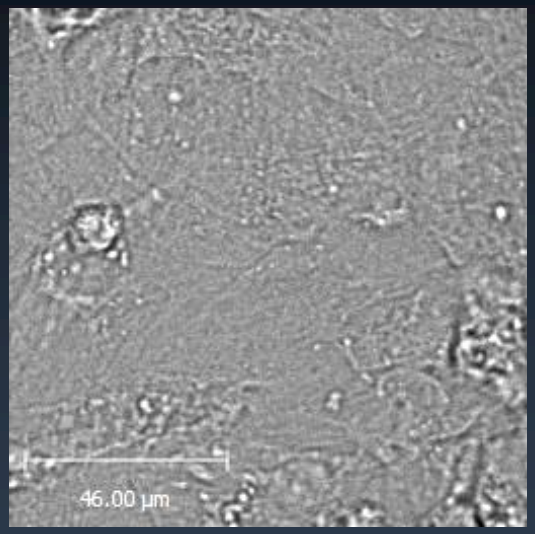
Existing Stem Cells:
Hematopoietic
Mesenchymal
Neuronal
Cardiac

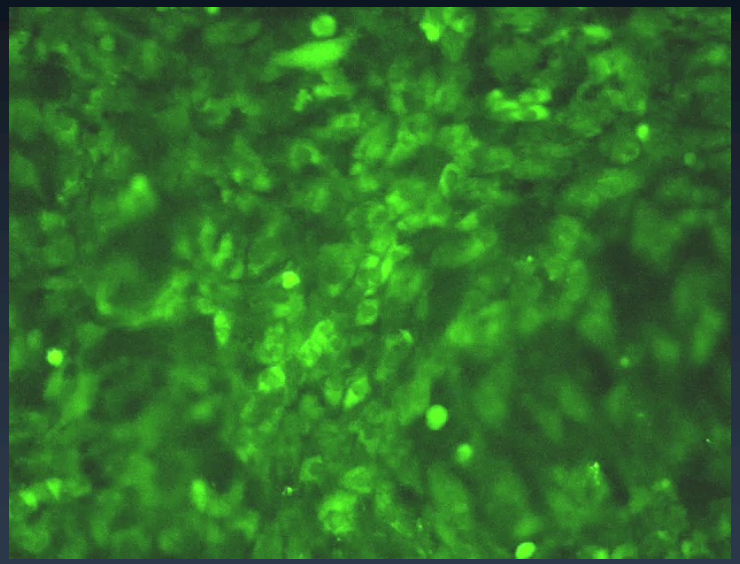
Umbilical
eSC
iPSC


Induced Pluripotent Stem (iPS) Cells
Integration-free Reprogramming
Blood or Skin
Genetic Editing
Differentiation
Cardiomyocytes
Hepatocytes
Neurons
Pancreatic Beta Cells
Differentiation
Primary Cell Isolation
Endothelial Cells
Osteoblasts
Mesenchymal Stem Cells
Keratinocytes

Utility of Stem Cell-Derived Tissue

Disease and Tissue Modeling: Cardiac



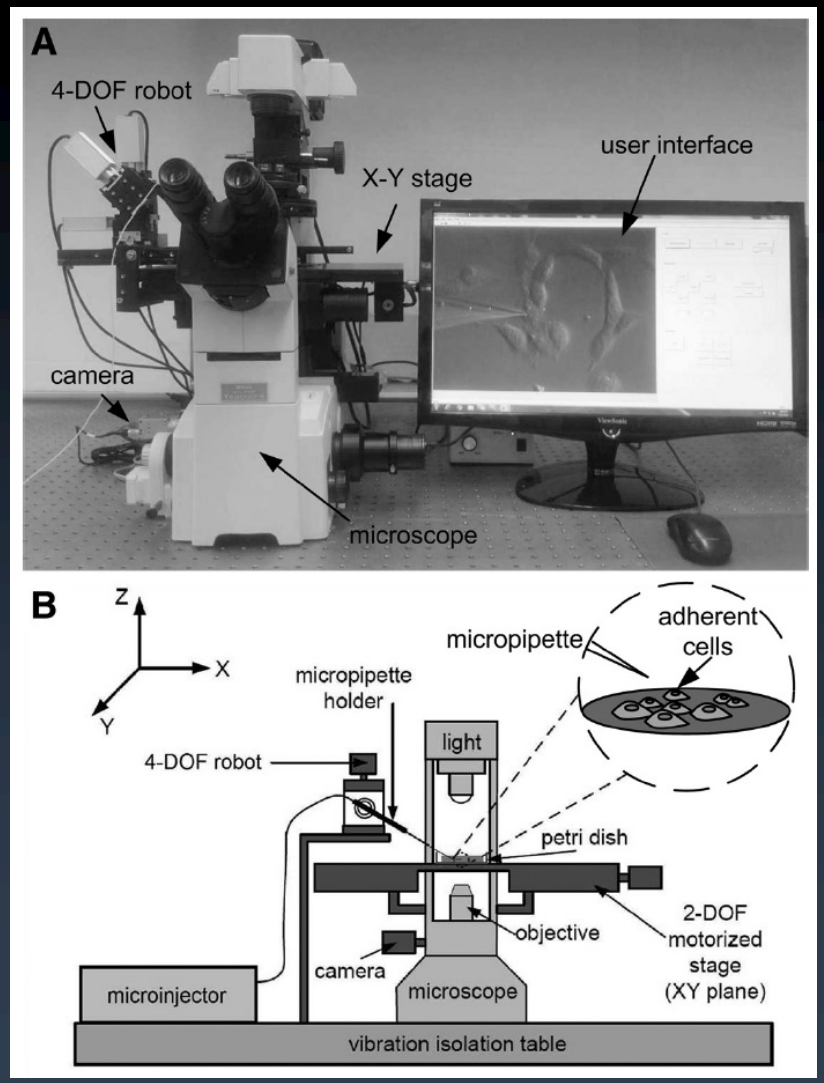


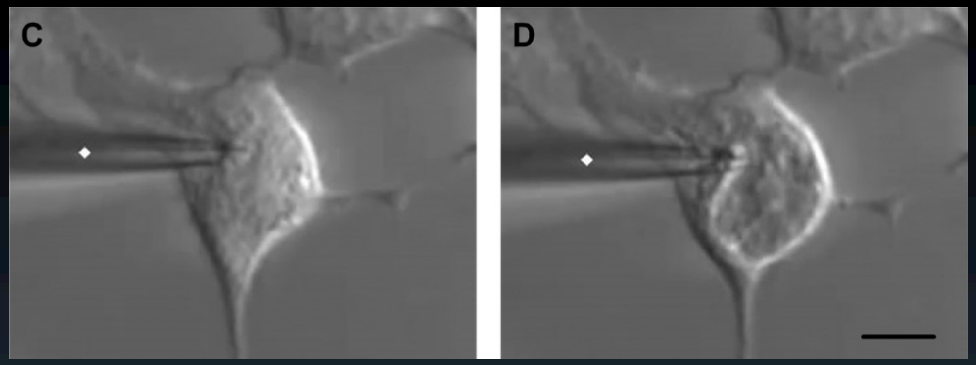
Complex Dynamic Tissue: rate, rhythm, contractility, conduction

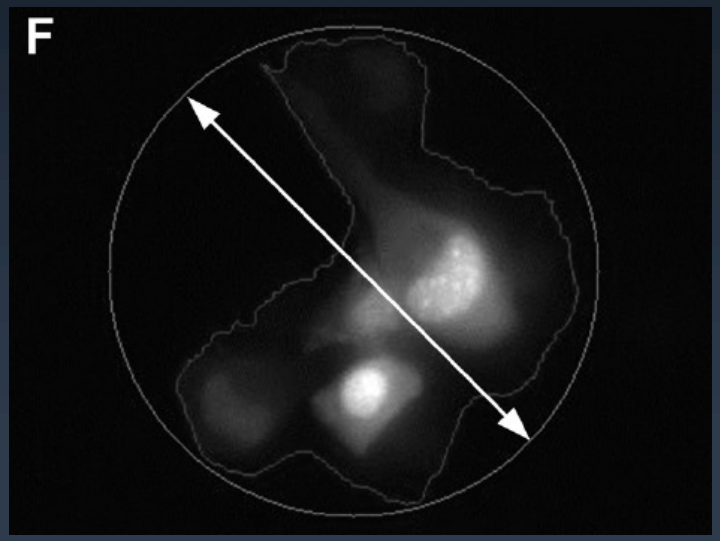
Quantifying Cardiac Conduction

- > 70% of drug recalls are for unexpected cardiotoxicity
  - Comprehensive in Vitro Proarrhythmia Assay (CiPA)
- A majority are for arrhythmogenic side effects
- Cardiac conduction diseases - ARVC
- Anesthesia pharmacopeia has high cardiac interactions

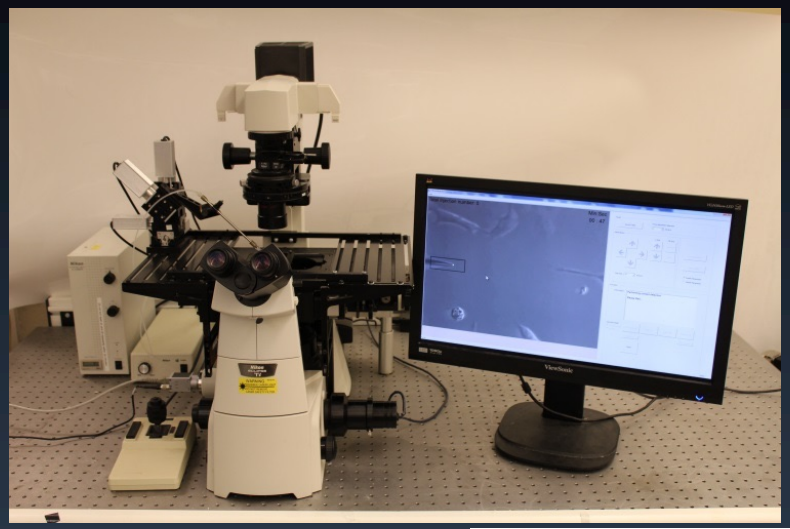
- To measure cardiac conduction, you need functional tissue (not isolated cells, patch clamping)

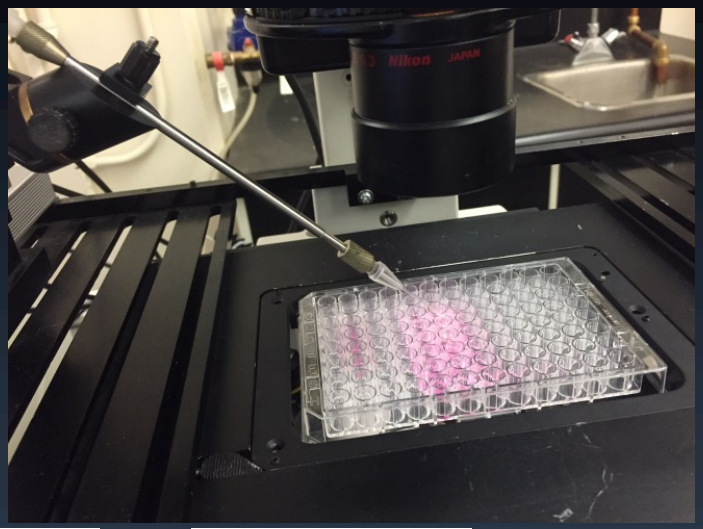

A
4-DOF robot
user interface
X-Y stage
camera
microscope
B
Z
X
Y
micropipette
adherent cells
micropipette holder
light
4-DOF robot
petri dish
objective
2-DOF motorized stage (XY plane)
camera
microinjector
microscope
vibration isolation table

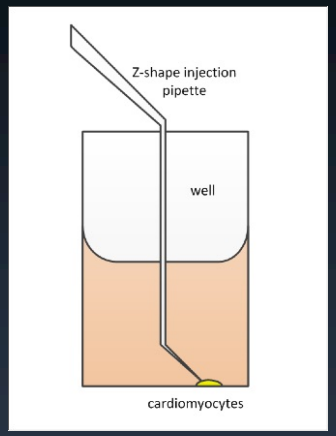

C
D

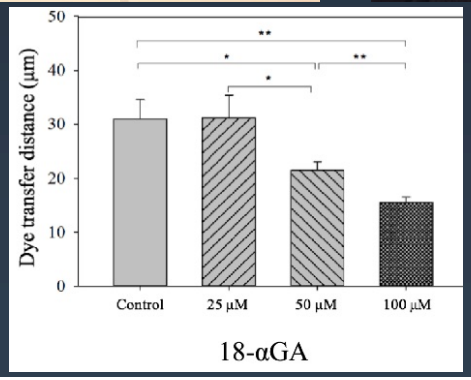

F

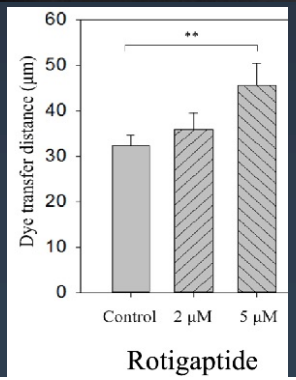
Quantifying Cardiac Conduction





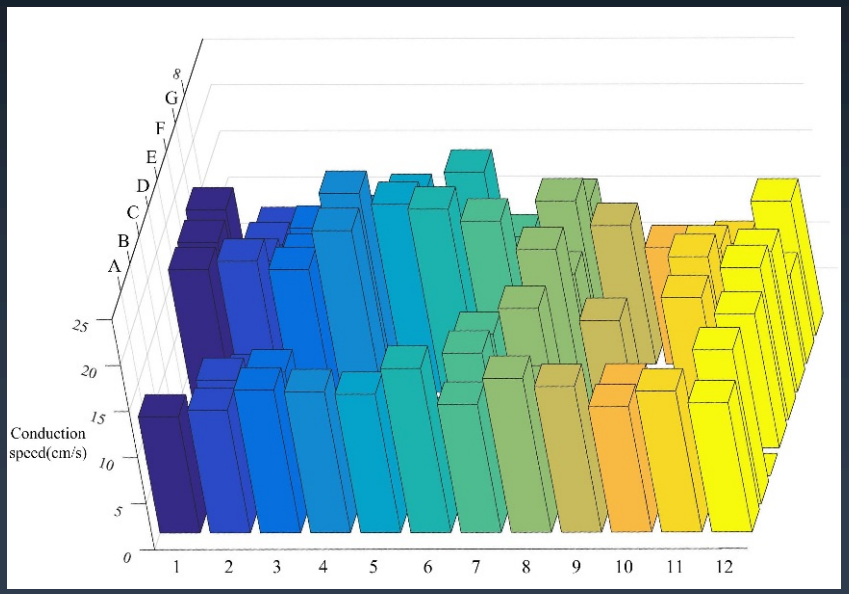


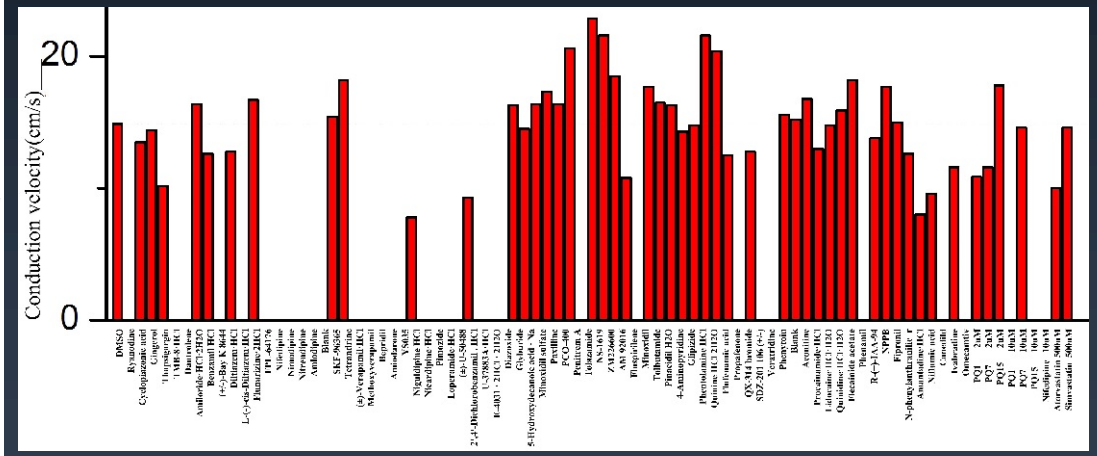




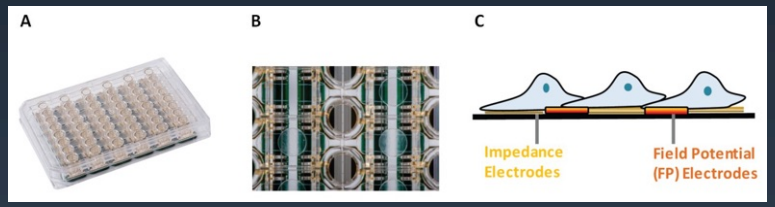
Quantifying Cardiac Conduction

- Mutations in Plakophilin 2 (PKP2) cause ARVC
- Screened 1000 compounds known to affect ion channels

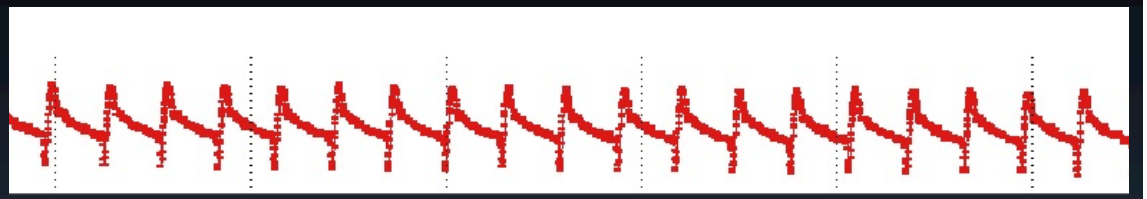




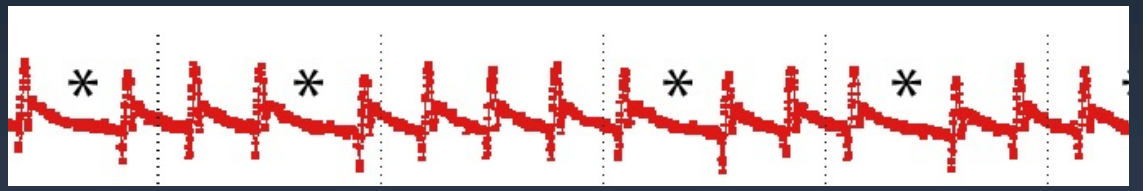
Comprehensive Disease Modeling

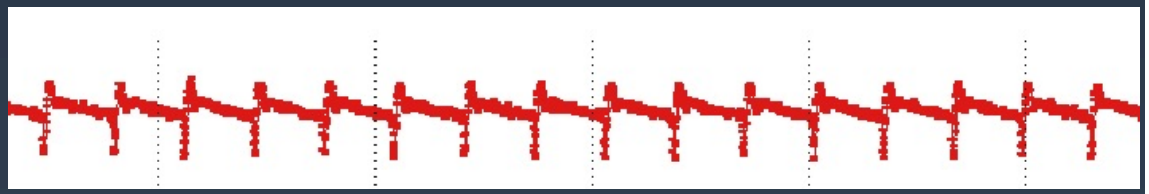


Baseline

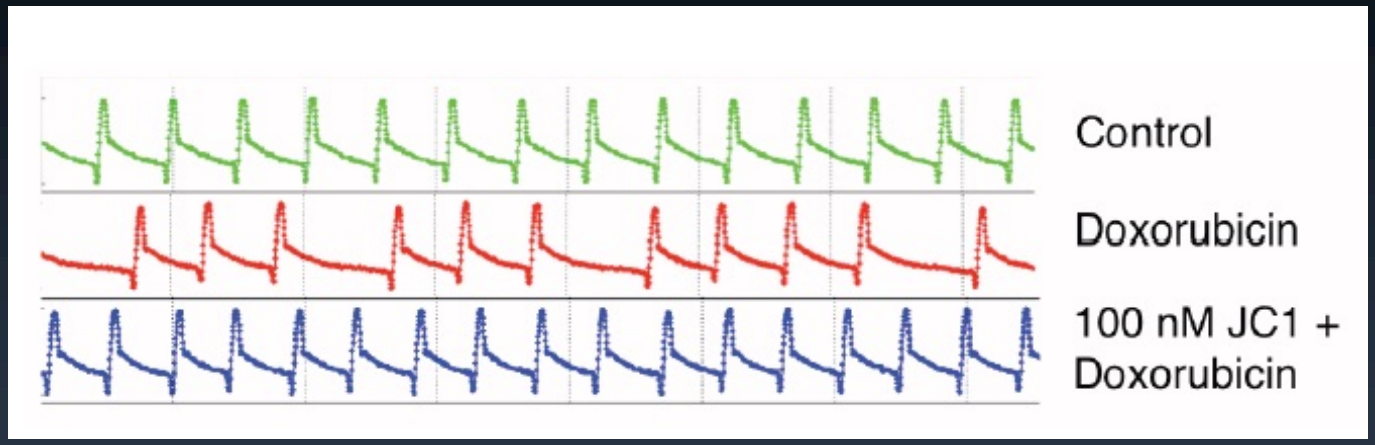


Post-Doxorubicin

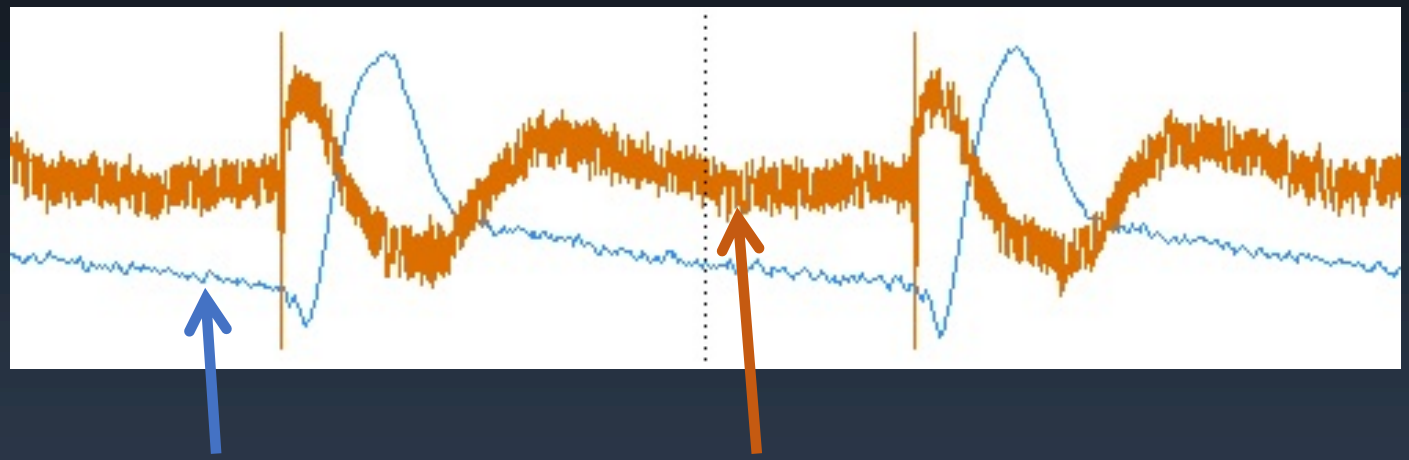




Therapeutic Design



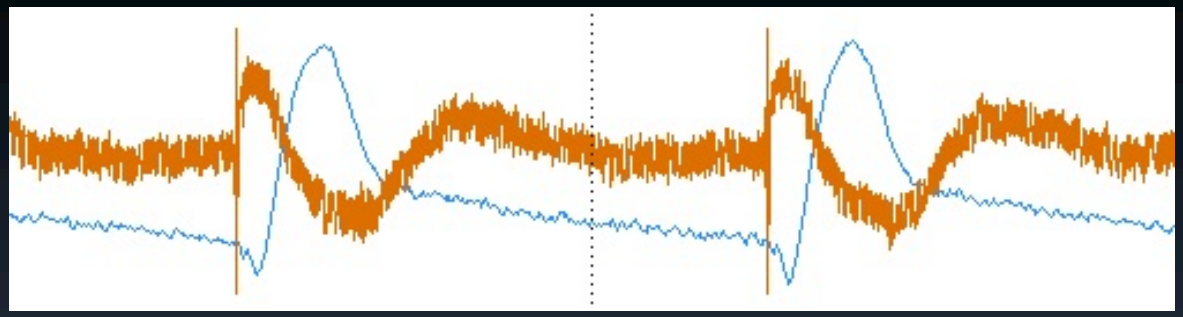
Healthy Cardiomyocyte iPSC-CM Layers



Physical Contraction

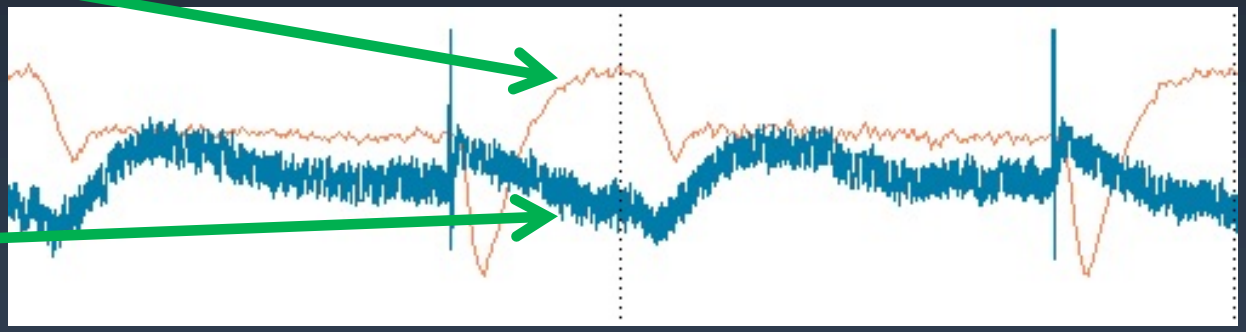
Ion Channel Activity

Healthy Cardiomyocyte iPSC-CM Layers



Impaired Physical Contraction

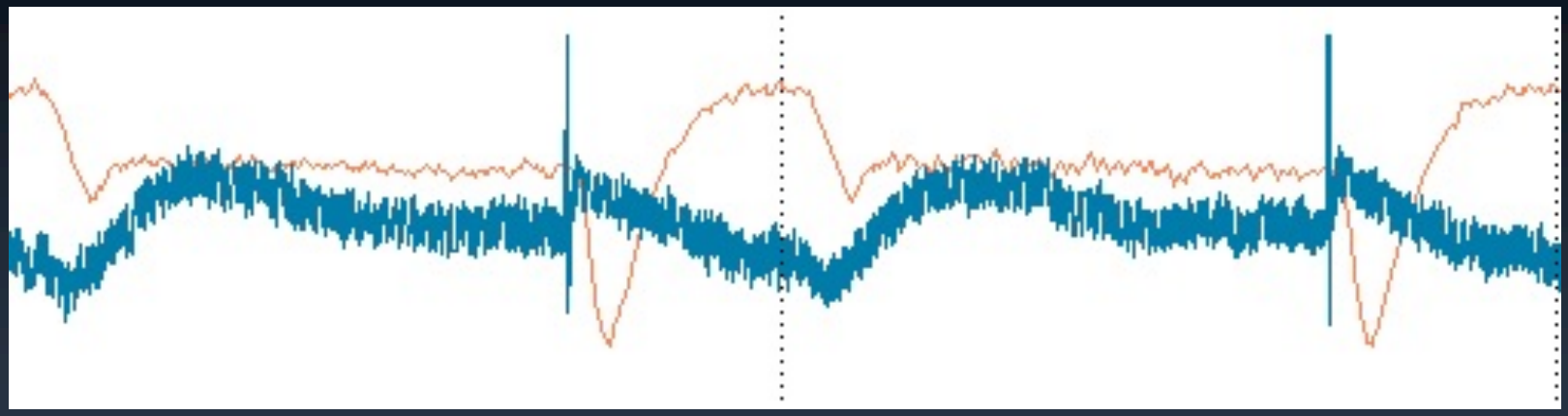
Hypertrophic Cardiomyopathy iPSC- CM Layers (MYH7)



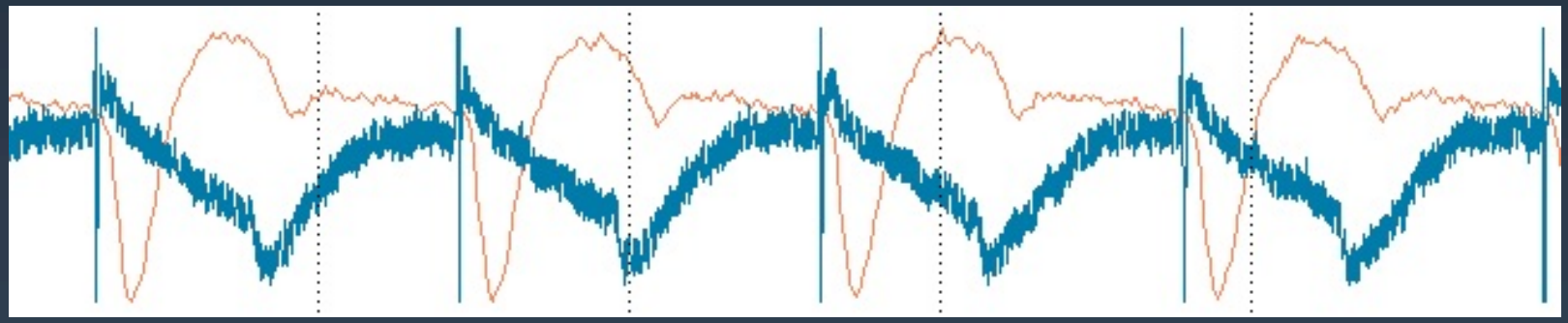
Loss of Ca++ Wave

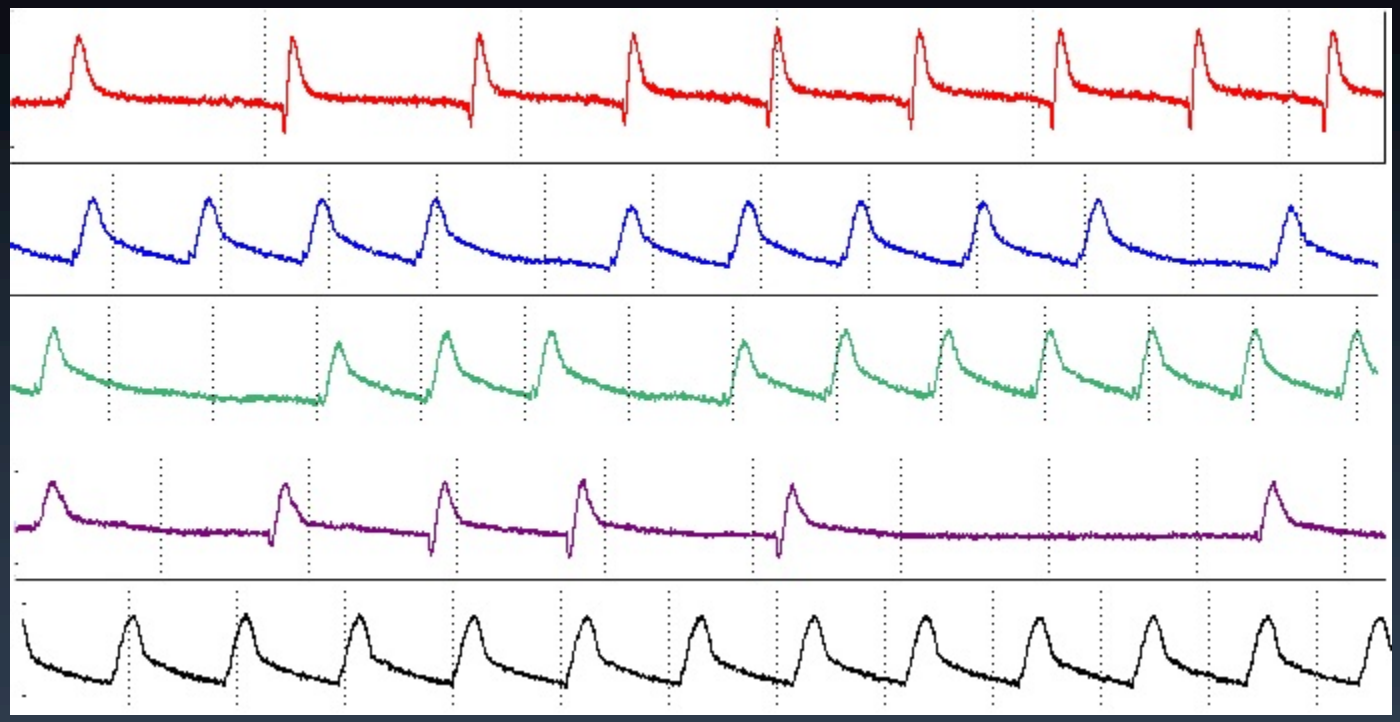
JC-1 Treatment Improves Ca++ Cycling in HCM

Baseline



+JC-1





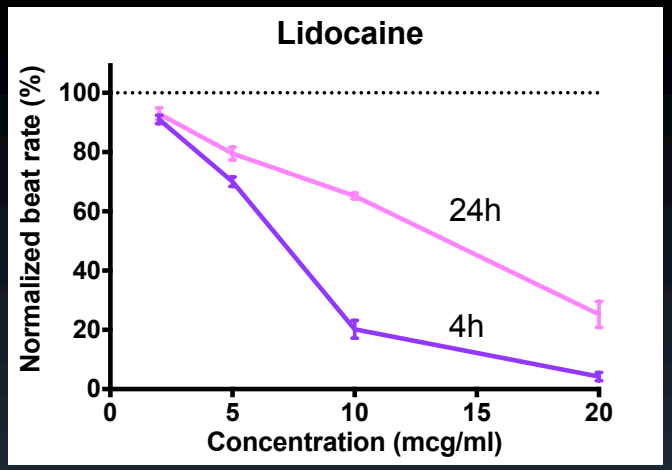
Bupivacaine 3 mg/L

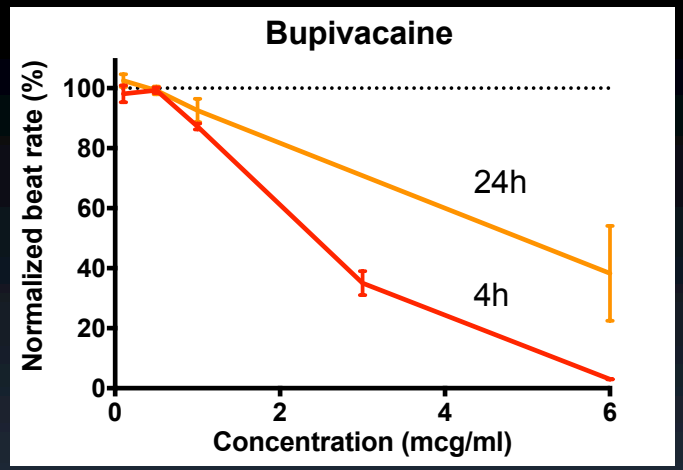
Levobupivacaine 3 mg/L

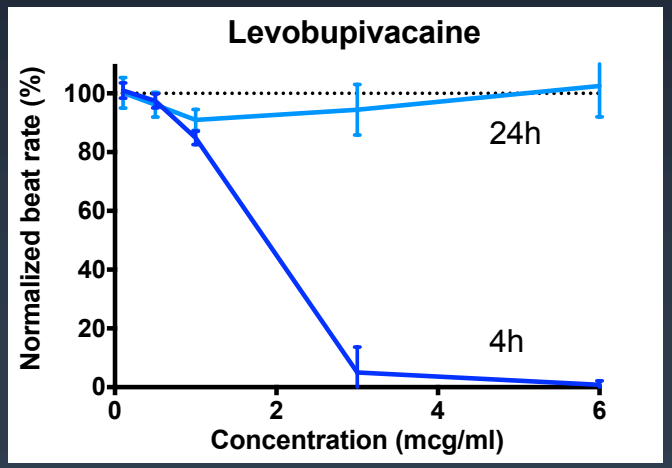
Ropivacaine 3 mg/L

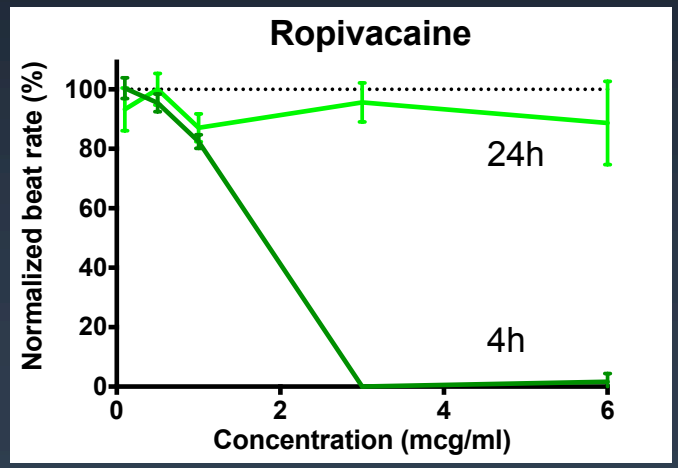
Lidocaine 10 mg/L

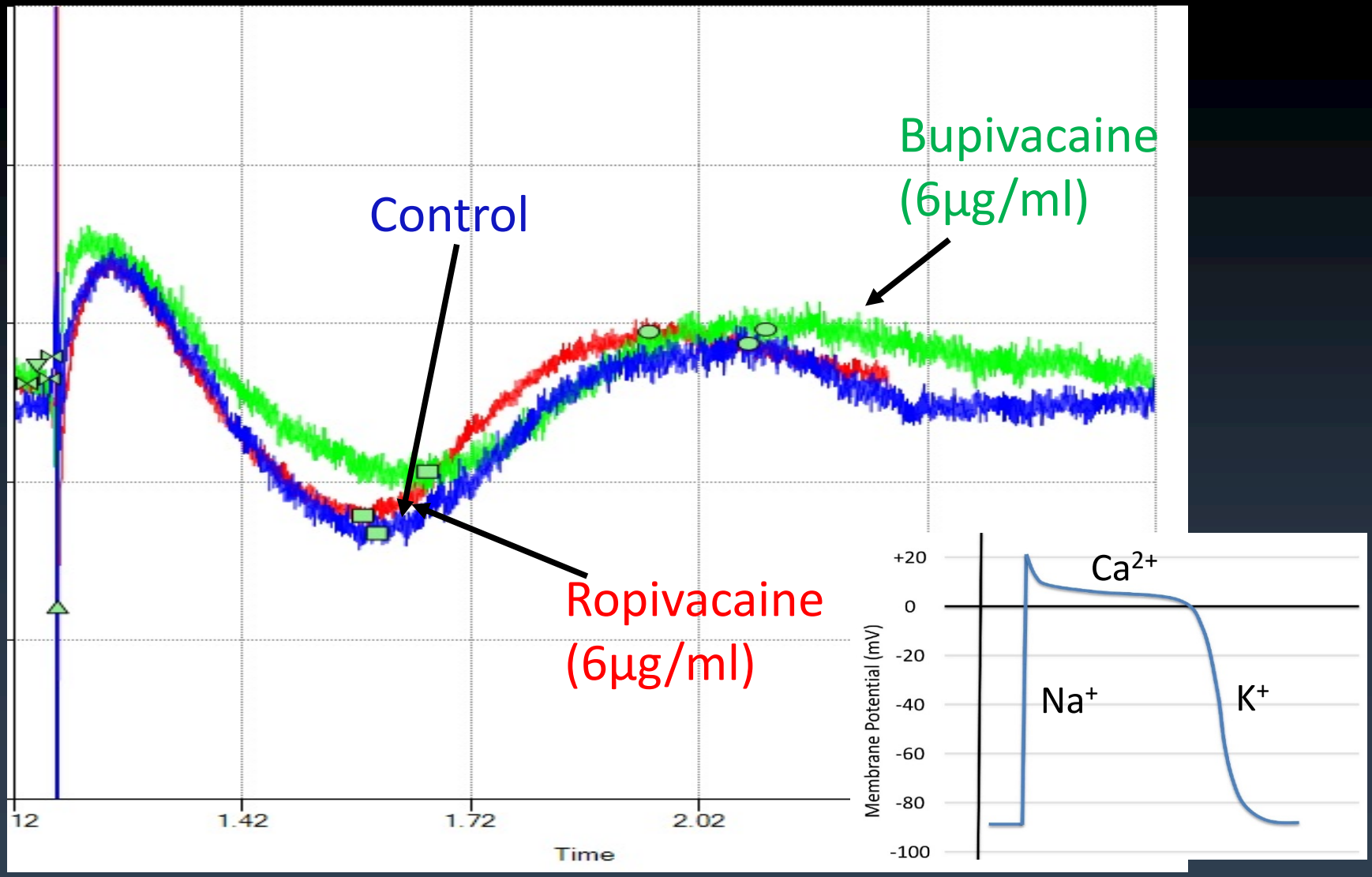
Control


Lidocaine
Normalized beat rate (%)
100
80
60
40
20
0
0
5
10
15
20
Concentration (mcg/ml)
24h
4h

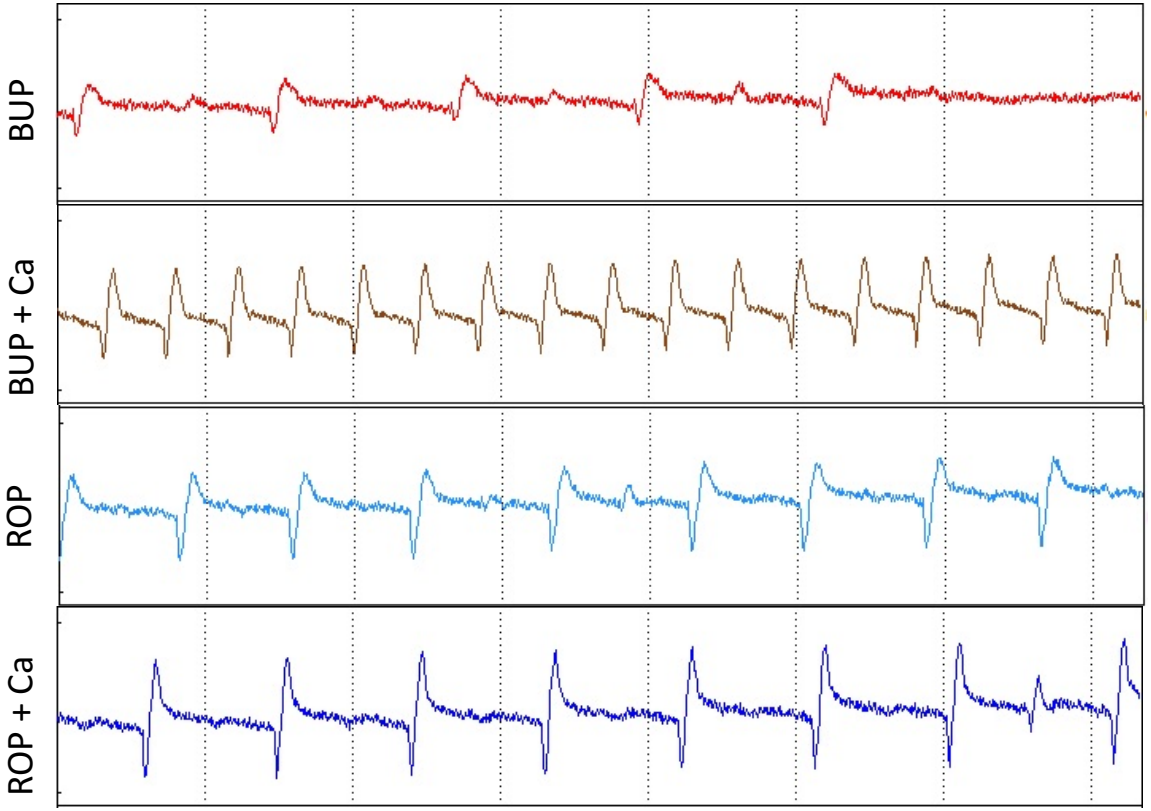

Bupivacaine
Normalized beat rate (%)
100
80
60
40
20
0
0
2
4
6
Concentration (mcg/ml)
24h
4h

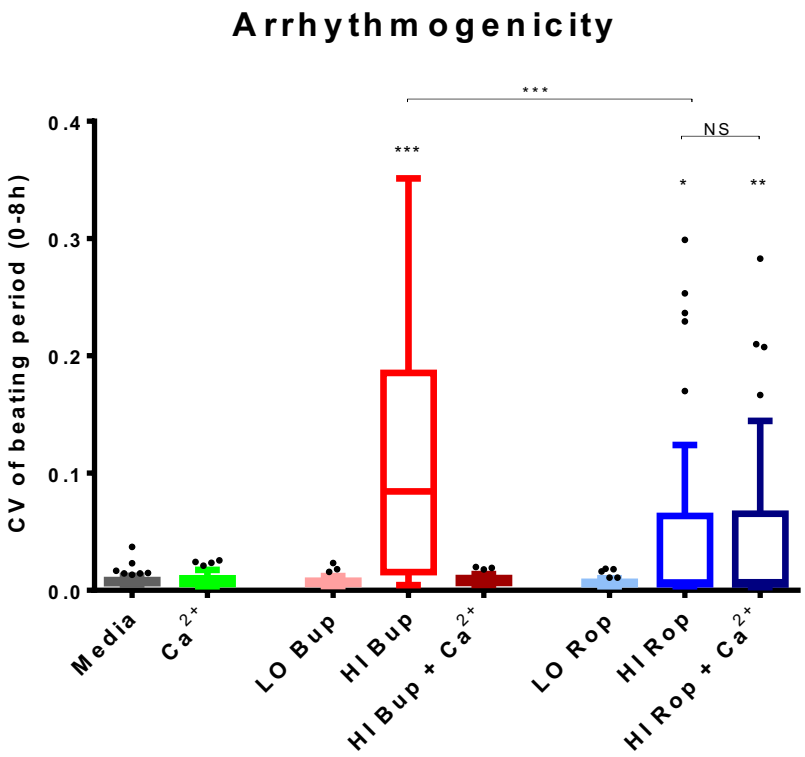

Levobupivacaine
Normalized beat rate (%)
100
80
60
40
20
0
0
2
4
6
Concentration (mcg/ml)
24h
4h

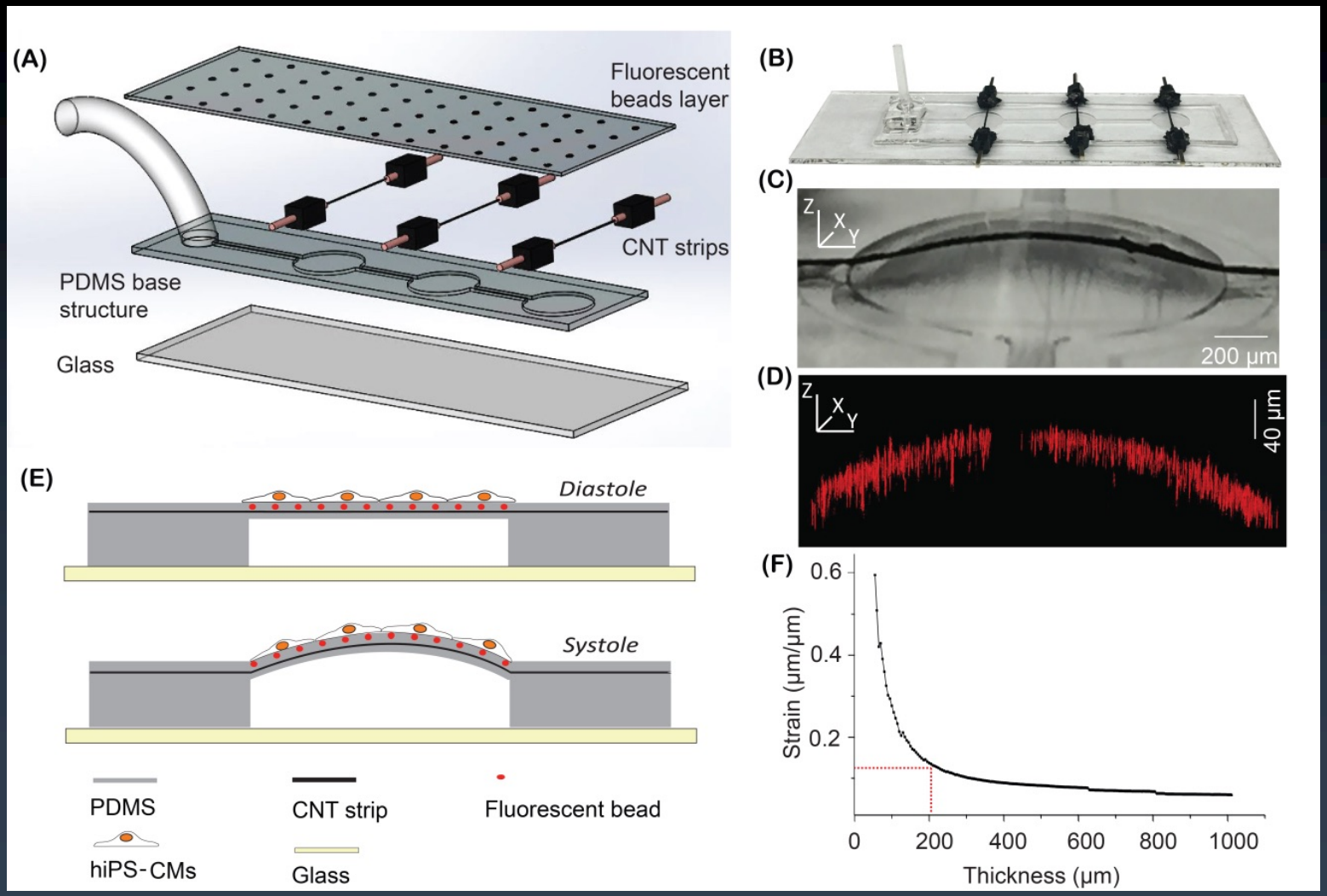

Ropivacaine
Normalized beat rate (%)
100
80
60
40
20
0
0
2
4
6
Concentration (mcg/ml)
24h
4h

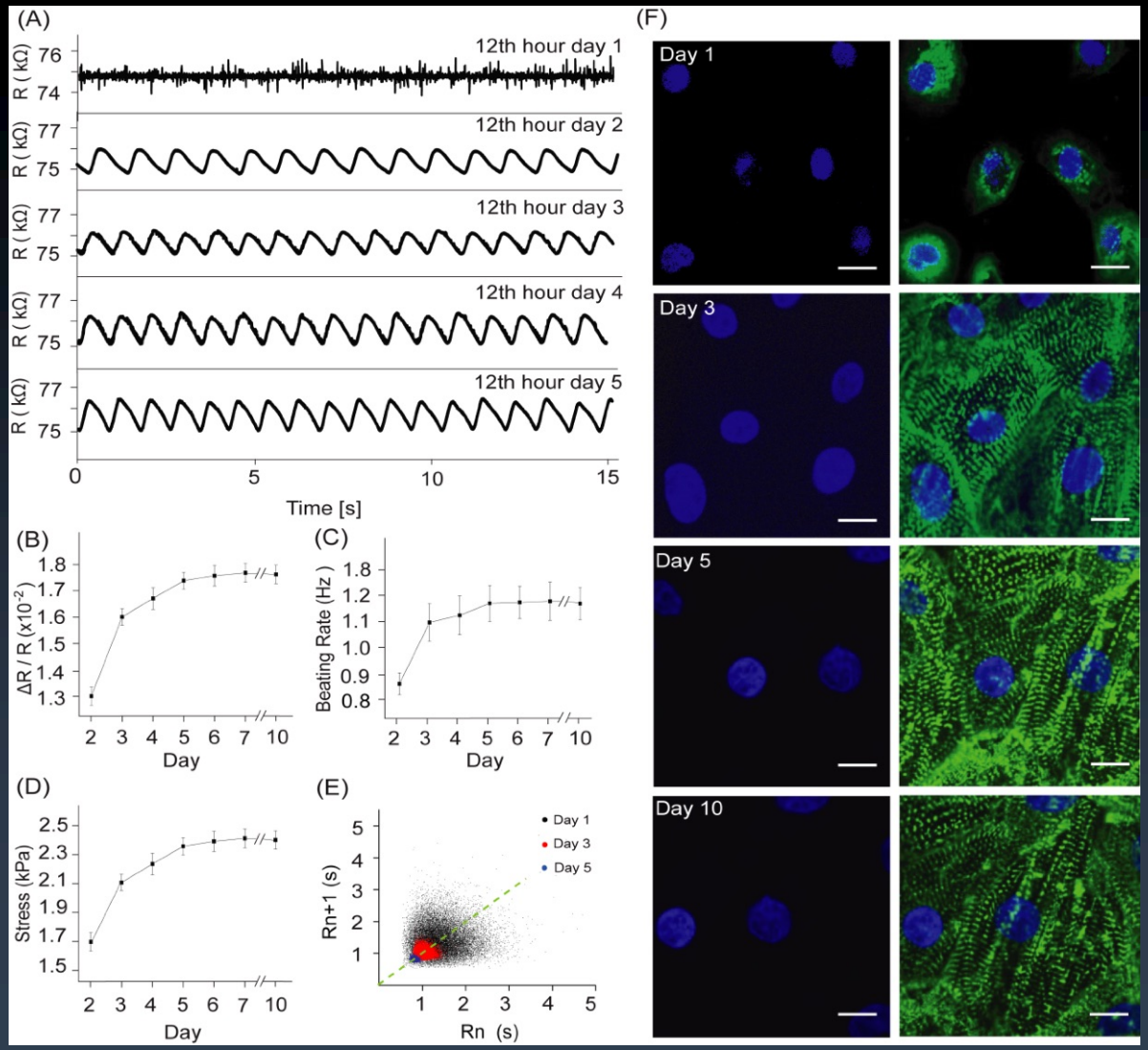

Bupivacaine
(6µg/ml)
Control
Ropivacaine
(6µg/ml)
12
1.42
1.72
2.02
Time
+20
0
-20
-40
-60
-80
-100
Membrane Potential (mV)
Ca2+
Na+
K+

Ca^{2+} Corrects Bupivacaine Arrhythmia

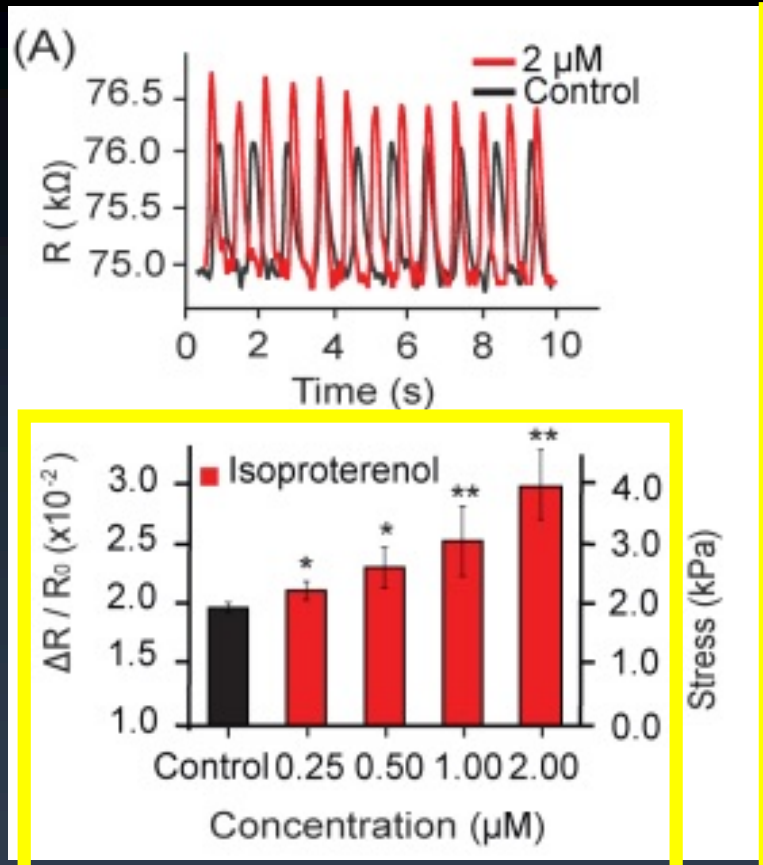




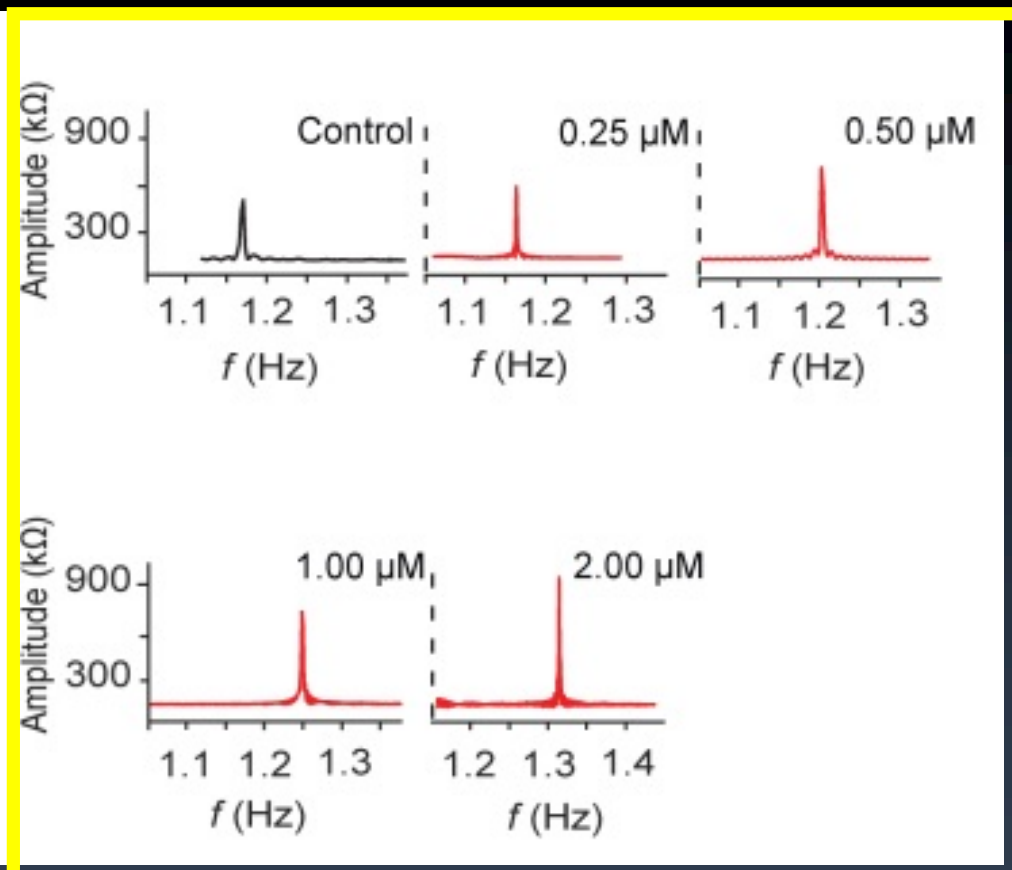

(A)
Fluorescent beads layer
CNT strips
PDMS base structure
Glass
(B)
(C)
Z
X
Y
200 µm
(D)
Z
X
Y
40 µm
(E)
Diastole
Systole
PDMS
CNT strip
Fluorescent bead
hiPS-CMs
Glass
(F)
Strain (µm/µm)
0.6
0.4
0.2
0
200
400
600
800
1000
Thickness (µm)


(A)
12th hour day 1
12th hour day 2
12th hour day 3
12th hour day 4
12th hour day 5
R (kΩ)
Time [s]
(B)
ΔR / R (x10⁻²)
Day
(C)
Beating Rate (Hz)
Day
(D)
Stress (kPa)
Day
(E)
Day 1
Day 3
Day 5
Rn+1 (s)
Rn (s)
(F)
Day 1
Day 3
Day 5
Day 10

Isoproterenol

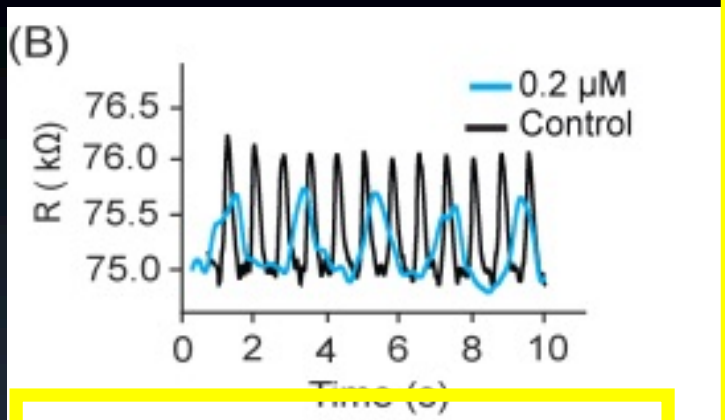


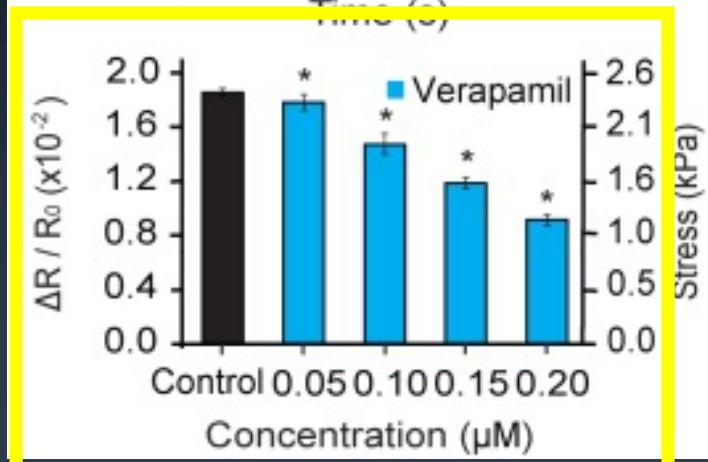
Contractility



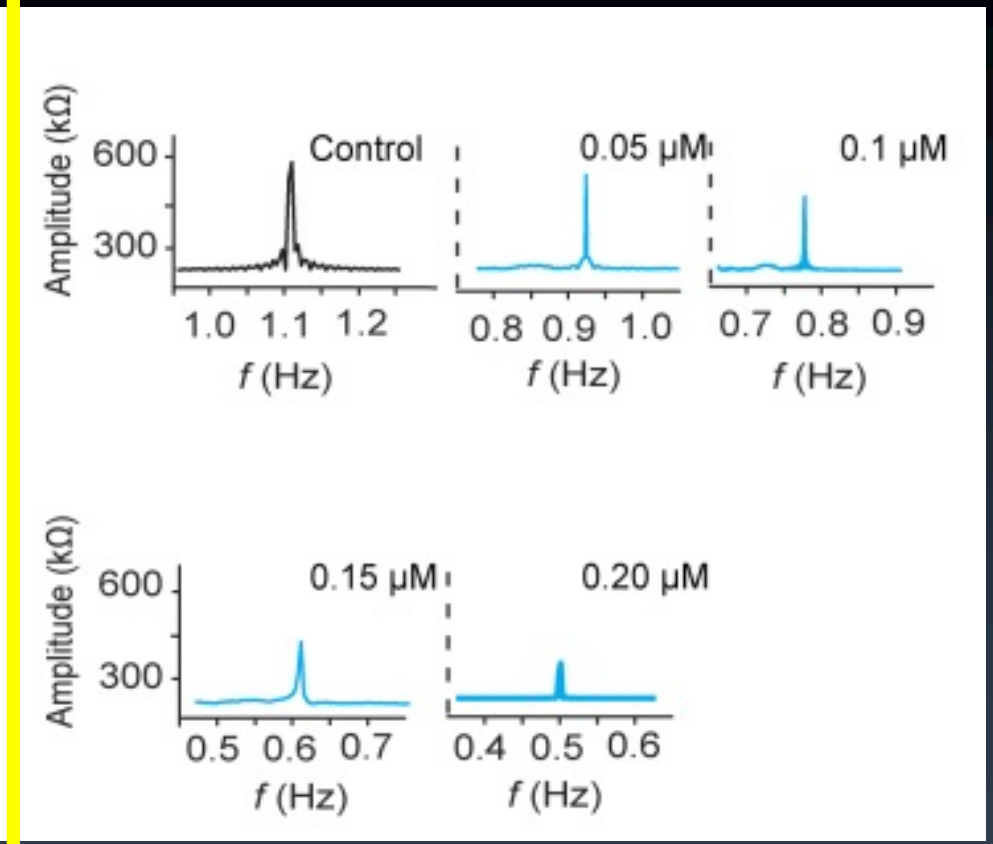
Rate (dose dep increase Hz)

Verapamil



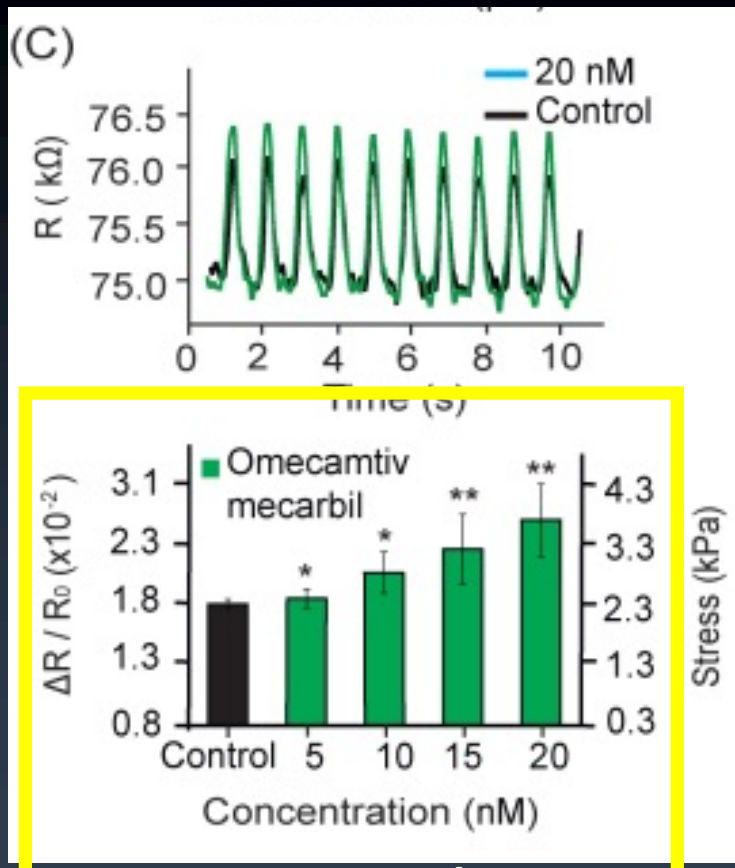


Contractility

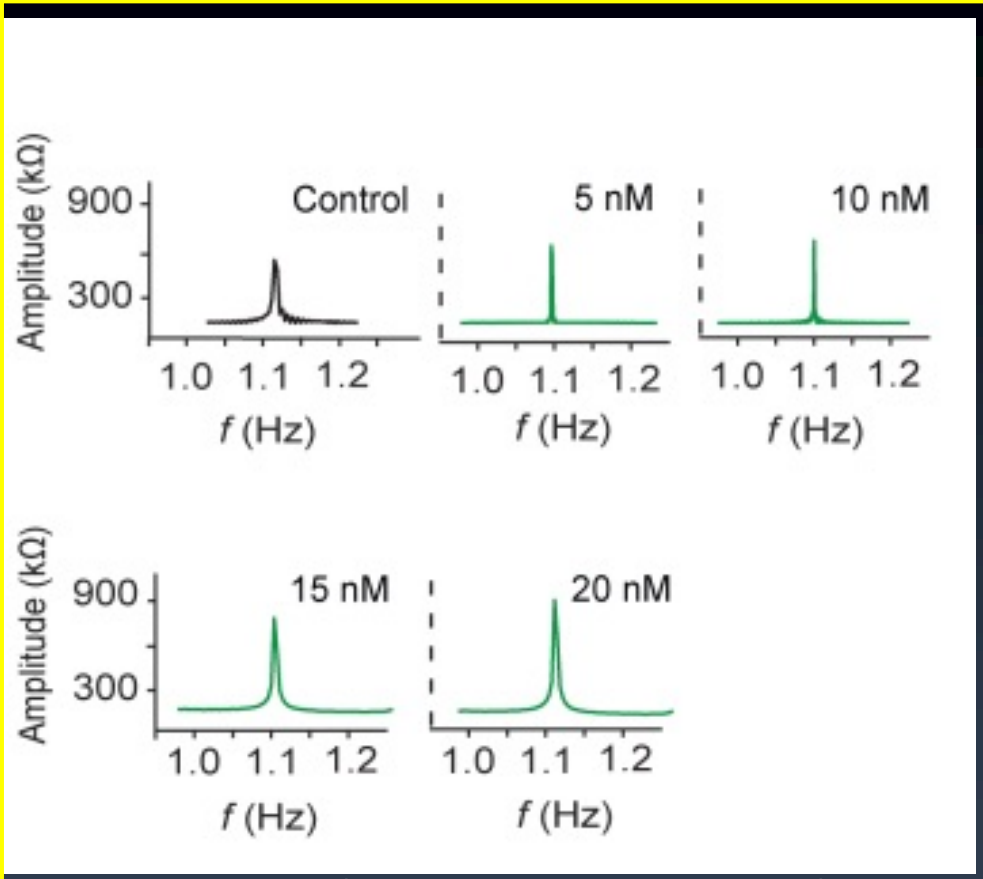


Rate (dose dep decrease Hz)

Omecamtiv

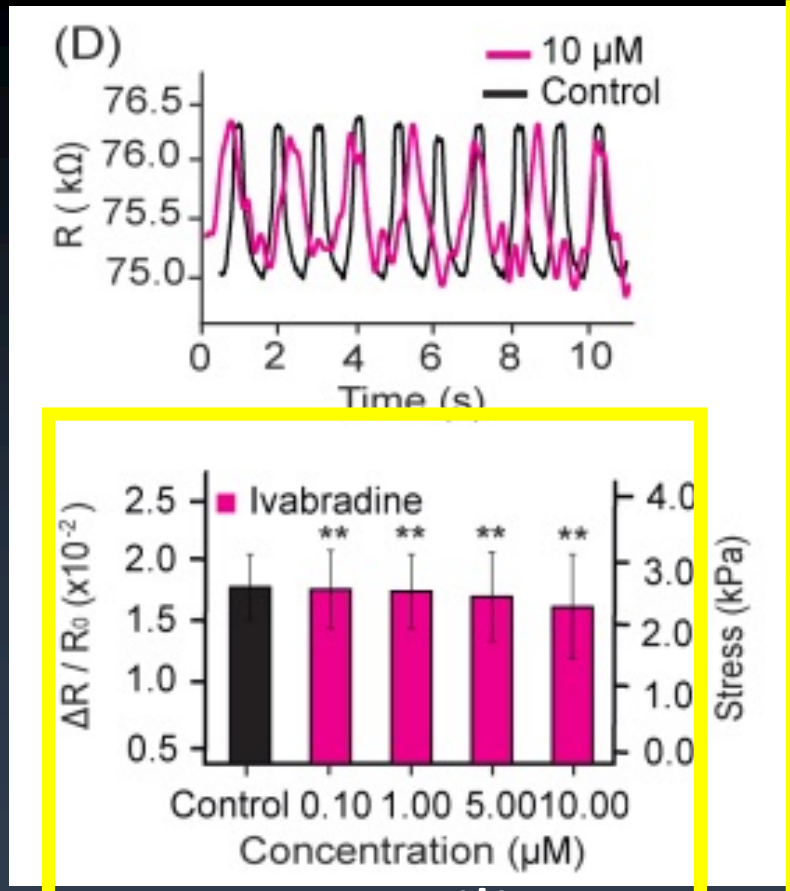


Contractility

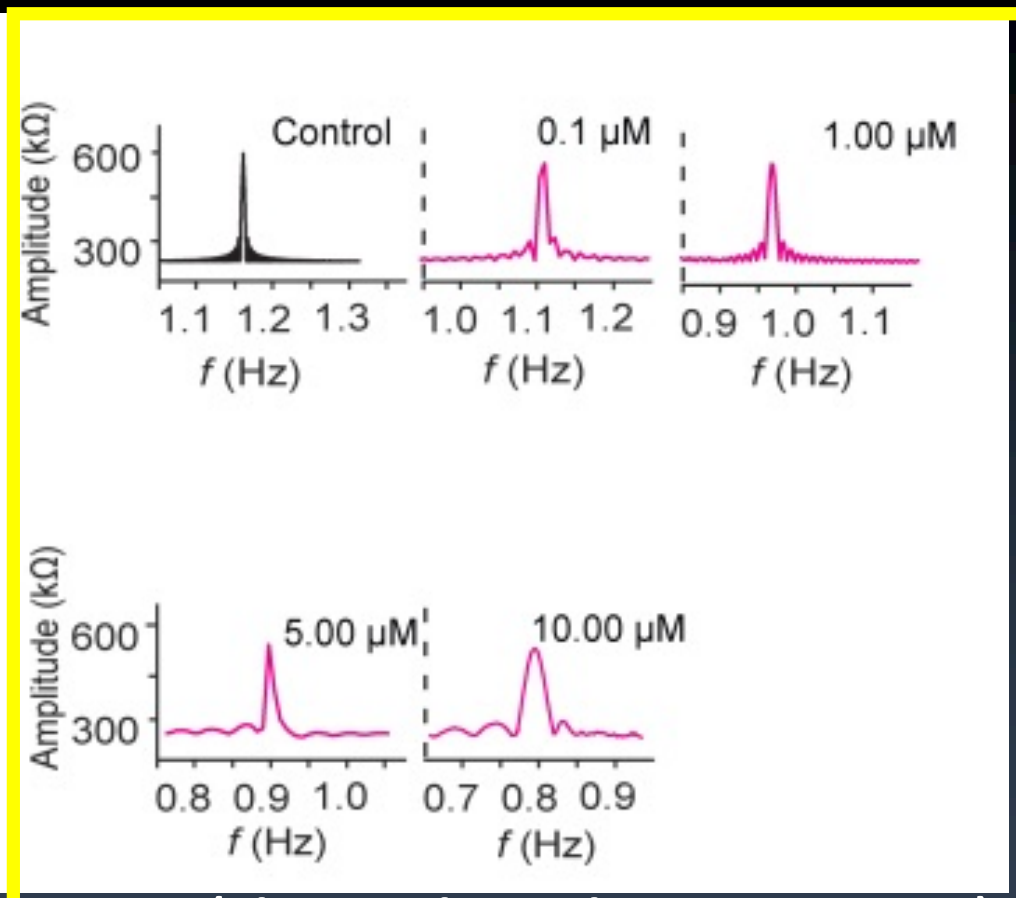


Rate (no change Hz)

Ivabradine



Contractility



Rate (dose dep decrease Hz)

Dangerous cut

The numbers of surgeons involved in research are falling — the trend must be reversed.

27 APRIL 2017 | VOL 544 | NATURE | 393

carried out. More than half said that basic research was a priority in their departments — but just one-third said that it was realistic to expect surgeons to succeed in basic research. Most respondents said they had neither the time nor the motivation for research, and in any case lacked adequate departmental support and funding. Nearly two-thirds believed that basic research among trainees should be limited to a select few residents with the ambition and talent to be successful in future research activities.

"Policymakers must stop seeing surgeons as easy sources of revenue."

surgeons as easy sources of revenue."

that includes regenerative medicine and possibly xenotransplantation (transplantation of tissues and organs from other species). They are also much needed for crucial research into surgically treated diseases that only rarely hit the headlines — particularly in the correction of congenital birth defects, but also in adult

Stimulation of Innovation at The Hospital for Sick Children

- Innovation Fund: ~$150 000 per year in peer-adjudicated funding
  - starter funding to stimulate further applications
- Formal collaborative programme with BME:
  - Clinicians pitch ideas to ideas to students (course)
  - Graduate student collaborative programme (CIGITI)
  - Many graduate appointments of physicians
- High success rate summer student programme
  - Formal mentorship, educational initiatives
- Customized educational pathways for peri-operative physicians
- Research chair funding used for research

Conclusions

- Preclinical models for prediction of drug activity are poorly translational
- Significant inter-individual variance in drug response, with dynamic changes (not necessarily genetic)
- Stem-cell derived tissue can be used to model:
  - Conductive diseases
  - Cardiac physiology
- Drug discovery can be facilitated using iPSC tissue:
  - Impedance-based electrode devices
  - Dynamic CNT contractility systems
- Future: packaging of pro-regenerative neonatal cardiac secretomes into nanoparticles

Acknowledgements

- Maynes Lab:
  - Dr. Ramesh Vanama
  - Dr. Michael Tropak
  - Julia Plakhotnik
  - Manpreet Malhi
  - Dr. Yanan Tang
  - Dr. Olga Chernya
  - Kaley Hogarth
  - Henry Mah
  - Dr. Libo Zhang
  - Doorsa Tarazi
  - David Pompili
- Coles Lab:
  - Alex Traister
  - Chris Welsh
  - Sarv Patel
- Collaborators:
  - Boris Hinz
  - Yu Sun

oirm Ontario Institute for Regenerative Medicine

MaRS Innovation

HEART & STROKE FOUNDATION

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