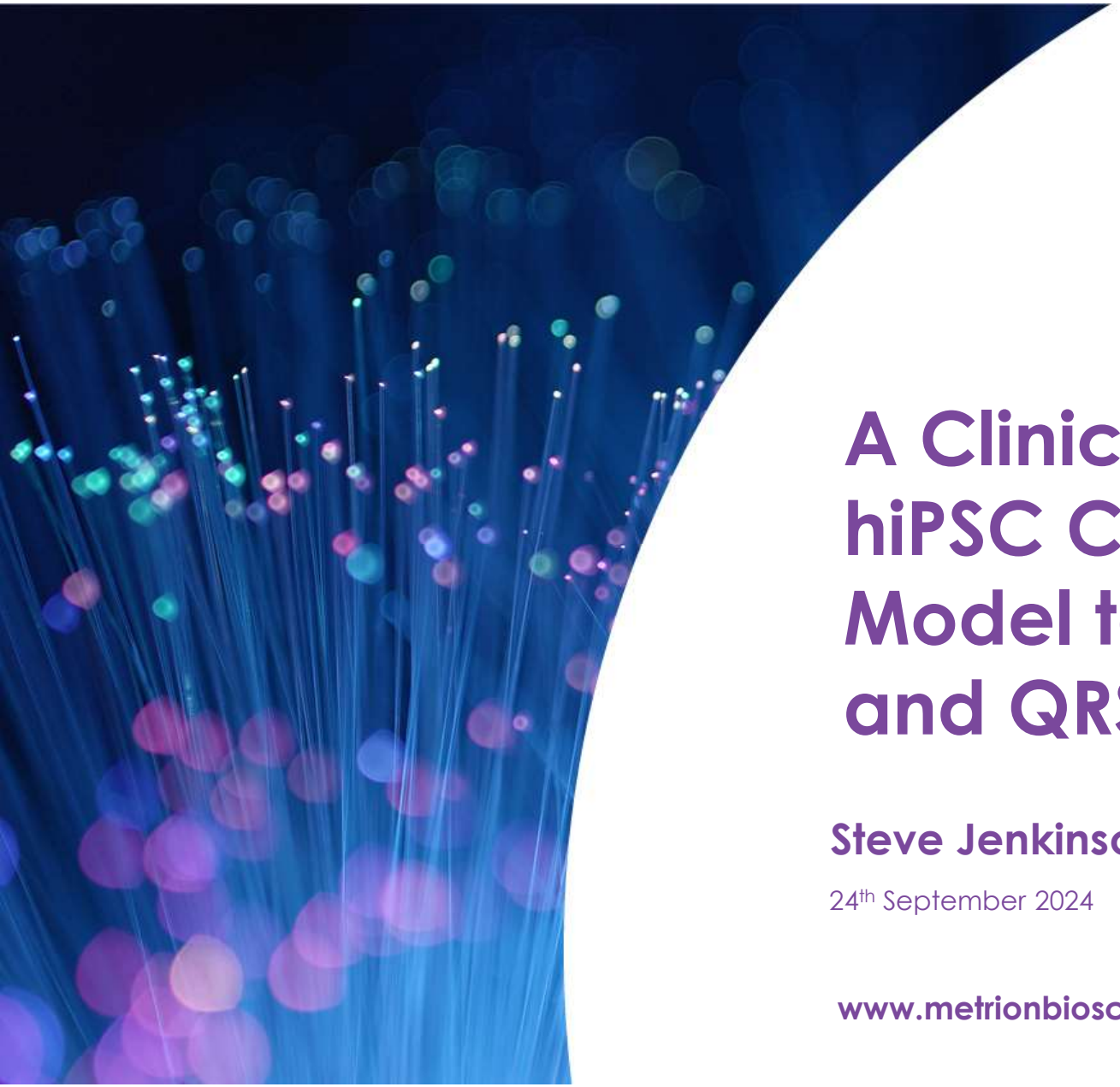

A Clinically Translatable hiPSC Cardiomyocyte Model to Predict QTc and QRS Cardiac Risk

Steve Jenkinson

24th September 2024

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hiPSC Cardiomyocytes in Defining CV Risk

Introduction

- hiPSC-CMs are now routinely used in industry to assess CV effects of novel compounds
  - Functional endpoints - Contractility
  - Electrophysiological endpoints – QTc/QRS risk assessment
- Importance noted by regulators resulting in change in ICH guidelines
  - Driven initially by CiPA initiative
  - Formalized by inclusion in revised ICH S7B Q&As
    - hiPSC-CM data can be used to support a Thorough QT (TQT) waiver application
- Key requirement of any hiPSC-CM assay is an understanding of its translation to the clinic

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Introduction

- Presentation describes a high throughput hiPSC-CM model that can
    - Predict exposures of a novel compound that would be associated with a 10 ms change in clinical QTc interval
    - Define the probability of QRS prolongation risk
    - Assess general CV toxicity liability
    - Acutely (30 min) and chronic (24 h) endpoints examined
    - Assay performed in serum free conditions

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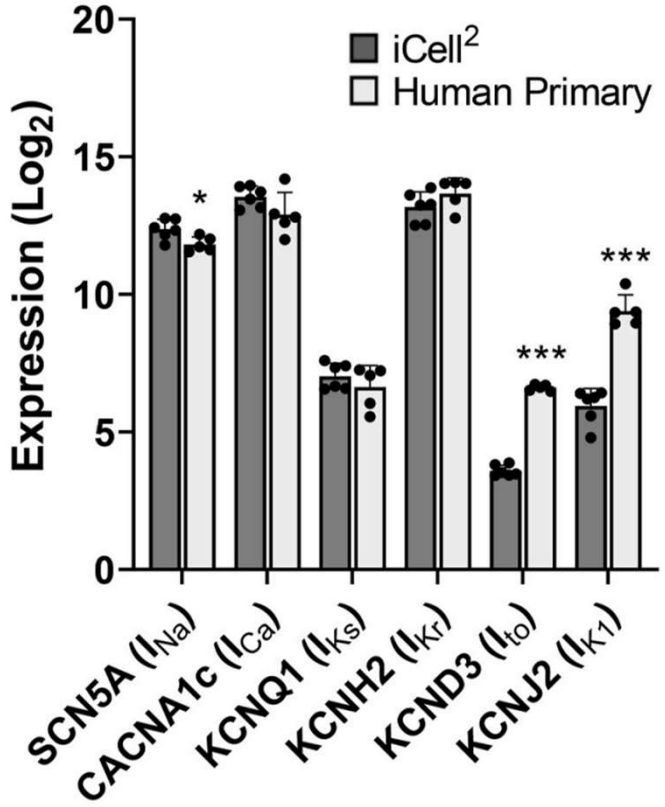
Cell System

iCell^2 cardiomyocytes
(FUJIFILM Cellular Dynamics Inc.)

- Key ion channels involved in cardiac AP generation show similar expression to purified human ventricular cardiomyocytes



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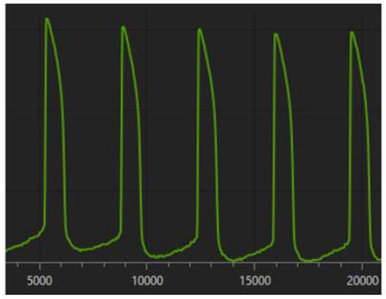
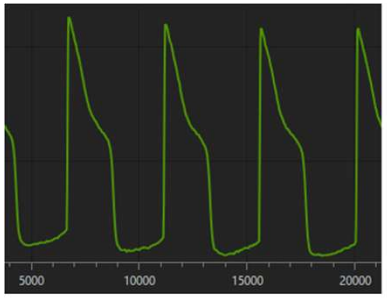
The Technology

Action potential recordings from intrinsically paced hiPSC-CMs

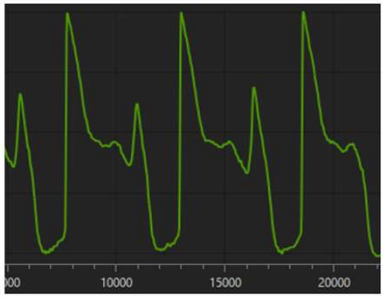
- Volta Fast Optical Reader (Lumencor Inc.)
- Utilizes voltage sensitive membrane dye (BeRST)
- Simultaneous reads 96 well plate at 10,000 Hz
- Recordings equivalent in quality to patch clamp



Vehicle



AP Prolongation



Early After Depolarizations

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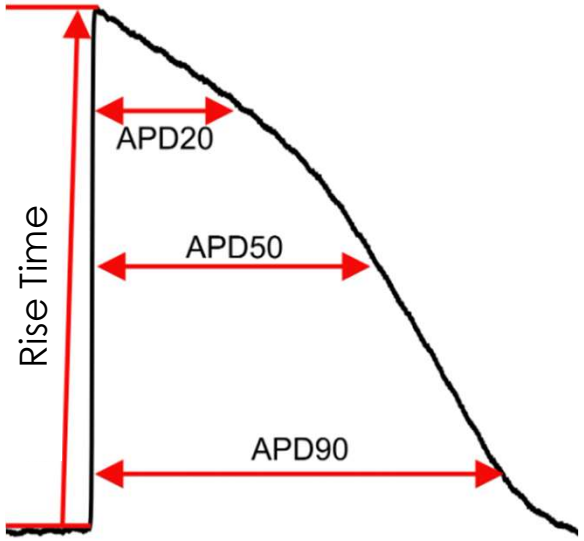
Assay Endpoints

Concentration-response curves for multiple endpoints

- 10-point concentration response curves
- Endpoints measured
  - Action potential duration (APD)
  - Rise time
  - Beat rate
- Acute (30 min) and chronic (24 h) measurements
- Protein/serum free assay



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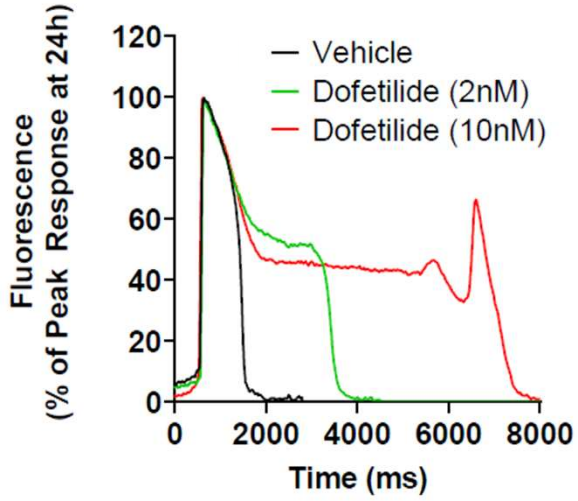
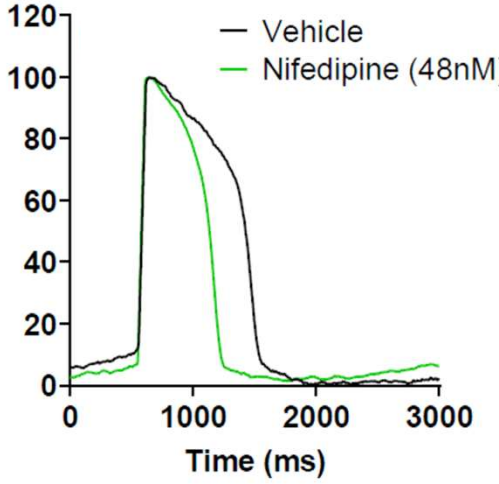
Example Data

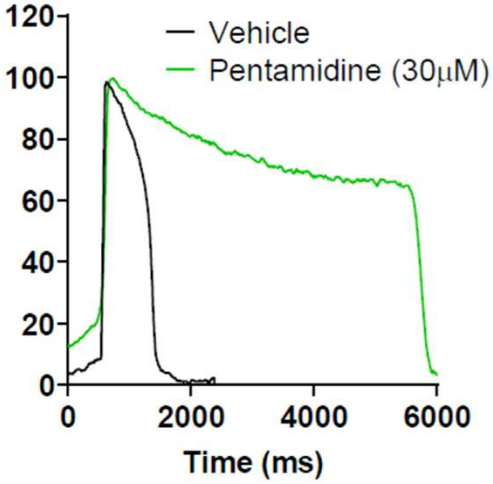
hERG Inhibition



$Ca_V1.2$ Inhibition



hERG Trafficking Inhibition



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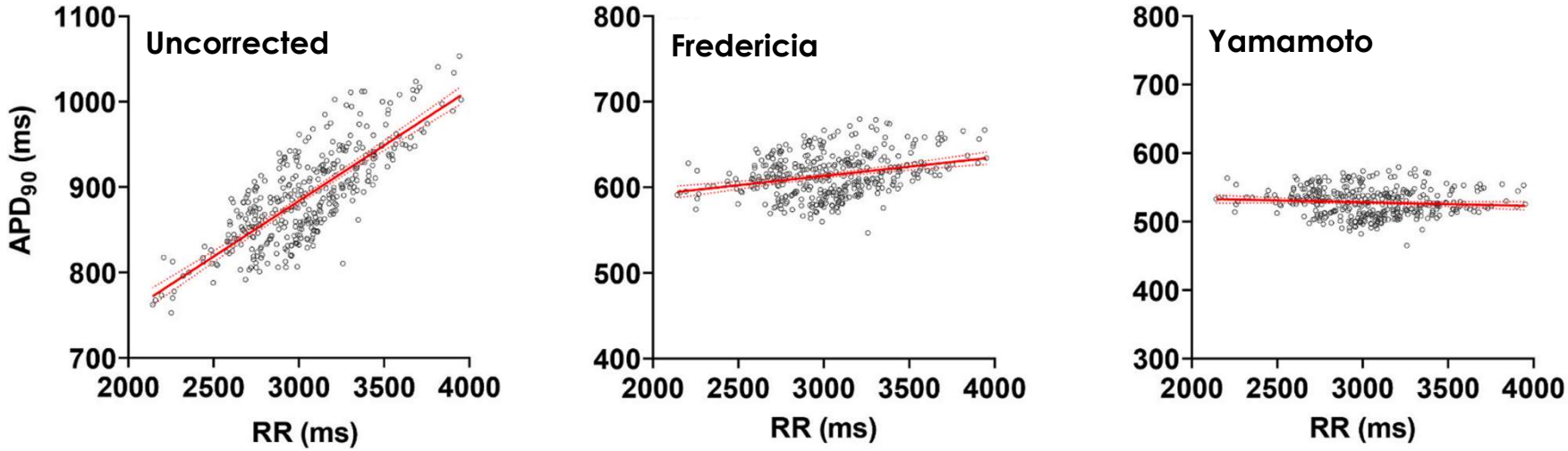
Rate Corrected APD_{90}

APD_{90} rate correction

- Yamamoto correction ($ycAPD_{90}$) is optimal method for hiPSC-CMs
- $ycAPD_{90}$ is a surrogate of clinical QTc



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Predicting Clinical QTc Risk Using hiPSC-CMs

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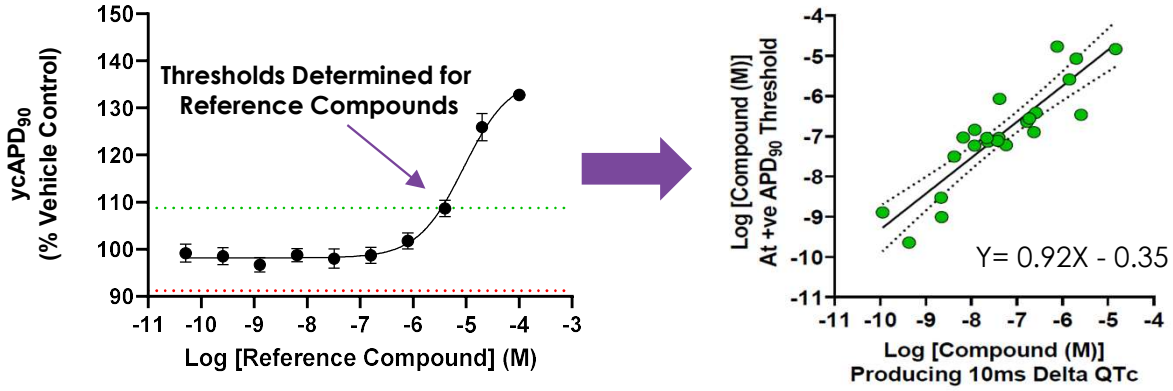
Prediction of Clinical QTc

- Ability to determine QTc risk was assessed using a series of reference compounds
  - 23 QTc positive reference compounds with available clinical data
- The analysis compared
  - The concentration associated with hiPSC-CM APD_{90} threshold value
    - 3x Vehicle Std Dev
  - Free clinical exposure producing 10 ms QTc change



Reference Compounds		
Azimilide*	Ibutilide*	Ribociclib
Bepridil*	Mesoridazine	Sotalol*
Cisapride*	Moxifloxacin	Terfenadine*
Citalopram	Odansetron*	Terodiline
Dofetilide*	Procainamide	Thioridizine
Droperidol	Quinidine*	Tolterodine
E4031	Quinine	Vandetanib*
Halofantrine	Ranolazine*	

*CiPA 28 compound

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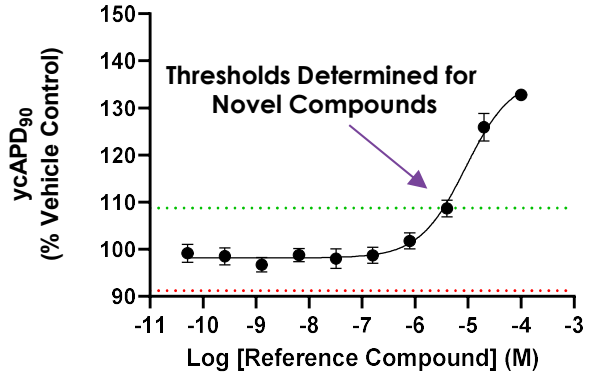
Prediction of Clinical QTc

Ability to predict the free clinical exposure associated with a 10 ms change in QTc for novel compounds



Input Threshold Value Into Equation (Y=0.92X – 0.35)

Prediction of Free Clinical Exposure Producing a 10 ms QTc Change

- Predictivity confirmed using a 4-fold cross validation analysis
- hiPSC-CM data available for an additional 43 reference compounds (66 in total)
  - Including all CiPA-28 compounds

Assessing the Probability of Clinical QRS Risk Using hiPSC-CMs

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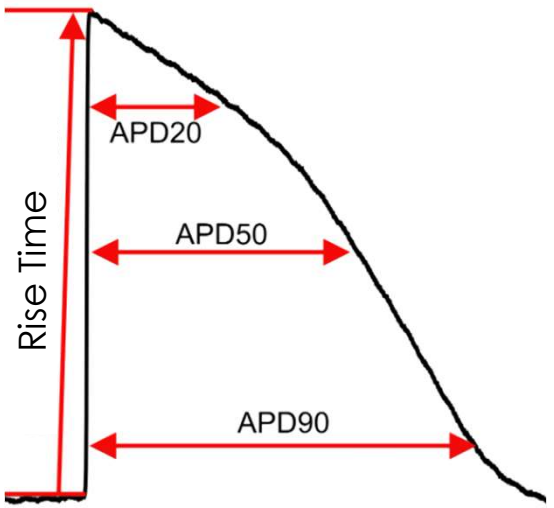
Probability of Clinical QRS Liability

- Action Potential Rise Time is associated with $Na_v1.5$ channel activity
- $Na_v1.5$ block is associated with QRS prolongation in the clinic
- hiPSC-CM assay utilizes Rise Time to assess clinical QRS risk

- Available clinical QRS data less defined than for QTc
  - Clinical data used in analysis
    - Clinician defined effect on QRS (Binary - Yes/No)
    - Free clinical exposure associated with finding
  - Stem cell data used in analysis
    - Rise Time threshold concentration



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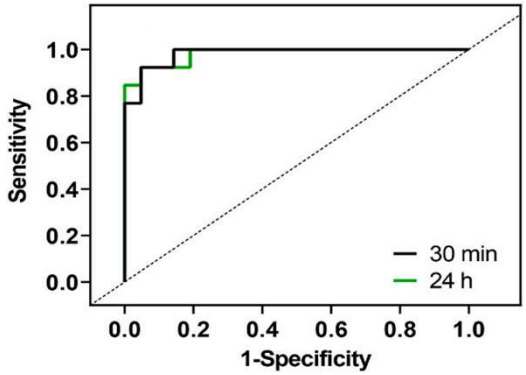
Probability of Clinical QRS Liability

Performed ROC analysis to define hiPSC-CM/clinical data association

- 22 QRS positive / 12 QRS negative compounds
- Compared QRS effect (Yes/No) versus ratio of stem cell rise time threshold concentration versus free clinical exposure associated with finding



Incubation	AUROC (95% CI)	Sensitivity (95% CI)	Specificity (95% CI)	Cut Point	N Value
30 min	0.98 (0.95, 1.01)	0.92 (0.64, 1.00)	0.95 (0.76, 1.00)	33.0	34
24 h	0.98 (0.95, 1.02)	0.92 (0.64, 1.00)	0.95 (0.76, 1.00)	56.4	34

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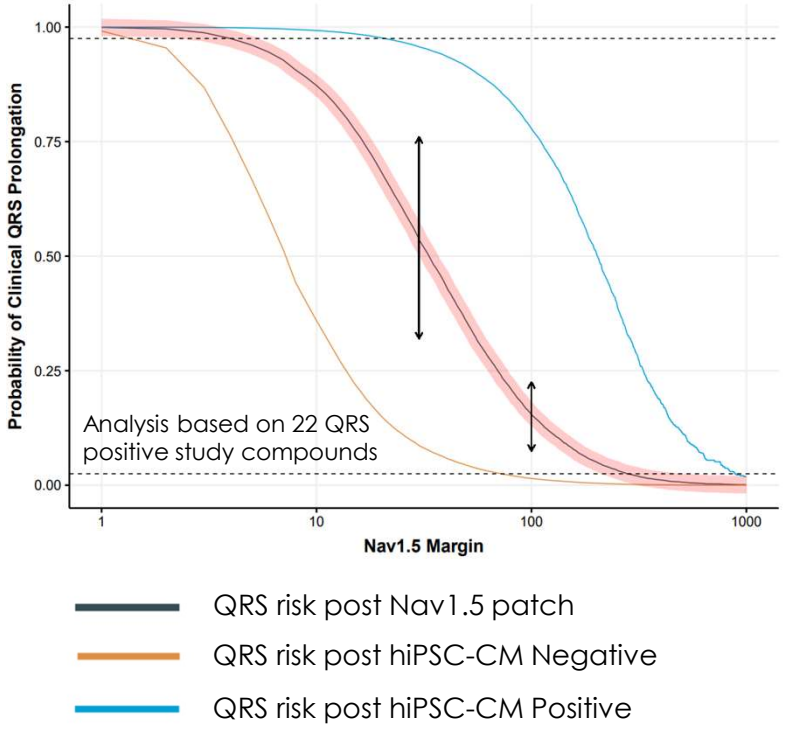
Probability of Clinical QRS Liability

Defining QRS probability

- QRS probability at a clinical exposure equivalent to
  - hiPSC-CM Rise Time threshold concentration
    - 74 - 86 %
  - 33-fold (30 min) or 56-fold (24 h) lower than Rise Time threshold concentration
    - 1.2 - 2.4%
  - Analysis assumes Nav1.5 prevalence of
    - 12.7 - 23.5%

Impact of hiPSC-CM Data On Defining QRS Risk



Case Study

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Case Study

- Compound ion channel profile
  - hERG IC_{50} = ~20 µM
  - $Na_v1.5$ IC_{50} = ~115 µM
  - $Ca_v1.2$ IC_{50} = ~280 µM
- QTc and QRS prolongation observed in the clinic
  - Free clinical exposure ~1 µM
- QRS liability was unexpected based on $Na_v1.5$ patch clamp data
  - hiPSC predicted 10 ms change in QTc plus QRS risk around 1 µM free exposure
- Further analysis suggests that $Na_v1.5$ block had components of state, use/rate dependency
- **Holistic hiPSC-CM model was able to predict effects missed in routine ion channel profiling**

hiPSC Cardiomyocytes in Defining CV Risk

Summary

- hiPSC-CM model is valuable in assessing CV risk
  - Ability to predict exposure associated with a 10 ms QTc change in the clinic
  - Assess probability of clinical QRS risk and associated exposure
  - Also valuable in assessing long term general toxicity
- Provide insight into potential mechanism of CV effects (hERG, $Na_v1.5$, $Ca_v1.2$)
- Assessment of acute (30 min) or chronic (24 h) effects of novel compounds
- Assay is valuable in assessing CV liabilities of novel compounds in a holistic integrated system

Questions?

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