A Human Cardiomyocyte-Based Platform for the Profiling of Drug-Induced Effects on Cardiac Contractility: Predicting Inotropic Mechanisms of Action

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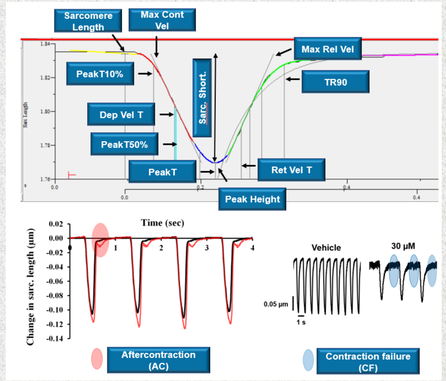
Introduction and Methods

Drug-induced effects on cardiac contractility can lead to serious adverse events including heart failure and therefore limit the utility of innovative treatments. We sought to develop a human cardiomyocyte contractility assay that has the potential to simultaneously predict drug-induced inotropic risk and generate multi-parameter data to profile different inotropic mechanisms of action. Adult human primary cardiomyocytes from ethically consented organ donors were used to measure contractility transients using an imaging-based platform[1]. We tracked changes in contractility parameters to infer both drug-induced inotropic effect (sarcomere shortening) as well as the mechanisms of action based on cluster analysis of a set of 12 contractility parameters. We addressed the relevance of this approach using a panel of 26 inotropes (17 positive, 9 negative) covering diverse mechanisms of action. Each inotrope was tested separately at multiple concentrations.

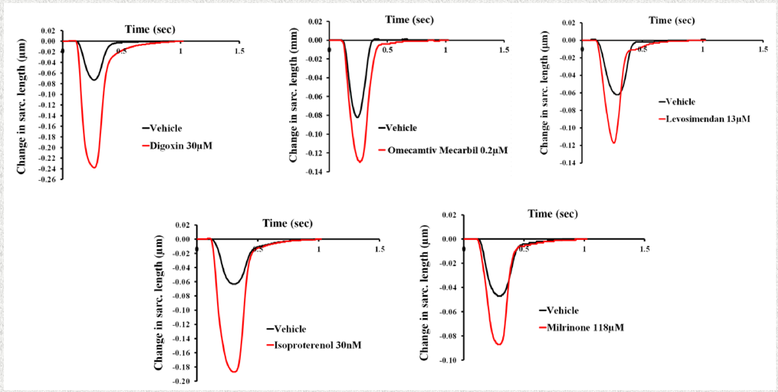
Panel of inotropes with diverse mechanisms of action

Inotropic Effect	Mechanism of Action	Drug
Positive	Na^+/K^+ pump inhibition	Digoxin
Positive	Na^+/K^+ pump inhibition	Ouabain
Positive	Na^+/Ca^{2+} exchanger inhibition	SEA-0400
Positive	Myosin activation	Omecamtiv Mecarbil
Positive	Myosin activation	EMD-57003
Positive	Ca^{2+} sensitization	Levosimendan
Positive	Non-selective b-adrenoceptor activation	Isoproterenol
Positive	Non-selective b-adrenoceptor activation	Epinephrine
Positive	b1-adrenoceptor activation	Dobutamine
Positive	PDE3 inhibition	Milrinone
Positive	PDE inhibition	IBMX
Positive	Ca^{2+} channel activation	Bay-K 8644
Positive	Adenylyl cyclase activation	Forskolin
Positive	Adenylyl cyclase activation	NKH-477
Positive	Calcemia	CaCl2
Positive	SERCA activation	N106
Positive	RyR activation	Caffeine
Negative	SERCA inhibition	Thapsigargin
Negative	RyR inhibition	Ryanodine
Negative	Ca^{2+} channel inhibition	Nitrendipine
Negative	Ca^{2+} channel inhibition	Nifedipine
Negative	Ca^{2+} channel inhibition	Diltiazem
Negative	Ca^{2+} channel inhibition	Mibefradil
Negative	Ca^{2+} channel inhibition	Verapamil
Negative	Na+ channel inhibition	Mexiletine
Negative	Na+ channel inhibition	Flecainide

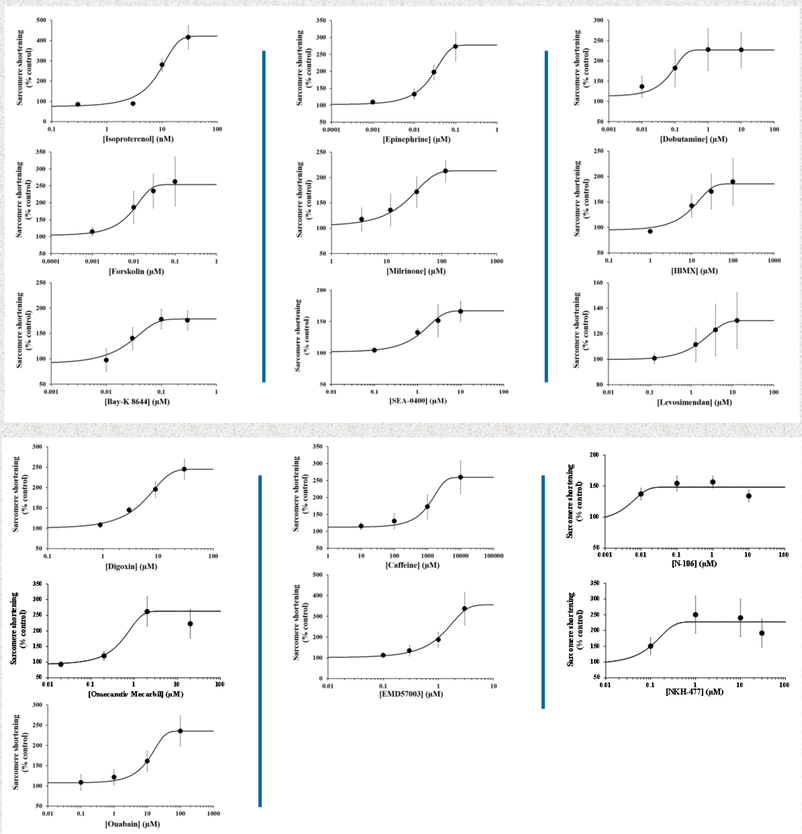
Contractility transient parameters



Identification of drug-induced positive inotropic effects

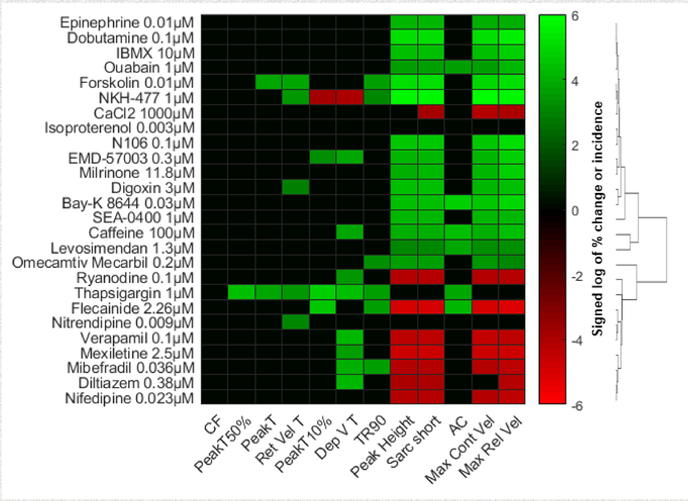


Concentration-Effect curves

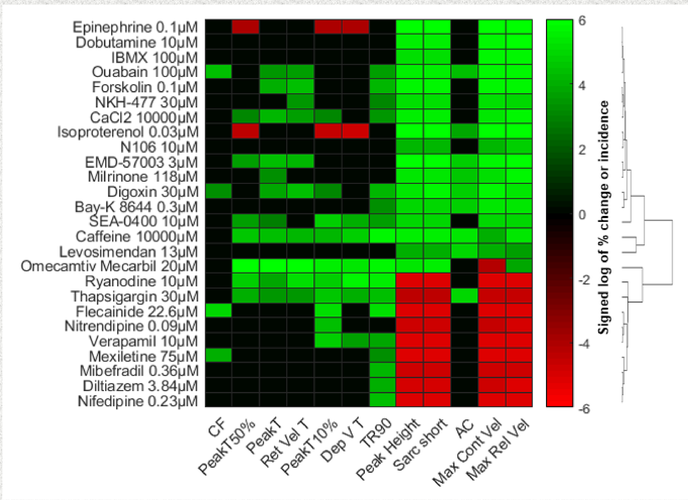


Differentiation of drugs operating via distinct mechanisms

Heatmap generated from 2nd concentration data



Heatmap generated from 4th concentration data



Right-skewed percentage data of incidence (AC and CF) and change (all other transient parameters) were normalized with a signed log transformation. Heatmap representations were created with a cluster analysis using dendrograms and partitions given the elbow criterion. Red and green colors indicate decrease and increase of >25% and >10% change, respectively. **Black** colors indicate no effect (<-25% < % change < 10%). Treatment % effects were expressed relatively to the myocyte's specific baseline control period. AC and CF were expressed as incidence: number of cells showing events normalized by the total number of cardiomyocytes.

Summary

The adult human primary cardiomyocyte-based platform:

1. Enables the identification of the inotropic potential of novel molecules.
2. Facilitates informed mechanistic-based decision making and risk management at the preclinical stage.
3. Scalable, efficient and predictive.

1- Nguyen N et al., Front Physiol 8 (2017) 1073 doi: 10.3389/fphys.2017.01073. eCollection 2017