

Combining Physiological Relevance and Throughput for In Vitro Cardiac Contractility Measurement

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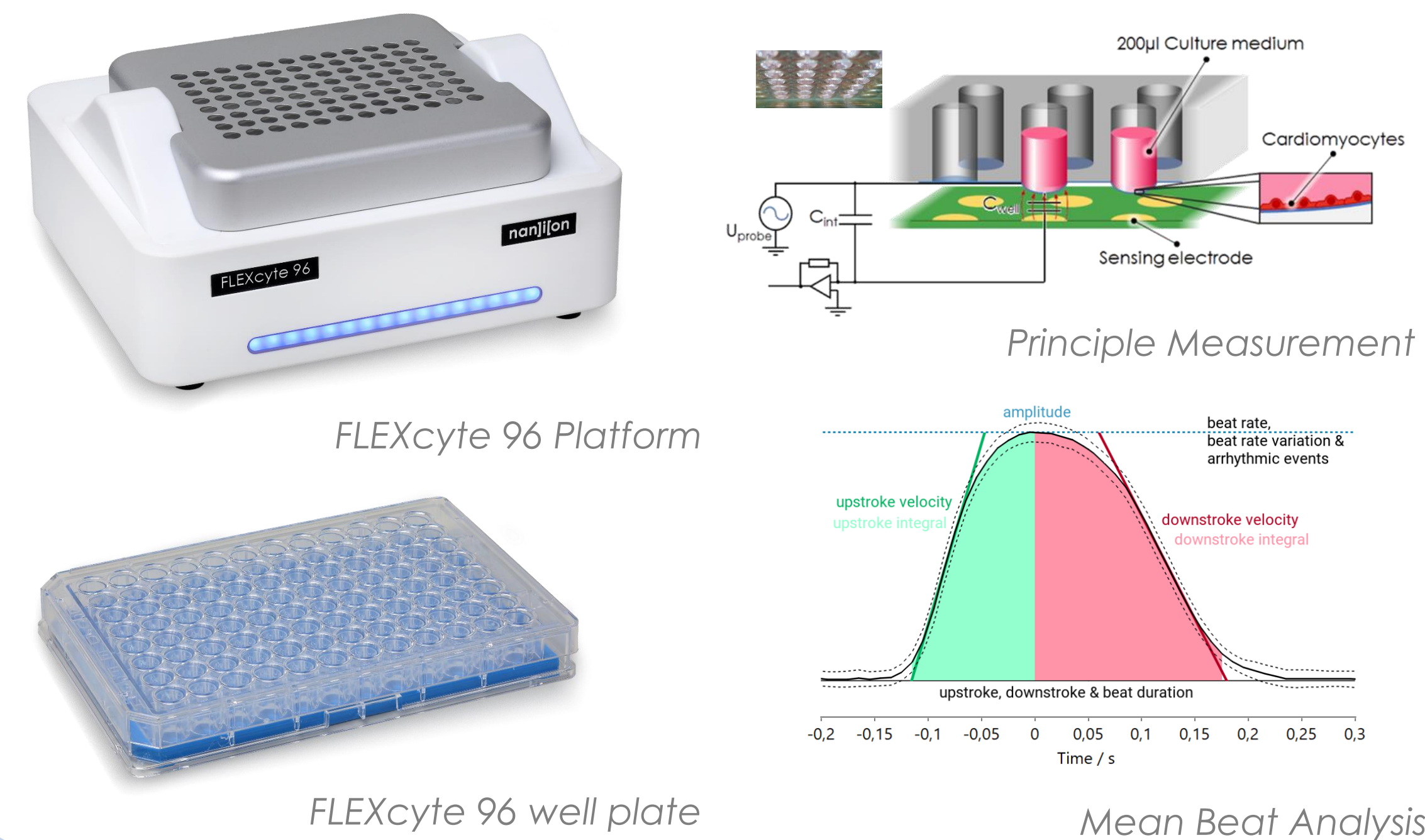
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Introduction

Despite increasing acceptance of human induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CMs) in safety pharmacology, controversy remains about the physiological relevance of existing in vitro models for their mechanical testing. We hypothesize that existing signs of immaturity of the cell models, e.g. negative inotropy upon adrenergic stimulation, result from an improper mechanical environment. We cultured hiPSC-CMs in a 96-well format on hyperelastic silicone membranes imitating their native mechanical environment, resulting in physiological responses to compound stimuli.

FLEXcyte 96 Platform

Contractility data derived from hiPSC-CMs in an environment that reflects the mechanical properties of real human cardiac tissue in a higher throughput format (FLEXcyte 96) is physiologically relevant.



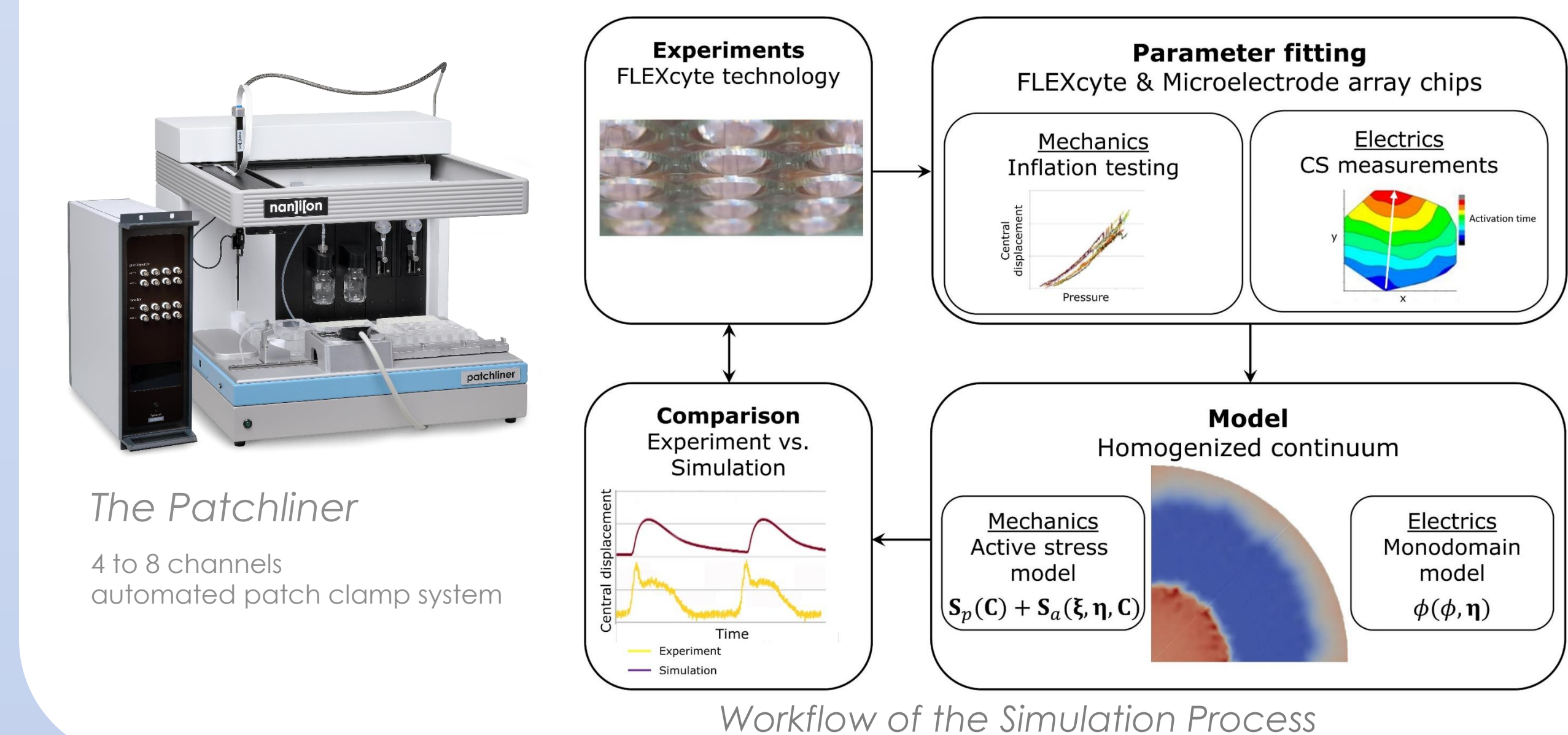
FLEXcyte 96 technology allows :

- ✓ contractility and contraction force on flexible substrate
- ✓ physiological environment & label-free measurements
- ✓ provides a reliable model for adult tissue-specific function
- ✓ Accurate detection and quantification of inotropic and chronotropic effects
- ✓ predictive recordings for early stage drug testing
- ✓ validated for hiPSC cardiomyocytes

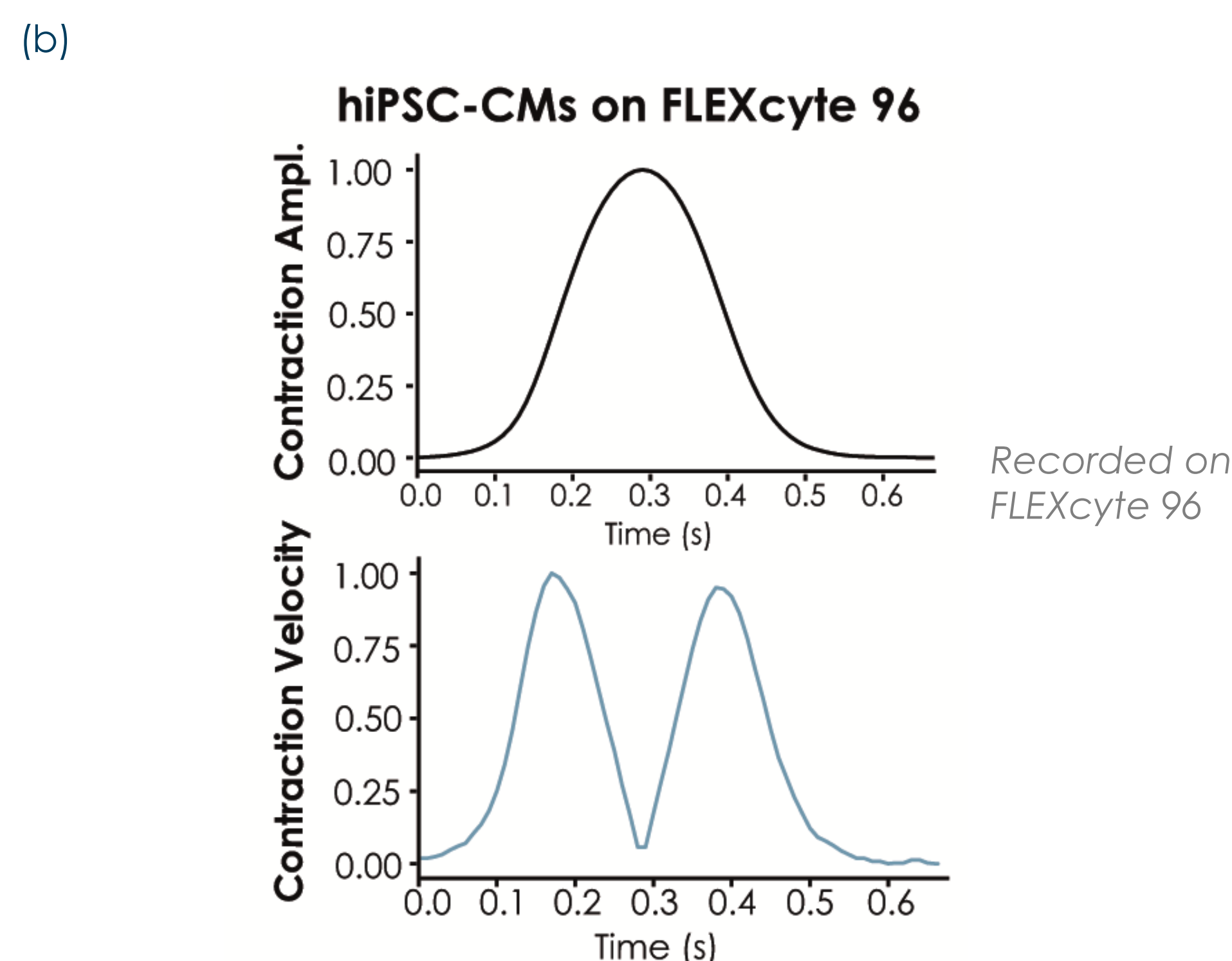
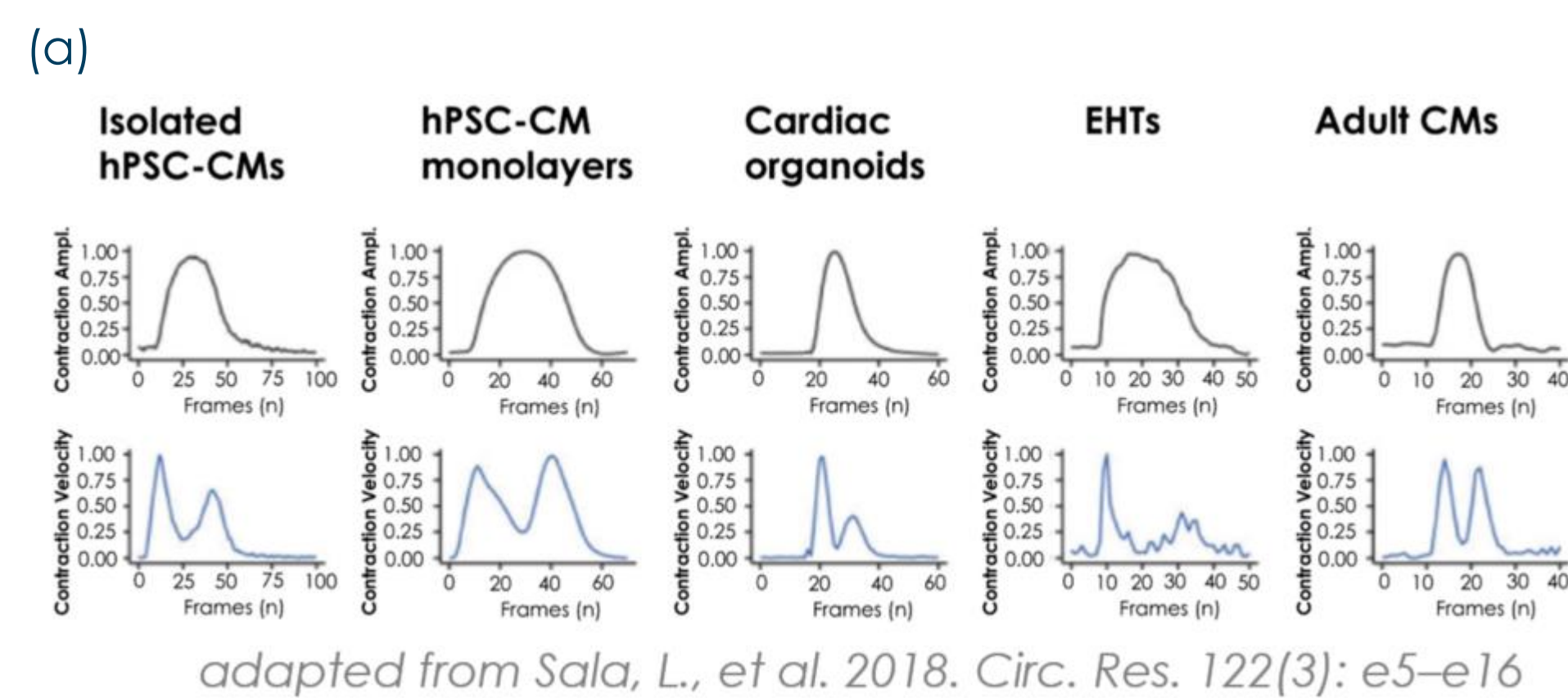


Electromechanical model simulation

The measurements were complemented with electromechanical models based on electrophysiological recordings of the used cell types. Electrophysiological properties were recorded by automated patch-clamp (Patchliner) and results integrated into the electromechanical model (Jung & Staat, GAMM-Mitt., 2019; e201900002).

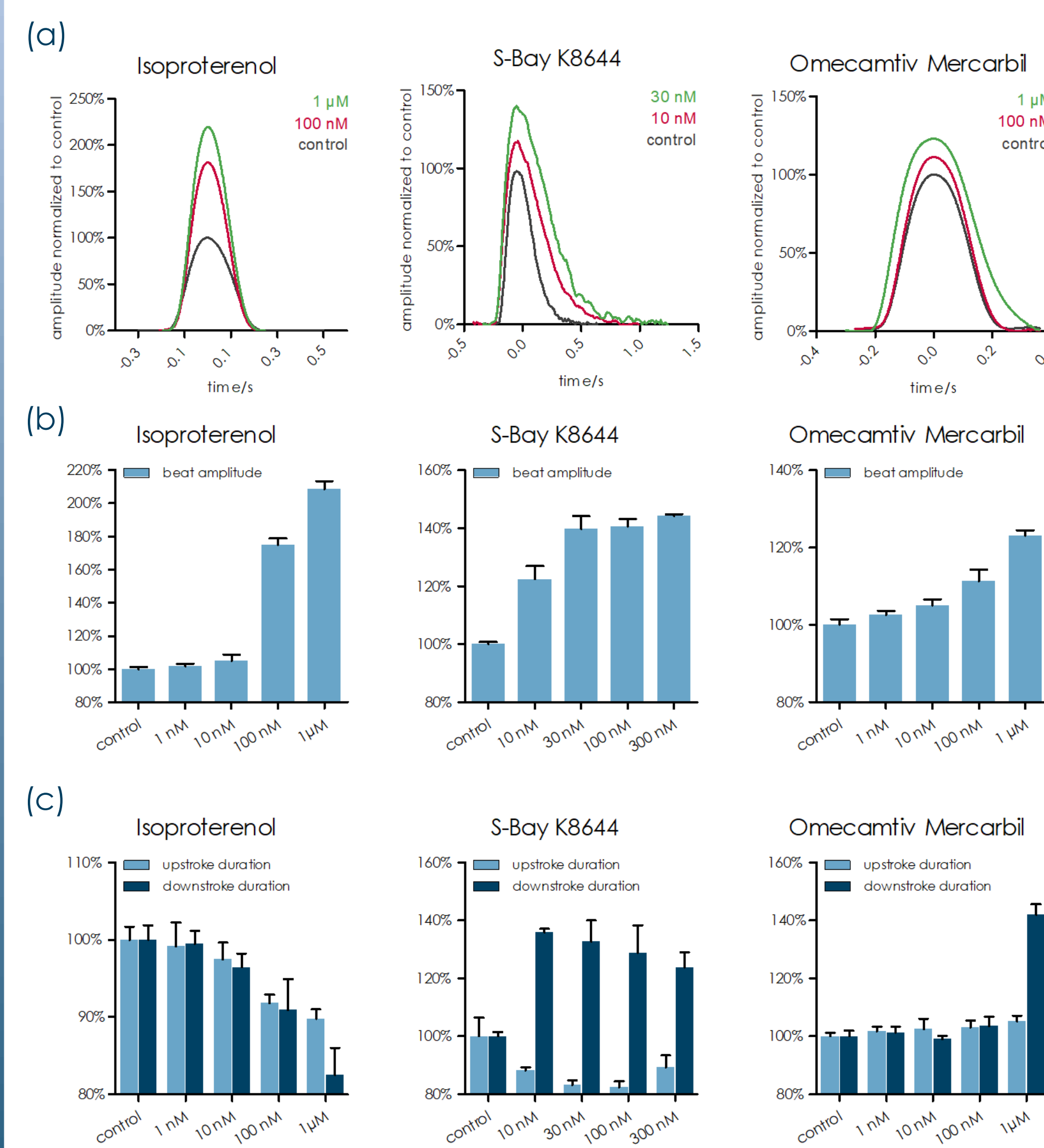


Contraction amplitude and velocity of hiPSC-CMs on the FLEXcyte 96



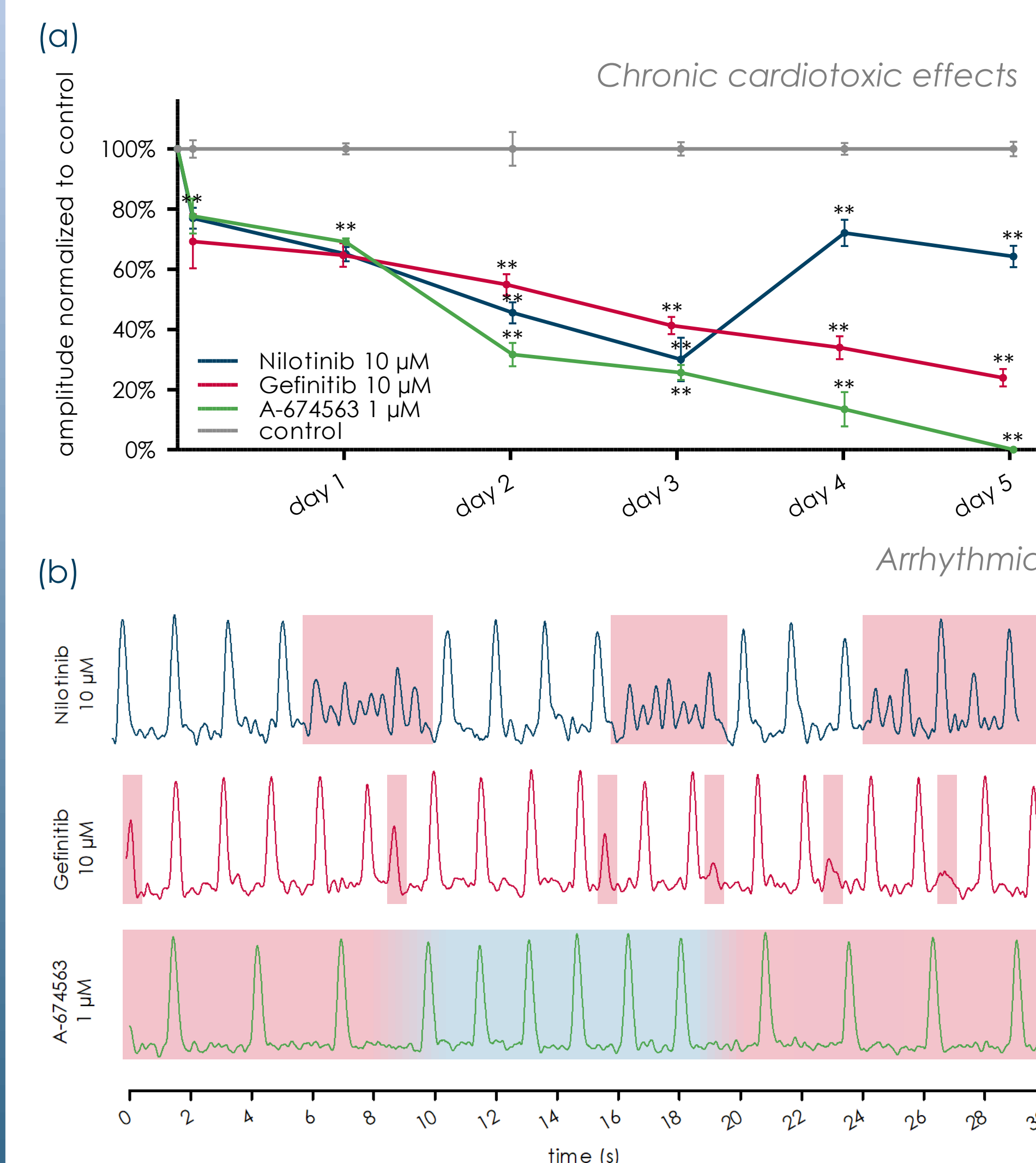
Contraction and velocity profile of hiPSC-CMs (Nexel, Cardiosight-S®) grown on the flexible substrate of the FLEXcyte 96 (b) are highly comparable to adult cardiomyocytes (a) indicating a more mature phenotype of these cells (adapted from Sala, L., et al. 2018. Circ. Res. 122(3): e5-e16).

Acute Effects of Positive Inotropic Compounds



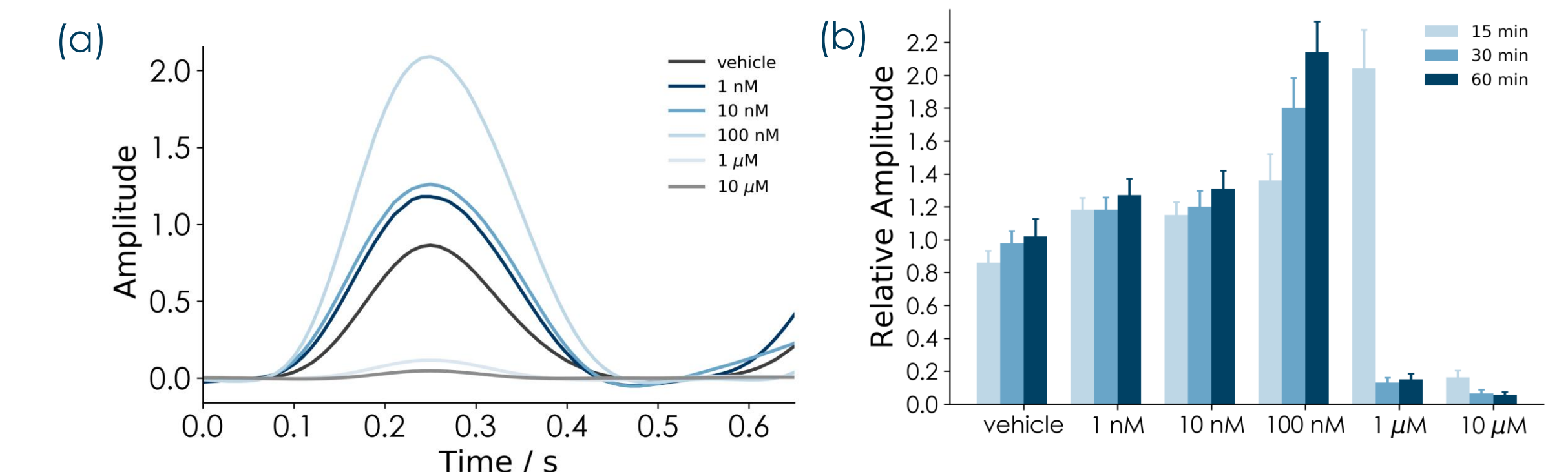
β-adrenergic agonist isoproterenol, L-type calcium channel agonist S-Bay K8644, and cardiac myosin activator omecamtiv mecarbil induced significant positive inotropic responses without additional external stimulation (a). These positive inotropic compounds induce an increase in beating amplitude (b) but show distinct effects on beating shape, upstroke and downstroke duration (c). Data recorded from hiPSC-CMs (FCDI - iCell® Cardiomyocytes² and Nexel - Cardiosight-S®). Bar graphs represent mean±SEM.

Chronic Cardiotoxicity Detection



Chronic treatment with kinase inhibitors induced cardiotoxicity during 5 day incubation (top) as well as various arrhythmia types (bottom). Kinase inhibitors displayed cardiotoxic effects on a functional level as chronic application induced significant reduction in beat amplitude of hiPSC cardiomyocytes. The system-integrated analysis detected quantifiable alterations in beating shape as well as frequency and arrhythmic events. Traces represent beating patterns after distinct kinase inhibitors were applied. Arrhythmias are marked in pink. Data recorded from hiPSC-CMs (FCDI - iCell® Cardiomyocytes²).

Acute Cardiotoxic Effects



Acute effects of digoxin on hiPSC cardiomyocytes (Nexel - Cardiosight-S®). (a) Overlay of mean beats at 60 min after application of several digoxin concentrations (1 nM – 10 µM). (b) Bar graphs indicate development of beating amplitude over 60 min for the same digoxin concentrations as in (a). Bar graphs represent mean±SEM.

Conclusion

In summary, the FLEXcyte 96 system is a reliable high throughput tool for *in vitro* cardiac contractility research, providing the user with data obtained under physiological conditions which resemble the native environment of the human heart. We show that the results obtained for both acute and chronic compound administration show consistency with the respective physiological responses in humans.

FLEXcyte 96 Whitepaper



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