



FP6 EU STREP Project



Invitroheart

**hESCreg Symposium
Berlin 18-19 January 2008**

**Carl-Fredrik Mandenius
Linköping University, Sweden**



FP6 EU STREP Project

INVITROHEART

**Reducing Animal Experimentation in Drug Testing by
Human Cardiomyocyte *In Vitro* Models Derived from Embryonic Stem Cells**



Start: 1 January 2007

Duration: 3 years

EC contribution: 2.701.611 Euro

Contractors: 9

**Coordinator: Carl-Fredrik Mandenius,
Linköping University**

CONTRACTORS

Linköping University, Sweden

Cellartis, Sweden

**Multi Channel Systems,
Germany**

Pharmacelsus, Germany

PreSens, Germany

Lundbeck, Denmark

Sahlgrenska University

Hospital, Sweden

Saarland University, Germany

ECVAM, Italy



INVITROHEART



Overall Objectives

- ***Replacing animals*** in preclinical pharmaceutical development and chemical substance toxicity testing by human cell culture systems
- ***Supporting the predictability*** of the drug discovery and development of cardiovascular pharmaceuticals by allowing more reliable and relevant testing in the preclinical phase and hinder weak lead candidates to enter clinical phases with innovative human cardiomyocyte cell systems
- ***Delivering an in vitro testing system*** with sensor methodology pertinent for *validation* in GLP/SOPs environment for cardiac safety
- ***Delivering in vitro testing systems*** with sensor methodology for chemical substance toxicity testing within REACH initiative
- The ***long-term aim*** is to significantly reduce the use of animals in drug and chemical substance testing, refine the model system under consideration and to replace the animal models currently used



INVITROHEART



Specific technology-related objectives

- **Establishing relevant hES cell derived cardiomyocytes** cultures that allow preclinical lead testing programs with higher predictability to be carried out
- Development of **real-time sensing methods** for *in vitro* model for cardiac side-effects that mimics the function and complexity of the cardiomyocyte tissue *in vivo*
- **Establishing a versatile biosensor platform** based on the developed cell lines cultivated advanced miniaturised systems with non-invasive measurement techniques for *in vitro* testing of cardiotoxicity

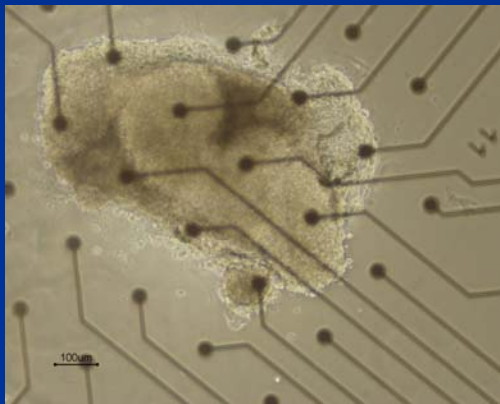


INVITROHEART



Multi Electrode Array / QT-Screen

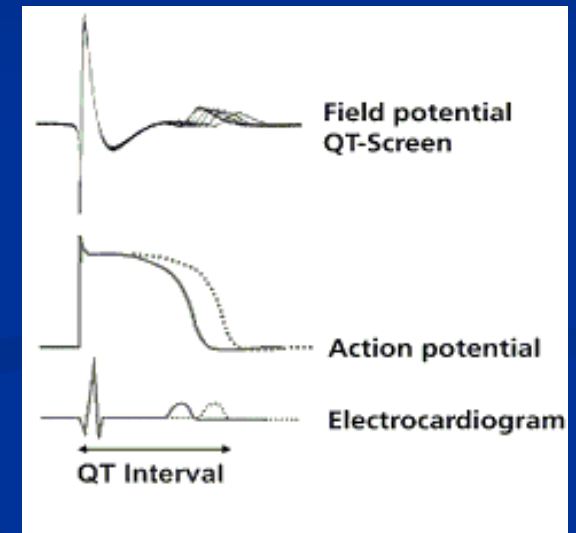
Monitoring drug-induced effects on cardiomyocyte depolarization and repolarization



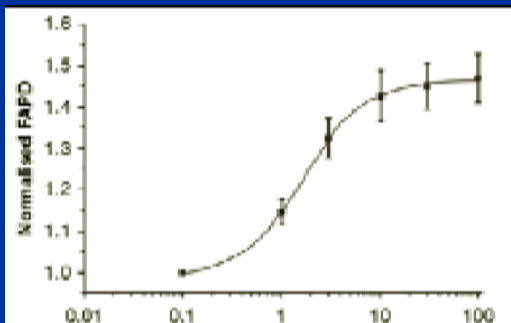
hESC-derived cardiomyocyte embryoid body on a MEA recording dish



Electrodes in a 96-well QT-Screen plate from MultiChannelSystems



Recording of the field potential during depolarization and repolarization with QT-Screen technology. Comparison with single cell action potential recording and *in vivo* ECG recording Drug-induced prolongation of repolarization illustrated with dotted line



Effect of hERG-blocker E4031 on the field potential repolarization phase of hESC-derived cardiomyocytes as compared with controls



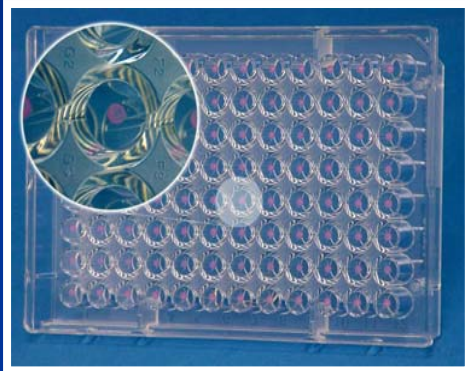
INVITROHEART



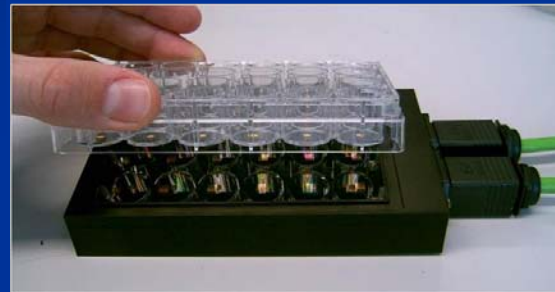
FP6 EU STREP Project

OxyPlate SensorDishReader

Monitoring of drug-induced effects on oxygen consumption for determination of LC_{50} values



96-well OxyPlate with oxygen-sensitive fluorescent dots



SensorDishReader from PreSens

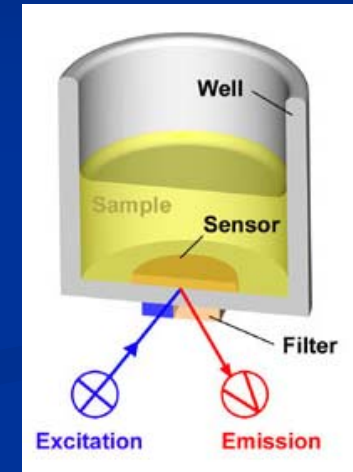
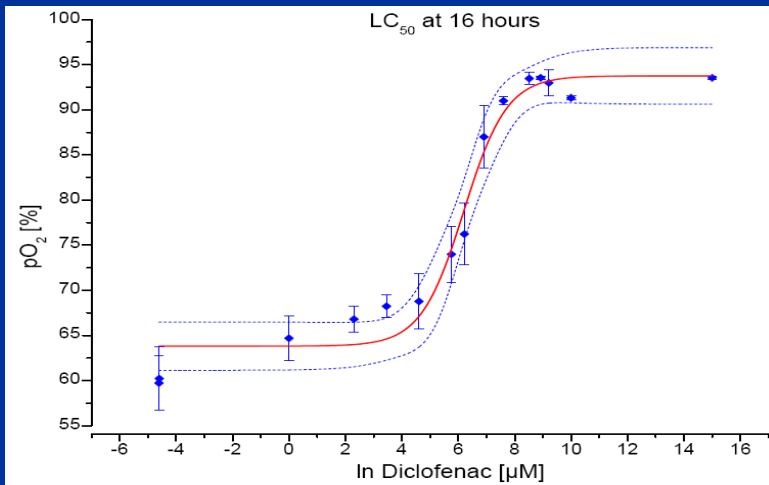


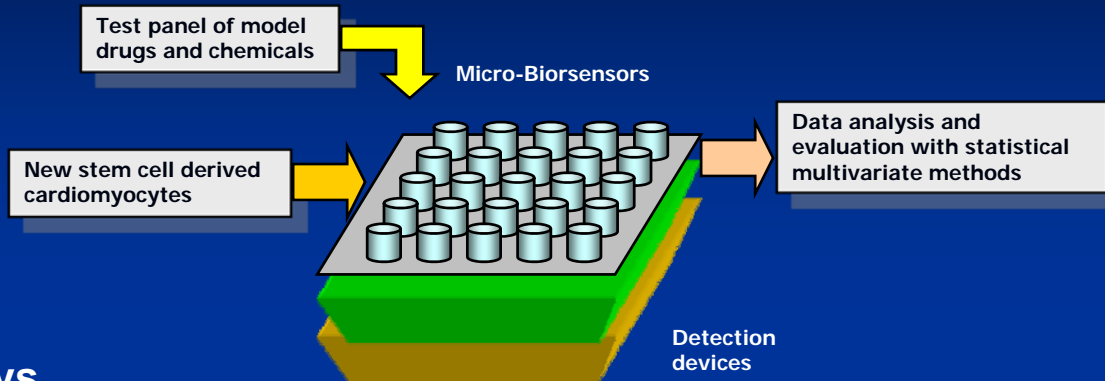
Illustration of optical monitoring of fluorescence from under each well in order to determine culture medium oxygen levels



Dose-response curve of drug-induced reduction of oxygen consumption for determination of LC_{50} value



INVITROHEART



Bioassays

- Electrophysiological recordings with Multi Electrode Arrays and QT-Screen Technologies
- Monitoring oxygen consumption with optical sensors
- Measuring drug-induced effects on the metabolome using ^{13}C -labeled substrates
- Measuring secreted factors with surface plasmon resonance technology
- Determination of cytotoxic effects and effects on membrane integrity with established methods

**Satellite meeting to
ESTIV2008 – 15th International Congress on In Vitro Toxicology
September 24, 2008
Djurönäset Conference Center, Stockholm
www.estiv2008.org**

**Development of *in vitro* test systems for hepatotoxicity and cardiotoxicity
based on human embryonic stem cell-derived cells**

SESSIONS

- Potential of stem cell-based *in vitro* test systems
- Stem cell-derived cardiomyocytes: state-of-the-art
- Stem cell-derived hepatocytes: state-of-the-art
- 3D-bioreactor test systems
- Optimization of differentiation by chemometric modeling
- Specialization of stem cells by recombinant techniques
- *In vitro* assessment of QT-prolongation
- Monitoring metabolic processes for toxicity testing
- Toxicity test methods employed by CROs
- Interindividual variability in xenobiotic metabolism

