Cell and Tissue Engineering
Cell-Based Assays
  • Controlled Cell Environment
  • Microelectrode array based assays
  • Microinjection based assays

Scaffolds for cell cultivation and implants

Surfaces for cell attachment
STRONG PARTNERING NETWORK
IN LIFE SCIENCES

University of Tampere

- **Regea**: Institute for Regenerative Medicine
- **IMT**: Institute of Medical Technology – joining BCC directly
- **Ficam**: Finnish Centre for Alternative Methods

**FinTiB**: Research Tissue Bank Finland

**SILK**: Research, Product Development and Innovation Centre of Ophthalmology

**Vactia**: Vaccine, inflammation and immune disease research and product development centre
Cell-Based Assays

Based on

- Controlled cell environment
- MicroElectrode Arrays (MEA)
- Microinjection

Especially suited to:

- Difficult-to-culture cells
- High-content screening

Seeking for public-private partnership
• Environmental control
  Perfusion, Gas exchange, Temperature, pH, pO₂ control

• Stimulation
  Mechanical, Electrical, Chemical

• Measurements and analysis
  Electrical, Electrophysiological, Chemical, Morphology,…

Research since 2000
Long-term Cell-Based Assays

- Continuous perfusion with growth medium
- Carbonation
- Control over environmental conditions in the wells
- No need for a separate incubator

Maturation of a neuronal network during the 2nd (A) and 3rd (B) weeks of culturing on MEA dish. Neuronal cells were mostly viable (green color) after 6 weeks of culturing (C). Inter-electrode distance = 200 μm (Heikkilä 2009)
We apply various sensing technologies
  • to follow changes in cell environment
  • to allow control of cell environment

Technologies include commercial measurement techniques as well as own developments.

General measured values: Temperature, pH, pO$_2$
Cell-specific measured values: Urea, gaba
We utilize the properties of elastomers to create “the lungs and the heart” of the environmental control.

Our know-how in microfluidics enables us to minimize the reagent consumption and control the dosage of tested substances (toxins, drugs).

We are able to generate a range of cell environments including hypoxia.
Controlled Cell Environment

Selected Publications

Cell-Based Assays

MEA-Based Technology

Biomimetic Active Environment for Differentiating and Maturing of Functional Neurons and Cardiomyocytes from Stem Cells

One of the largest academy projects in Finland

In co-operation with Institute for Regenerative Medicine
Signal from cardiac cell population on a 60-electrode MEA platform.

Microelectrode arrays (MEAs) or multielectrode arrays are devices that contain multiple plates or shanks through which electrical signals are obtained or delivered to or from the cells, essentially serving as interfaces that connect cells to electronic circuitry.
Cell networks can provide information on phenomena occurring in the tissue level:
- Such as arrhythmia-prone electrical loops, which are not detectable on single cell level
- Cell network can be native or engineered (such as using patterning)

Co-cultures of cell networks can provide information on humoral effects of cells.

Use of primary and human Induced Pluripotent Stem (hIPS) cells enables personalized medicine.

In-vitro models are in concordance with the EU policy to reduce, refine and replace animal testing.
MEA-Based Platforms
Pharmacologic modulation of hESC-derived neuronal network activity

Addition of the pharmaceuticals and recording of the network activities:
(A) Baseline activity
(B) Activity was partly suppressed by CNQX
(C) CNQX and D-AP5 together blocked all activity
(D) After a washout, activity reappeared
(E) GABA inhibited all activity
(F) Activity did not return after a washout
(G) Addition of bicuculline restored the activity to a higher level than at the baseline (A). (Note the different z-axis scale in G.)
(H) Raster plot of the bicuculline-induced synchronous activity.

(Ref. Heikkilä 2009)
Next, we aim to further develop current 1-well system into a high-throughput high-content system.

- **Current**: 1 well system with electrodes/sensors and microfluidics
  - Well diameter of 5 cm (small Petri dish size)
  - [www.tut.fi/bme/stemfunc](http://www.tut.fi/bme/stemfunc)

- **Next step**: Minuaturized 1 well system with multilayered structure
  - Well diameter of 1,5 cm (96-well plate well size)

- **Proof-of-concept**: 8-well system featuring the technology
  - 8 wells, each 1,5 cm in diameter

- **Final**: 96-wells / 256 array

Tested and developed with stem cell derived cardiomyocytes and neural cell cultures.
• Lahti A. et al. “iPS cells derived from a LQTS patient carrying a mutation in hERG gene differentiate to cardiomyocytes and exhibit the diseased phenotype in cell culture conditions”, manuscript, 2009
• Mari Pekkanen-Mattila et al. “Substantial Variation In The Cardiac Differentiation Of Human Embryonic Stem Cell Lines Derived And Propagated Under The Same Conditions ? A Comparison Of Multiple Cell Lines,”Annals of Medicine, in press.
• Kujala et al. ”Analyzing stem cell-derived cardiomyocytes with MEA platform” Tissue Engineering Symposium, 2009
CELL AND TISSUE ENGINEERING
Cell-Based Assays
Microinjection-Based Technology

“Integrated Smart Microinstruments for Automated Cell Manipulation and Analysis”

Research since 2000
Our goal is to build a platform capable of analyzing living cells on a single-cell level.

State-of-the-art devices
Separate instruments for measurements from the same population.

Our platform
One instrument for SIMULTANEOUS measurements and stimulation.

Micro injection
Visual tracking
Atomic Force Microscope
Micro Electrode Array

Electric Stimulation and Meas.
Force/Adhesion Meas.

Integrated System
Microinjection-Based Platforms

Our system can bring several tools to work SIMULTANEOUSLY

**Chemical** stimulation
(microinjection including electrical and mechanical measurement)

Cell isolation for gene expression analysis (aspiration and electrical measurement)

Electrical stimulation

Electrical measurement

Mechanical stimulation and measurement

Cell tracking

Biosensing Competence Centre  www.biosensing.fi
Our platform is interesting especially for the following studies

- Rapid automatic microinjections of cDNA, siRNA into living neural cells
  - We aim at throughput of 600 cells / hour
- Cell-to-cell stimulation and measurement studies
  - Followed by gene expression of stimulated / connected cells
- Injecting protein mass from primary cells into stem cells
- Cell-to-substrate adhesion studies
Why *capillary microinjection*?

Unspecific incorporation (electroporation, viral vectors) of various compounds to whole cell population at once

⇒ Incorporation of compounds to specific selected cells.

In methods such as electroporation, the delivered compounds are limited in size and chemical composition.

⇒ Microinjection does not limit the choice of compounds.

Capillary microinjection as a method has the highest potential for control over the injected volume.

A technique for the delivery of small volumes of compounds into suspended or adherent cells.
Why automated capillary microinjection?

Only a few skilled experts in the world are able to inject primary and especially primary neural cells.

- With a fully automated system, the success rate is independent from the level of experience of the user.
- Unprecedented repeatability.

State-of-the-art systems (= semi-automatic) achieve the throughput of 200 of cell injections per hour with an unreliable success rate due to missing contact detection and capillary condition diagnostics. State-of-the-art throughput for microinjections of primary neural cells is in tens of cell injections per hour depending on the skills of the operator/researcher.

- With our technology, we aim at throughput of 600 cells/hour (injection every 5 seconds) with the success rate of 80 %.
Automatic cell contact detection

State-of-the-art bottlenecks
Injection plane set for whole population. Contact of the capillary and the cell is not detected.

Our system facilitates automatic contact detection for each cell. Enabling technologies include visual, electrical and mechanical methods.

Part of cells remains uninjected

Proof-of-concept already achieved by electrical means.

Biosensing Competence Centre

www.biosensing.fl
Automatic capillary fault diagnostics

State-of-the-art bottlenecks:
Capillary condition (clogging of the tip with cell membrane or debris or breakage of the tip of the capillary) cannot be diagnosed. Undiagnosed faults result in low success rate and unreliable results. Injection volume cannot be controlled.

➔ Our system can distinguish between the clogging (automatic cleaning performed) and the breakage of the capillary (possibly automatic capillary change).
• Kovanen, K. et al. "Combined Cell Survival and Injection Success Rate in Microinjection of Living Adherent Cells", the 3rd European Medical and Biological Engineering Conference EMBEC'05, 2005.
Scaffolds for cell cultivation and implants

- 3D scaffolds using biocompatible materials for cell growth
- Biodegradable and bioactive implants
- (Bio)material testing
  - Standardized testing of biodegradation (e.g. ISO 13485)
Scaffolds
Selected Publications


Polystyrene surfaces have been modified to both reject and promote culturing of human embryonic stem cell (hESC)-derived neuronal cells.

Cells show a selective growth on defined part of the surface whereas no growth can be observed on parts designed to reject the growth.


In co-operation with Institute for Regenerative Medicine
Highly Specific Layers for Molecular Recognition

• Polymer:
  • N-[tris(hydroxy-methyl)methyl]acrylamide, lipoic acid conjugate, pTHMMAA

Biosensing Competence Centre
www.biosensing.fi

Visiting address:
Hermia 1, Hermiankatu 6-8

Postal address:
P.O.Box 692, 33101 Tampere
Finland

Hannu Helle
Chief Operating Officer, M.Sc.
hannu.helle@biosensing.fi
+358 40 849 0920

Johana Kuncová-Kallio
Chief Business Development, Ph.D.
johana.kuncova-kallio@biosensing.fi
+358 40 849 0015