DELIVERABLE REPORT

Grant Agreement Number: 812954

H2020-MSCA-ITN-2018

EUROoC

Interdisciplinary training network for advancing

Organ-on-a-chip technology in Europe

Deliverable 3.3: Slides and Directions for Public Lectures

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Project coordinator organisation name	Fraunhofer
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Dissemination Level of Report

PU	**Public**	**X**
PP	Restricted to other program participants (including the Commission Services)	
RE	Restricted to a group specified by the consortium (including the Commission Services)	
CO	Confidential, only for members of the consortium (including the Commission Services)	

Abstract

Deliverable 3.3 of the EUROoC project comprises 36 presentation slides developed for public use. They were designed to introduce the topic, present the participating institutions and organisations, to explain the different objectives and to provide deeper insights into the current state of research. As they address a broad audience, they will be used for classroom lectures for pupils, public evening lectures or non-specialist science events equally.

Moreover, they will be featured on the website and can be requested by schools, science museums, science cafes, public science festivals or similar events organised by stakeholders in the vicinity of the network partner locations. Respective PIs or ESRs at those locations will present the lectures and engage with the public.

Table of Contents

1 Lecture Slides

Slide Deck for Public Outreach

Deliverable 3.3

MSCA-ITN-ETN: EUROoC (GA-No. 812954)

Organ-on-a-chip – Definition

2

"a fit-for-purpose microfluidic device, containing living engineered organ substructures in a controlled microenvironment, that recapitulates one or more aspects of the organ's dynamics, functionality and (patho)physiological response in vivo *under real-time monitoring"*

As defined by the ORCHID Vision Workshop (Mastrangeli M. et al. Organ-on-Chip In Development: Towards a roadmap for Organs-on-Chip, doi:10.20944/preprints201903.0031.v1)

see also Mastrangeli M. et al. Building blocks for a European Organ-on-Chip roadmap. ALTEX. 2019;36(3):481–492.

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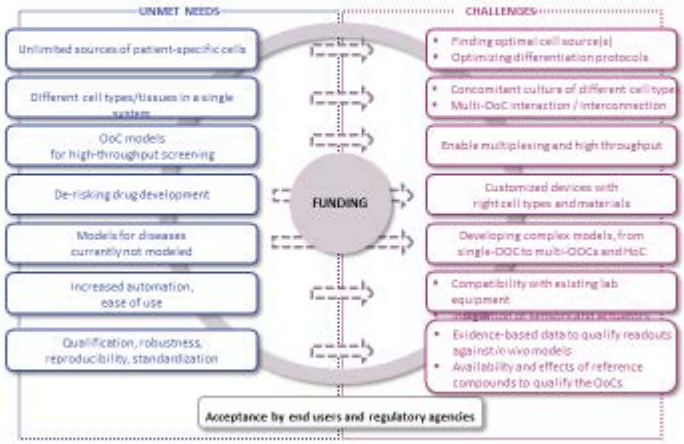
Organ-on-a-chip – Current state



Organ-on-a-chip – Crossing the Valley of Death

Academics
- Functional validation
- Mechanistic applications
- Some compounds tested
- Manual handling
- Proof of concept in developers lab

Support
- Joint projects
- Early regulator involvement
- Tissue Chip Testing Centers

Industry
- Large reference datasets
- Accredited systems
- High TRL
- Automated handling
- Inter/Intra-laboratory reproducibility

Issues
- Extensive screening not publishable / not interest of academics
- Missing funding mechanisms

Organ-on-a-chip – Working Together in an European Society

5

- launched at the International Organ-on-Chip Symposium on Nov. 8th
  - Development and coordination of Organ-on-Chip research in Europe
  - Annual EUROoC Conferences (2019 Graz, Austria; 2020 Upsala, Sweden)
  - Development Provide opportunities to share and advance knowledge and expertise in this field
  - Founding board: Christine Mummery (chair), Peter Loskill (vice-chair), Janny van den Eijnden, Albert van den Berg
  - Website: www.eurooc-society.eu/

EUROoCS
EUROPEAN ORGAN-ON-CHIP SOCIETY

Training future leaders in Organ-on-a-chip Research

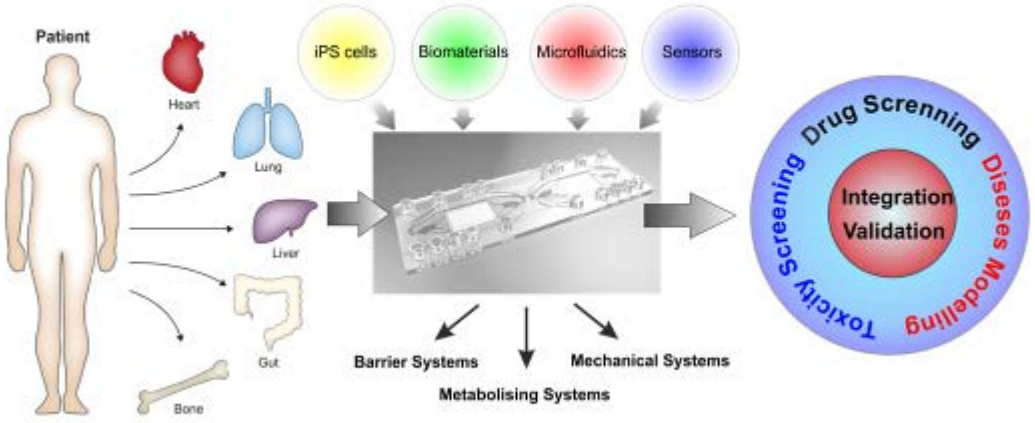
6

To support innovative research projects, which together target the development of advanced Organ-on-a-chip systems with higher physiological significance that go beyond the culture of monolayers on inert membranes and that directly integrate endpoint analysis

To train the next generation of interdisciplinary scientists to be adept in all aspects of Organ-on-a-chip development and utilization

To create a trans-European network of interdisciplinary specialists working on different aspects of Organ-on-a-chip development from academic institutions, industrial infrastructure providers, and regulatory agencies

Training future leaders in Organ-on-a-chip Research



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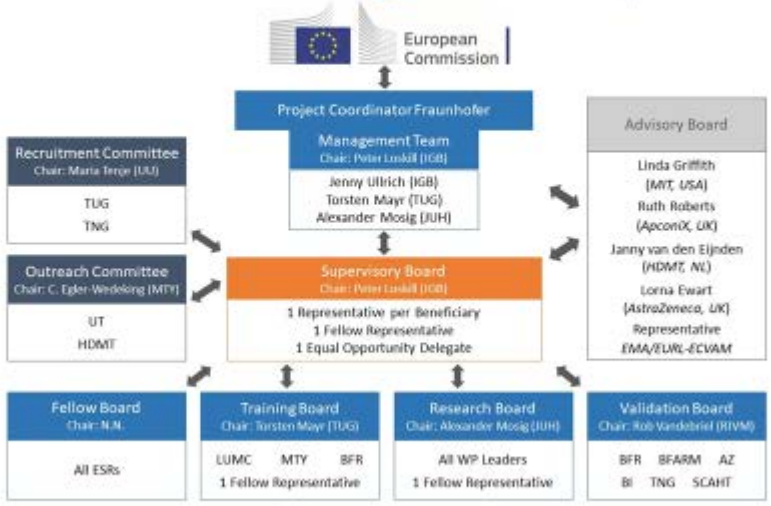
Training future leaders in Organ-on-a-chip Research



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Training future leaders in Organ-on-a-chip Research

9

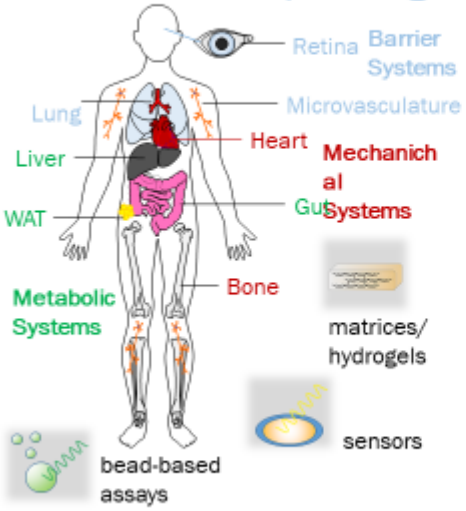
Beneficiaries

- Fraunhofer (**IGB**), Germany – *Peter Loskill & Jenny Ulrich*
- Universitaetsklinikum Jena (**JUH**), Germany – *Alexander Mosig*
- Technische Universitaet Graz (**TUG**), Austria – *Torsten Mayr*
- Universitaet Bern (**UB**), Switzerland – *Olivier Guenat*
- Uppsala Universitet (**UU**), Sweden – *Maria Tenje*
- Universiteit Twente (**UT**), Netherlands – *Andries van der Meer*
- Akademisch Ziekenhuis Leiden (**LUMC**), Netherlands – *Christine Mummery*
- Universite du Luxembourg (**UL**), Luxembourg – *Paul Wilmes*
- Ecole Polytechnique Federale de Lausanne (**EPFL**), Switzerland – *Matthias Lütolf*
- Bundesinstitut fuer Risikobewertung (**BFR**), Germany – *Marion Schneider & Andreas Haase*
- Mitenyi Biotec BV & CO KG (**MTY**), Germany – *Dominik Eckardt*

Partners

- Pyro Science GmbH (**PYR**), Germany – *Roland Thar*
- Rijksinstituut voor Volksgezondheid en Milieu (**RIVM**), Netherlands – *Rob Vandebriel*
- UPM - The Biofore Company (**UPM**), Finland – *Pia Nilsson*
- Transgene SA (**TNG**), France – *Jean-Marc Balloul*
- Boehringer Ingelheim Pharma GmbH & Co. KG (**BI**), Germany – *Stefan Kauschke*
- hDMT (**HDMT**), Netherlands – *Janny van den Eijnden-van Raaij*
- Bundesinstitut für Arzneimittel & Medizinprodukte (**BFARM**), Germany – *Susanne Brendler-Schwaab*
- AstraZeneca UK Ltd. (**AZ**), UK – *Lorna Ewart*
- Universitaet Basel (**SCAHT**), Switzerland – *Martin Wilks*
- Eberhard Karls Universitaet Tuebingen (**EKUT**), Germany – *Katja Schenke-Layland*
- AlveoliX Ag (**AlveoliX**), Switzerland – *Janick Stucki*
- Institut National de la Sante et de la Recherche Medicale (**INSERM**), France – *Maxime Mahe*
- Technische Universitaet Berlin (**TUB**), Germany – *Peter Neubauer*

EUROoC workpackages



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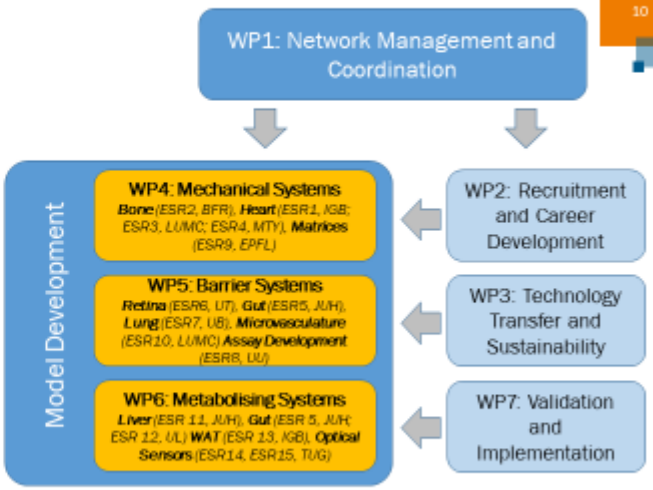

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The value of OoC for regulatory safety assessment

11

Where and how may OoC take a role in the transition to a more predictive, animal-free safety assessment for regulatory purposes?

Ten organs of priority for OoC development for regulatory use have been identified.

For lung, skin, liver, kidney, heart, and intestine, OoC are at rather advanced stages of development, such that involvement of regulators becomes of value in the optimization towards fitness-for-purpose of these methods.

For testis, spleen, brain, and stomach, OoC are more premature, if existing at all. Developmental work on OoC for these organs is expected to stay in the academic arena for some time.

We recommend the development of OoC to go together with the development of Adverse Outcome Pathways and combining them with other methods into integrated testing strategies.

Regular interactions in multi-stakeholder workshops on application of animal-free innovations such as OoC will be beneficial.

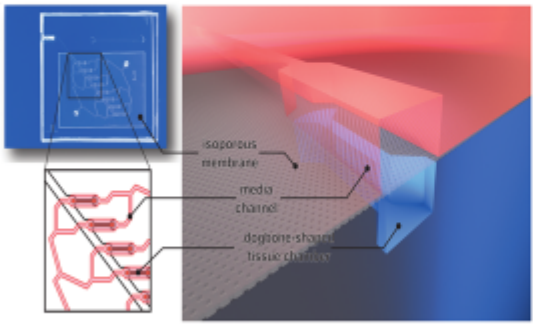
Heringa et al. ALTEX 2019

WP4 – ESR1: Heart-on-chip

12

Parallelisable microfluidic Heart-on-a-chip systems with integrated sensing capability to monitor maturation and functionality of cardiac microtissues.

Chip Design



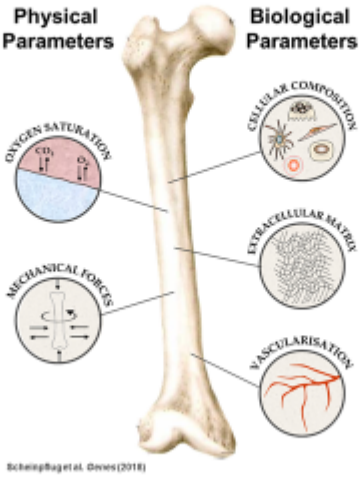
Milestones

- Defined cell composition (hiPSC-derived Cardiomyocytes/Fibroblasts).
- Integration of hydrogels.
- On-chip tissue maturation assessment.
- Set-up a toolbox of non-invasive read-out technologies (pO2 sensors, RAMAN, FLIM).

WP4 – ESR2: Bone-on-chip

13

- Hypoxic gradients (0.5–7% O_2) that also determine cell function (e.g. heamatopoetic stem cell niche)
- Subjected to mechanical load that is a regulator in bone remodelling through the balanced interaction of osteoclasts (bone resorption) and osteoblasts (bone formation)

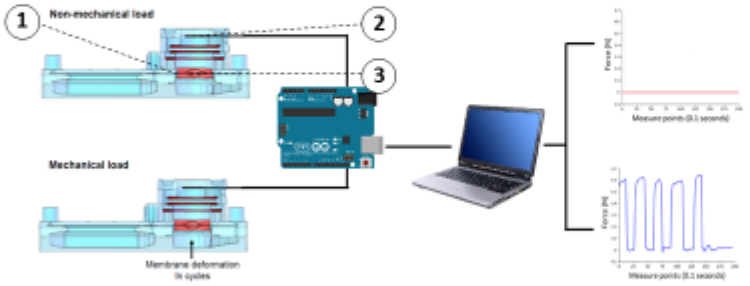


- Harbours a multitude of different cells including mesenchymal stromal cells, heamatopoetic stem cells, osteoblasts, osteoclasts etc.
- ECM is a composite material consisting of organic (90% Col I) and inorganic components (Hydroxyapatite: ([Ca3(PO4)2]Ca(OH)2))
- Highly vascularized, varying degrees of vascularisation within different tissue compartments (e.g. bone marrow vs. periosteum)

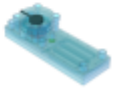
WP4 – ESR2: Bone-on-chip

14

Aim: To develop a OoC system that allows for the application and quantification of mechanical load to a 3D organoid to study matrix mineralization and remodelling, thereby enabling a physiologic model as an alternative to animal testing (reduce and replace) in the context of bone biology.

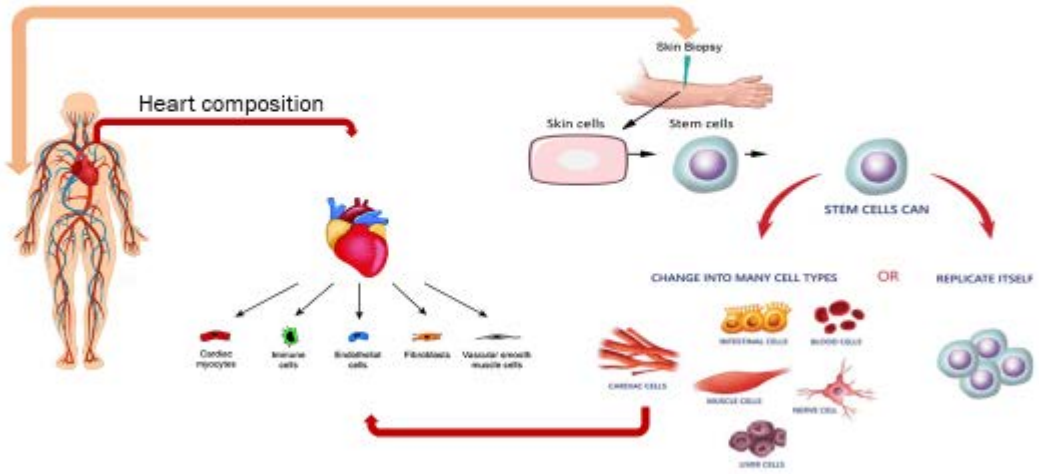


- Mechanical load is applied pneumatically via a flexible membrane that is situated below the organoids' culture compartment and covers a small chamber than can be pressurized (1)
- A sensor for force determination is incorporated in the devices' lid on the opposite site of the pressure chamber (2)
- The organoid (in red) is situated between pressure chambe (3)



WP4 – ESR3: Heart-on-chip

15



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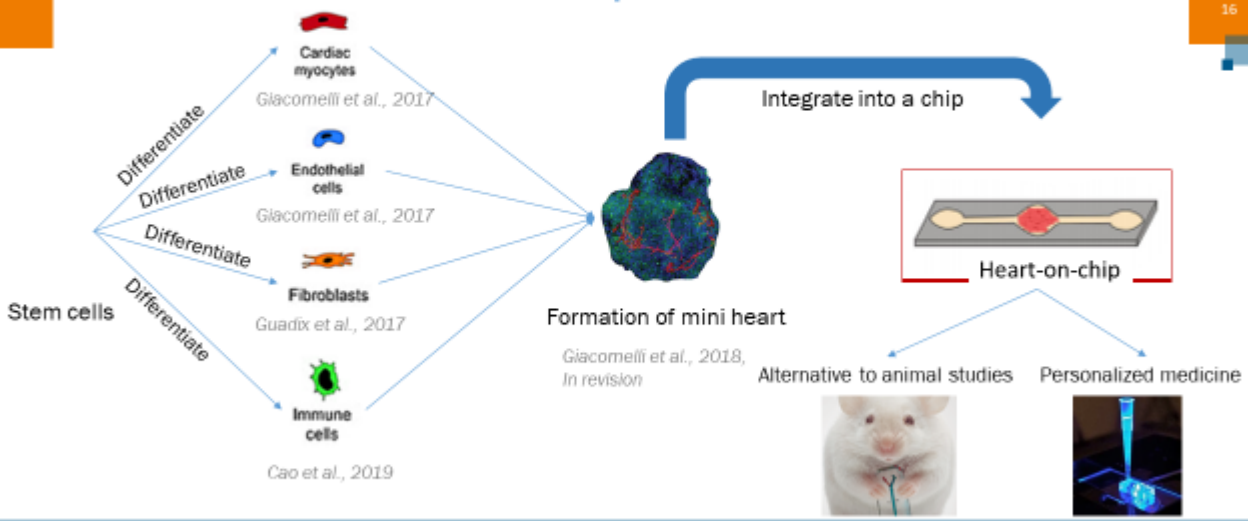
WP4 – ESR3: Heart-on-chip

16



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WP4 - ESR4: Heart-on-chip

17

Limited analysis of cell engraftment after transplantation into human tissue

?

Lack of human *in vitro* test systems

Development of a microfluidic-based heart-on-a-chip model as a device for modelling cellular therapy approaches

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WP4 - ESR4: Heart-on-chip

18

TISSUE
Protocol development for heart microtissue functioning for non-invasive analysis set up
2019/20

CHIP
Transfer of differentiation protocol and analytics to „on-chip" systems
2020/21

PROOF OF CONCEPT
Evaluation of the heart-on-a-chip system as a *in vitro* assay for cellular therapy products
2021/22

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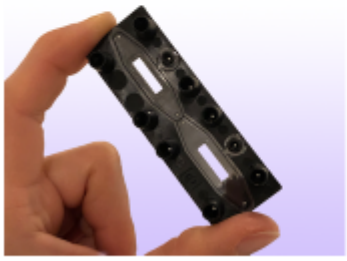
WP5 – ESR5: Gut-on-chip

19

Current limitations to investigate:

- Intestinal diseases
- Host-microbiome interactions
- Person-specific reactions to medicine



Aim of PhD project:

To create an immune-responsive and physiologically relevant intestinal model for personalised microbiome studies using a gut-on-chip platform

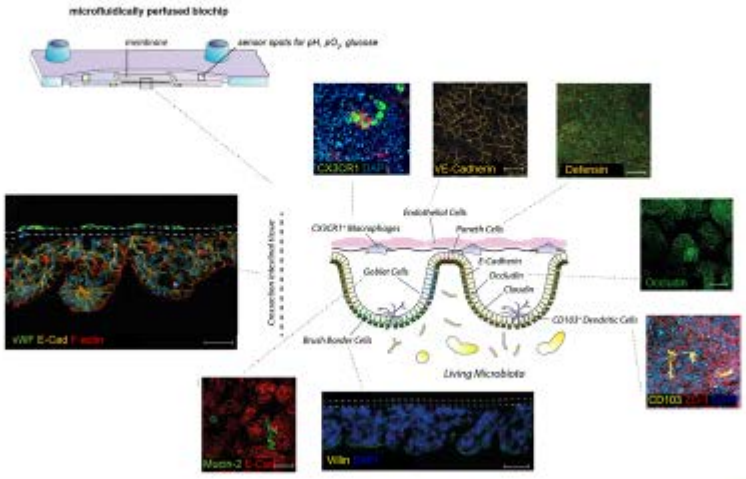
Objectives:

1. Establish a more physiologically relevant model of the gut
2. Create an hIPSC-based intestinal model
3. Study host-microbiome interactions

MSCA-ITN-ETN: EUROoC (GA-No. 812954)

WP5 – ESR5: Gut-on-chip

20



MSCA-ITN-ETN: EUROoC (GA-No. 812954)

WP5 – ESR5: Gut-on-chip

21

- **How**

Incorporate *in vivo* knowledge of physiologically relevant chemical and biophysical cues into the gut-on-chip model

Integrate the gut microbiome into this model and carry out studies of host-microbiome interactions

Implement and develop a physiologically relevant hIPSC model to enable future personalized studies

- **Future perspective**

Empower physiologically relevant *in vitro* investigations of the gut on a cellular level

Enable relevant *in vitro* studies of the gut and host-microbiome interactions

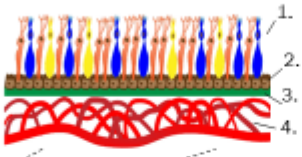
WP5 – ESR6: Retina-on-chip

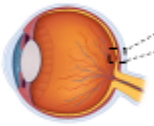
22

outer Blood-Retinal Barrier*

→ Essential for normal visual functions:

- Supply O_2 & Nutrients for neuronal Retina
- Disposal of waste products





1. Neuronal Retina
2. Retinal Pigmented Epithelium(RPE)*
3. Bruch's Membrane*
4. Choroid (vascular layer)*

- Vision loss and blindness affect million of people
- Blood-Retinal barrier involved in pathophysiology of various retinal diseases

But: disease mechanism not fully understood!

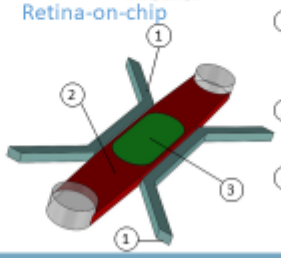
Problem: Lack of suitable animal models & limitations of current cell models to study disease mechanism

Aim: developing a 3D model of oBRB as a Organ-on-chip with integrated oxygen sensing

- Drug testing
- Retinal disease modelling

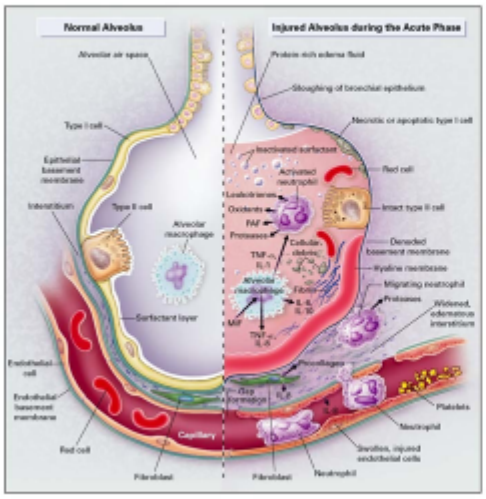
→ Find new treatment strategies against retinal diseases

Retina-on-chip



1. Microfluidic channels connected to medium reservoirs/pumps
   - to supply cells in inner chamber with O_2 & Nutrients
2. Open inner chamber filled with hydrogel and endothelial cells
   - Vascular network (Choroid)
3. RPE cells seeded on top of hydrogel in open inner chamber
   - Epithelium & Bruch's membrane

WP5- ESR7: Lung-on-chip



Acute Lung Injury (ALI)

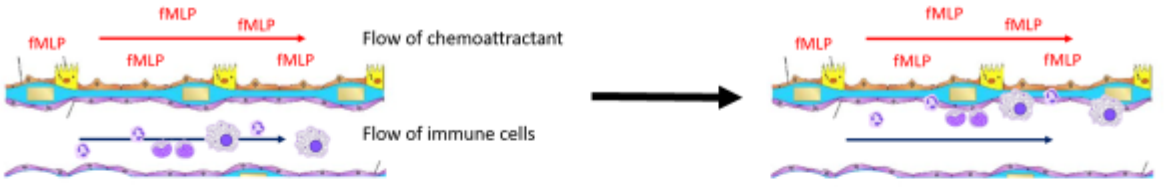
- Severe injury of the lung
  - Sepsis, infection, intubation, etc.
- 40-60% mortality
- Current treatment not effective
- Mechanism: barrier disruption and infiltration of immune cells
- **Aim: mimic ALI on chip**

Lorraine and Matthey, 2000

WP5- ESR7: Lung-on-chip

Aim: investigation of ALI on chip

Development of a perfusable lung-on-chip to study barrier disruption and immune cell migration



Perfusion of immune cells and chemoattractant

Immune cells

Immune cells adhere and transmigrate towards chemoattractant

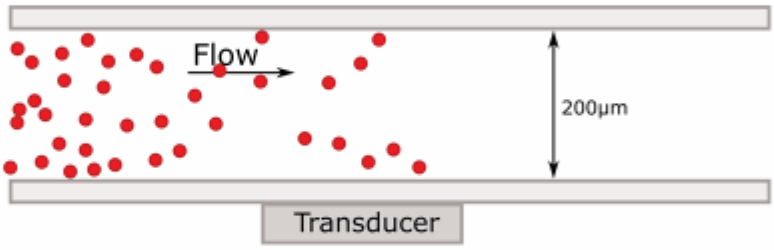
→ detection of expressed cytokines

WP5-ESR8: Assay Development

25

Acoustic trapping

- Particles can be trapped when a soundwave has the right pitch to fit inside a channel, like you see in the picture to the right.
- These particles can be designed to attach to proteins or molecules that would otherwise be to small to catch. This will concentrate the proteins and caught proteins will give a flourecent signal allowing a measurement.



Schematic drawing of the trapping phenomenon. [1]

[1] M. Tenje *et al.*, "Acoustic trapping as a generic non-contact incubation site for multiplex bead-based assays," *Anal. Chim. Acta*, vol. 853, no. 1, pp. 682–688, 2015.

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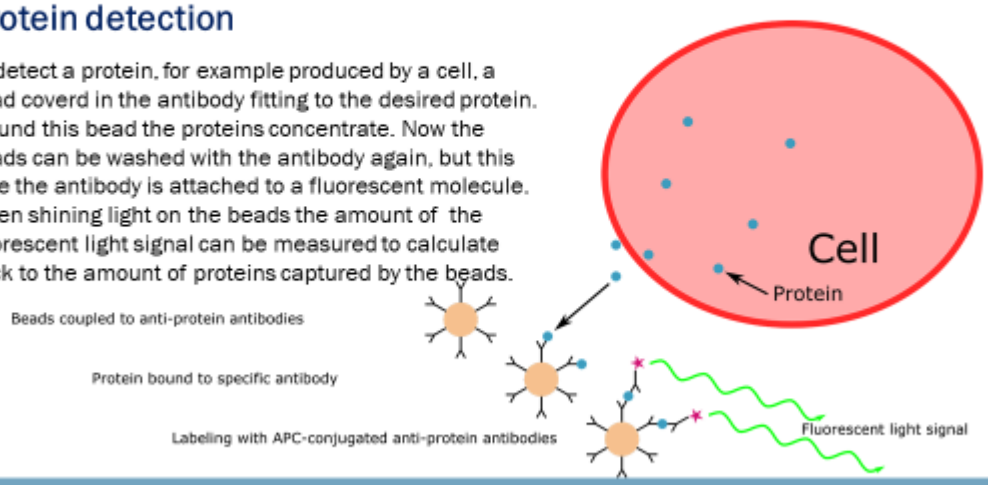
WP5-ESR8: Assay Development

26

Protein detection

To detect a protein, for example produced by a cell, a bead coverd in the antibody fitting to the desired protein. Around this bead the proteins concentrate. Now the beads can be washed with the antibody again, but this time the antibody is attached to a fluorescent molecule. When shining light on the beads the amount of the fluorescent light signal can be measured to calculate back to the amount of proteins captured by the beads.



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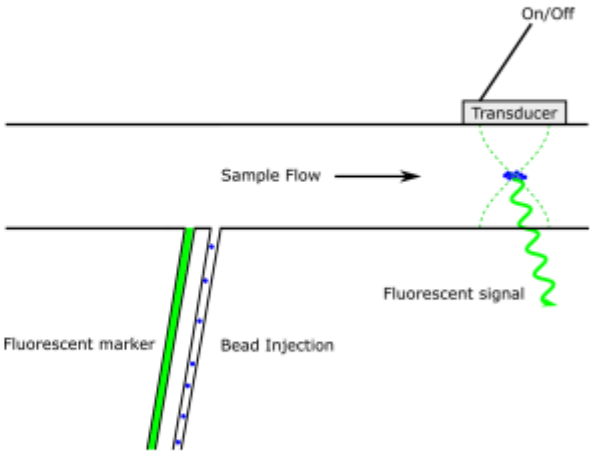
WP5-ESR8: Assay Development

27

Lab-on-Chip design

- The shown chip design will allow beads to be captured and exposed to the fluid containing the proteins from the cells
- After exposure the fluorscent markers can be injected and the fluorescent signal can be measured
- The goal is to detect the protein concentration in 10 minute intervalls, providing more insight on the cell behaviour during experiments, especially for Organ-on-Chip setups.
- The chip will be tested in collaboration with the group working on the Lung-on-Chip



WP4-ESR9: Defined Matrices

28

Motivation: Cells can be guided externally to make up complex organ-like systems

Healthy and Diseased

Better fitting than animal models

Stem Cells

Highly flexible cells that can produce multiple cell types

Defined "lab-homes"

Architecture: 3D
Furniture: ECM proteins
Supplies: Growth factors, hormones
Seasons: dynamicity (4D)

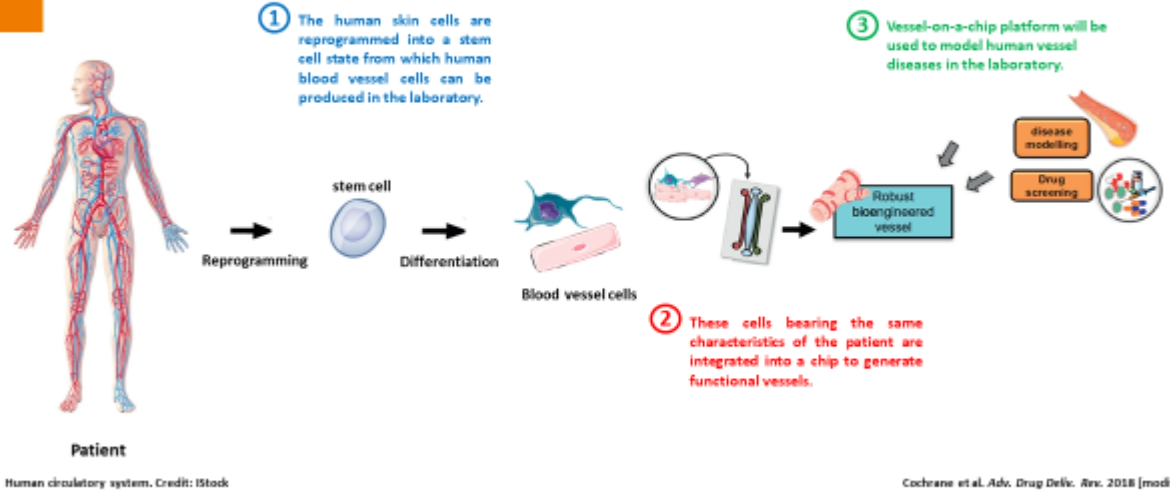
Miniature organs on-Chip

Different cell types
Roads: Vascular network
Unique to the donor

Synthetic "homes" and "villages" to grow mini-organs

WP4-ESR10: Microvasculature-on-chip

29



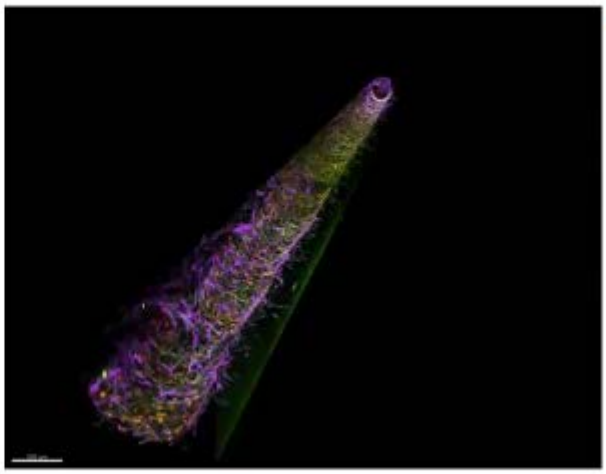
Human circulatory system. Credit: IStock

Cochrane et al. *Adv. Drug Deliv. Rev.* 2018 [modified]

WP4-ESR10: Microvasculature-on-chip

30

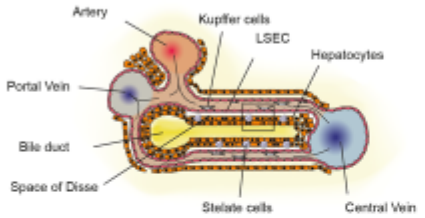
Let's flow through a vessel-on-a chip formed by human cells.



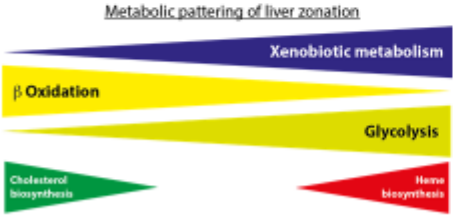
De Graaf et al. APL Bioeng. 2019

WP6 – ESR11: Liver-on-chip

31



Metabolic pattering of liver zonation



Aim of PhD project

To create an immune-responsive and physiologically relevant model of liver zonation able to recreate metabolic patterning

Objectives

1. Development of an hiPSC based liver-on-chip model
2. Emulating liver zonation be recreating metabolic patterning.
3. Improvement of microfluidic platform by integration of metabolic sensors.
4. Establishment of Multi-Organ-Systems: WAT-Liver-on-chip and Gut-Liver-on-chip

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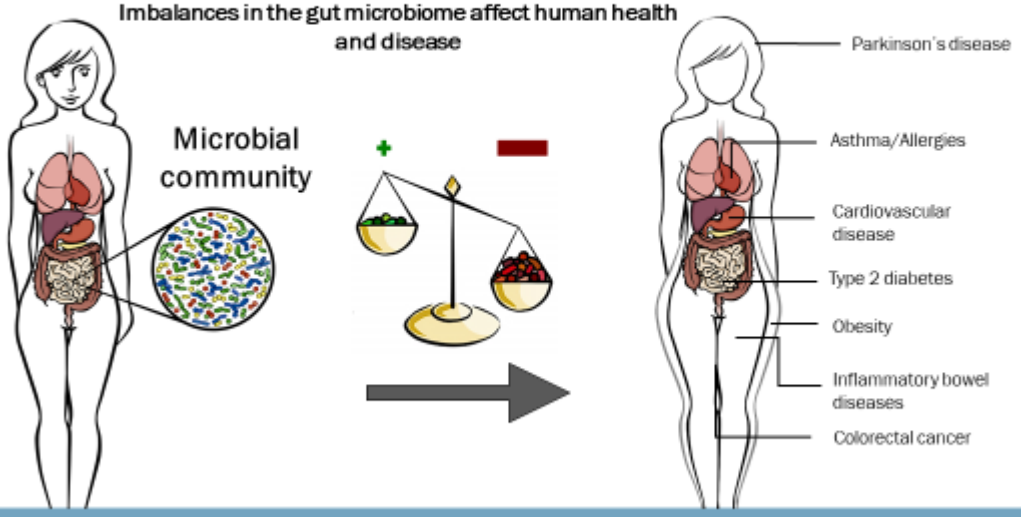
WP6 – ESR12: Gut-on-chip

32

Imbalances in the gut microbiome affect human health and disease



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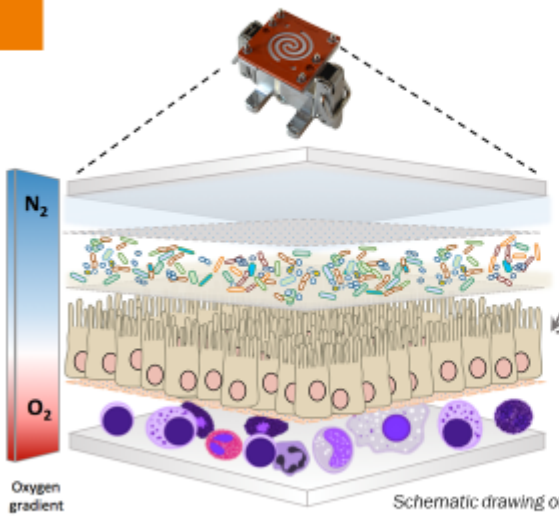
WP6 – ESR12: Gut-on-chip



Schematic drawing of HuMiX 3.0

- Integrate iPSC-derived intestinal epithelial cells to improve physiological resemblance — AIM 1
- Integrate on-chip functionality to monitor gut barrier function — AIM 2
- Interfacing of gut and liver model to mimic physiological organ communication — AIM 3

Study host-microbe interactions using Gut-on-chip: HuMiX 3.0

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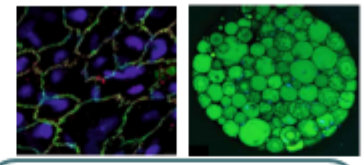
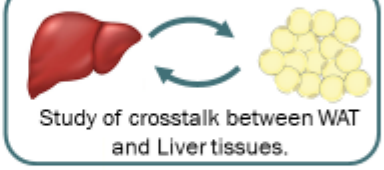
WP6 – ESR13: WAT-chip

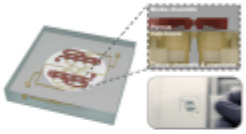
Focus of the project

- Physiologically relevant tissue crosstalk
- Patient specific immune cell responses
- Real-time sensing of glucose, lactate consumption and oxygen in metabolizing tissues

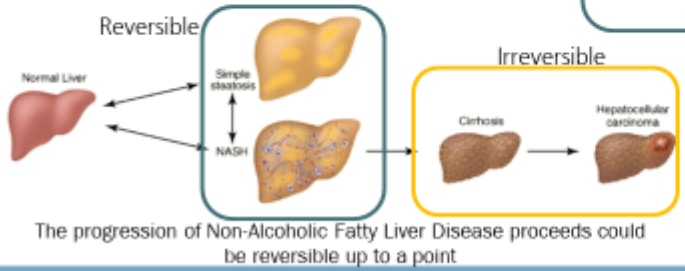




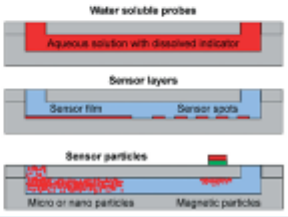
Study of crosstalk between WAT and Liver tissues.



WAT chip



The progression of Non-Alcoholic Fatty Liver Disease proceeds could be reversible up to a point



Sensors help to visualise parameters which will affect cell function in real time[4]

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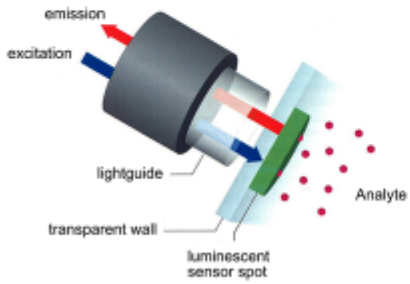
WP6 – ESR14 / ESR15 : Sensors

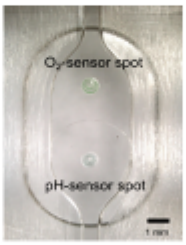
35

Integrated Optical Chemical Sensors

A luminescent sensor spot is read-out contact-less via an optical fibre



Microfluidic chips with integrated sensors



Advantages:

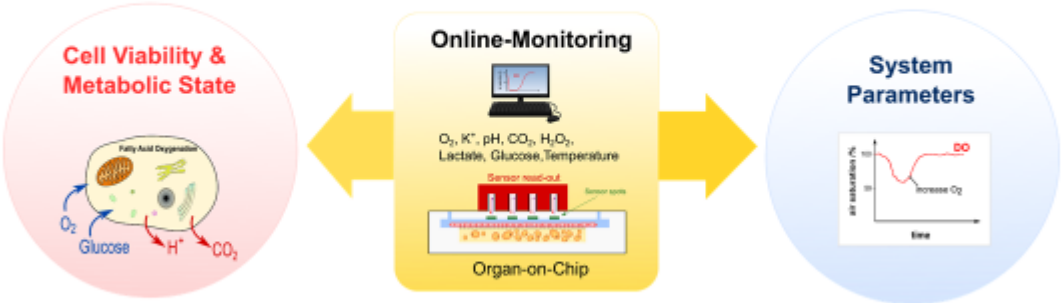
- *In-situ* measurement
- Real-time measurements
- Contactless measurements
- Non-invasive
- No reference-elements
- Simple fabrication steps
- Spot size 500 µm

in-situ measurement of O_2, pH, CO_2, Glucose, Lactate, Ions, Temperature

WP6 – ESR14 / ESR15 : Sensors

36

Online-monitoring in Organ-on-Chips with integrated optical sensors



In-line sensors enable:

- Monitoring and control of cell culture conditions
- Monitoring of tissue conditions to ensure functional organ activity
- Obtain metabolic data for disease/toxictiy models

Acknowledgements

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