HEMULE

A NOVEL MULTI-ORGAN MICROFLUIDIC CHIP:
ON THE WAY TO THE COMPLEXITY OF A LIVING ORGANISM

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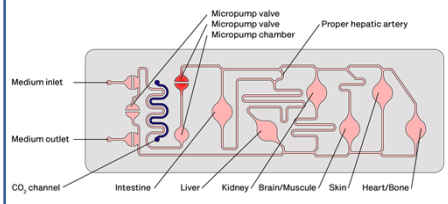
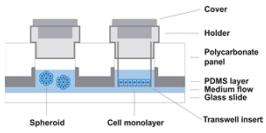
Currently an increasing number of physiologically relevant organ-on-chip platforms are reported. Most of them are focused on modeling particular organs or their functional elements. Here we present H-chip, a novel platform capable of culturing up to six different organotypic models integrated into a single microfluidic circuit (Samatov et al., 2015). The developed platform provides constant long-term circulation and automated replenishment of medium in the circuit. The key characteristics of the medium flow, including volumetric flow velocity and resulting shear stress, are similar to the ones found in a human organism. The medium flow-dependent increased viability of cultured cells supports the physiological relevance of the presented platform.

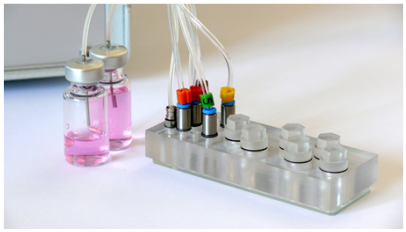
Design of H-chip



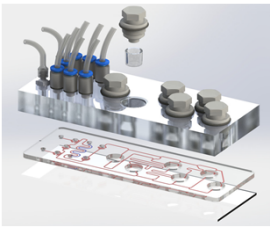
Schematic side view of cell chambers containing spheroids or cell monolayer



Connected H-chip

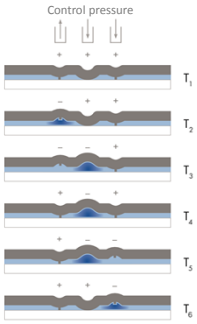


Exploded view

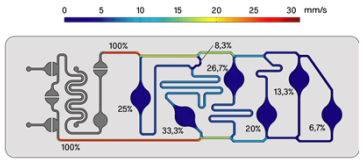


Circulation of medium

Pump cycle

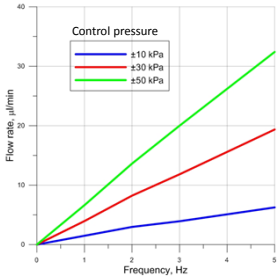


Flow distribution map



Proportions of mean flow rate are indicated in %
Instaneous flow rate (color-coded) is given for ±30kPa

Pump control parameters and resulting mean flow rate



Basic parameters of H-chip

Total volume	546 µl
Circuit volume	27 µl
Surface	1,761 mm²
Volume/surface	0.31 mm
Circuit/cell monolayer volume ratio	90
Circuit/cell spheroid volume ratio	137
Shear stress, dyne/cm²	0.2-2.0 (monolayer), 0.9-8.7 (spheroid)
Capacity of a single well	Monolayer of 50,000 cells; 20 spheroids (∅200 µm each)

Parameters of medium circulation

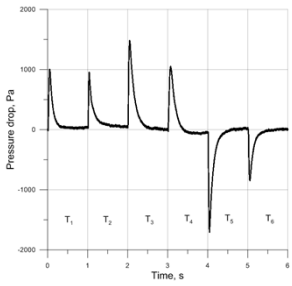
Pump pressure		±10 kPa					
Organ		Intestine	Liver	Kidney	Brain/Muscle	Skin	Heart/Bone
Maximum instantaneous velocity, mm/s	1 Hz	0.402	0.522	0.424	0.329	0.204	0.104
	2 Hz	0.406	0.528	0.429	0.333	0.206	0.106
	3 Hz	0.406	0.528	0.429	0.333	0.206	0.106
	5 Hz	0.423	0.549	0.446	0.346	0.215	0.110
Average velocity, mm/s	1 Hz	0.015	0.019	0.015	0.012	0.007	0.004
	2 Hz	0.029	0.038	0.031	0.024	0.015	0.008
	3 Hz	0.039	0.050	0.041	0.032	0.020	0.010
	5 Hz	0.062	0.080	0.065	0.050	0.031	0.016

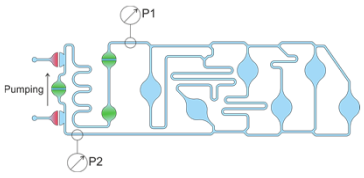
Pump pressure		±30 kPa					
Organ		Intestine	Liver	Kidney	Brain/Muscle	Skin	Heart/Bone
Maximum instantaneous velocity, mm/s	1 Hz	0.780	1.013	0.823	0.639	0.396	0.203
	2 Hz	0.815	1.058	0.859	0.667	0.414	0.212
	3 Hz	0.854	1.110	0.901	0.699	0.434	0.222
	5 Hz	0.945	1.228	0.997	0.774	0.480	0.246
Average velocity, mm/s	1 Hz	0.032	0.042	0.034	0.026	0.016	0.008
	2 Hz	0.081	0.105	0.085	0.066	0.041	0.021
	3 Hz	0.116	0.151	0.123	0.095	0.059	0.030
	5 Hz	0.191	0.248	0.201	0.156	0.097	0.050

Pump pressure		±50 kPa					
Organ		Intestine	Liver	Kidney	Brain/Muscle	Skin	Heart/Bone
Maximum instantaneous velocity, mm/s	1 Hz	1.106	1.437	1.167	0.906	0.562	0.287
	2 Hz	1.181	1.535	1.247	0.967	0.600	0.307
	3 Hz	1.284	1.668	1.355	1.052	0.652	0.334
	5 Hz	1.440	1.871	1.519	1.179	0.731	0.374
Average velocity, mm/s	1 Hz	0.065	0.085	0.069	0.053	0.033	0.017
	2 Hz	0.134	0.174	0.141	0.110	0.068	0.035
	3 Hz	0.197	0.256	0.208	0.161	0.100	0.051
	5 Hz	0.319	0.415	0.337	0.261	0.162	0.083

Pump pressure		±10 kPa	±30 kPa	±50 kPa
Circuit, s	1 Hz	1091	497	243
	2 Hz	544	196	119
	3 Hz	411	137	81
	5 Hz	258	83	50
Flow rate, µl/min	1 Hz	1.48	3.25	6.63
	2 Hz	2.966	8.226	13.62
	3 Hz	3.924	11.82	20
	5 Hz	6.25	19.36	32.41

Pressure drop between two sensors during the pump cycle

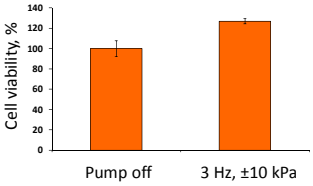




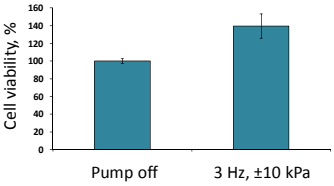
Cells in H-chip

Neutral red uptake based viability assay (Repetto et al., 2008) of hepatocytes incubated in H-chip for 48 h

Differentiated hepatocytes in 3D culture



Non-differentiated hepatocytes



Future perspectives

- Co-culture of micromodels of various organs
- Modelling the pathological processes

References

Repetto G, del Peso A, Zurita JL. Neutral red uptake assay for the estimation of cell viability/cytotoxicity. Nat Protoc. 2008;3(7):1125-31

Samatov TR, Shkurnikov MU, Tonevitskaya SA, Tonevitsky AG. Modelling the metastatic cascade by in vitro microfluidic platforms. Prog Histochem Cytochem. 2015 Feb 7. pii: S0079-6336(15)00002-9. doi: 10.1016/j.proghi.2015.01.001