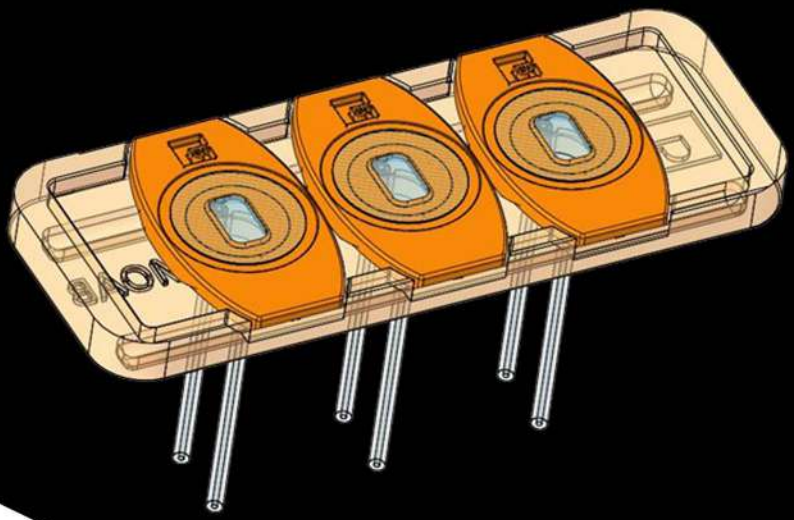
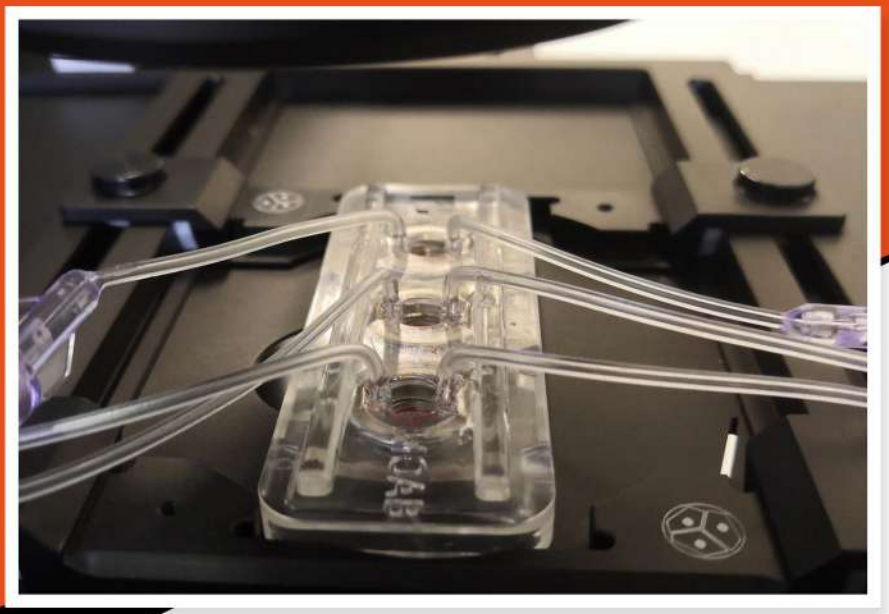
MOAB

Miniaturized Optically Accessible Bioreactor

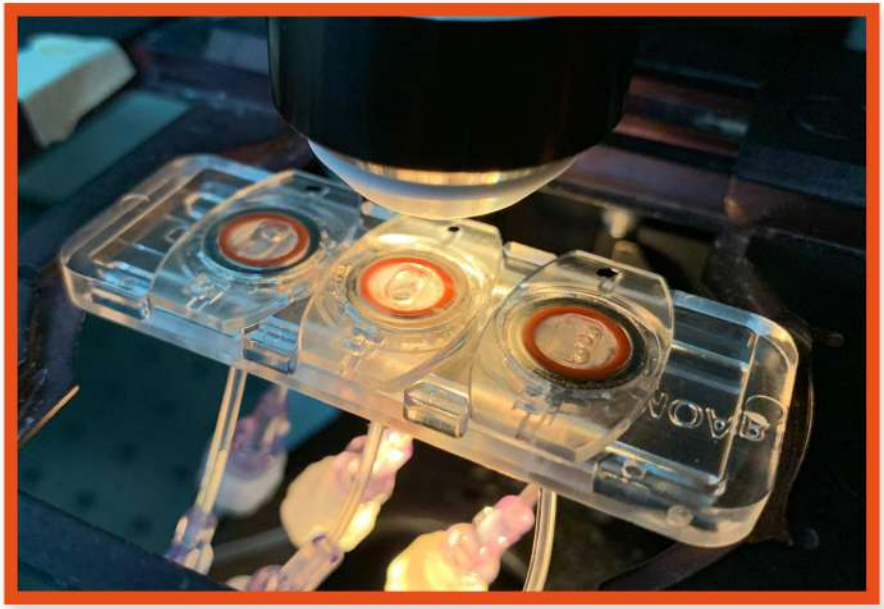


The first fully optically accessible millifluidic bioreactor for scaffold-based 3D cell culture

MOAB mounted on an inverted microscope



MOAB mounted on an upright microscope



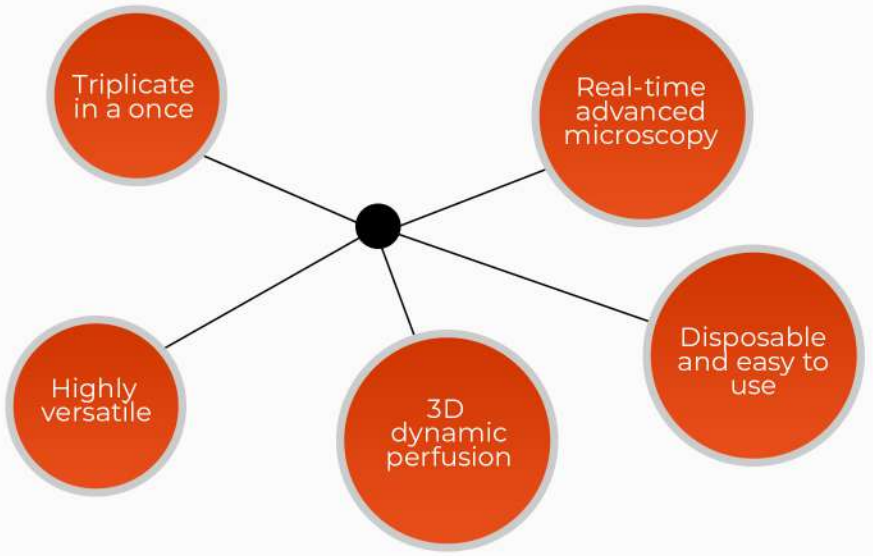
MOAB the smart bioreactor

The **Miniaturized Optically Accessible Bioreactor (MOAB)** is a smart multi-chamber dynamic cell culture system for 3D, long-term engineered tissue growth.

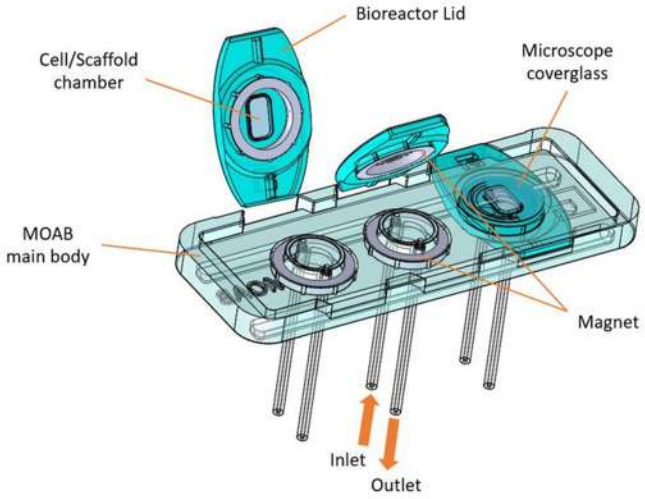
MOAB provides three indipendent perfusion lines with optically accessible micro-chambers in a very compact design which allows real-time, non invasive investigation through the most advanced microscopy techniques.

The possibility of finely controlled culture conditions by wide range of flow-rates and pressures, the micro-3D environment, the flexibility and ergonomics makes MOAB a powerful tool for tissues in vitro modelling and pharmacokinetics studies.

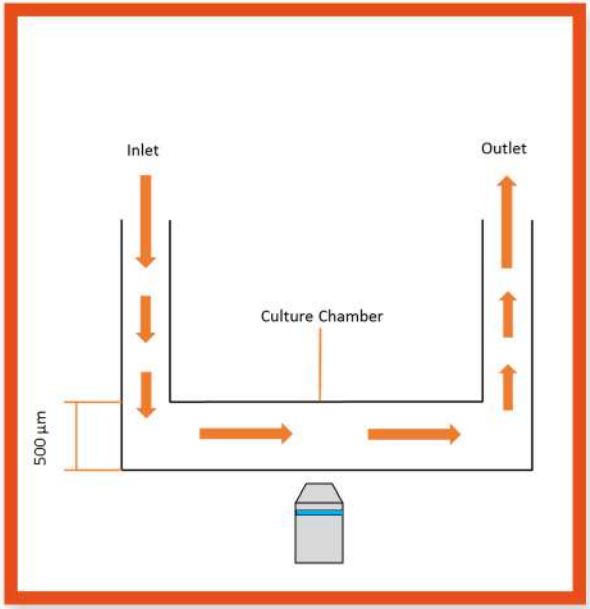
Triplicate could be retrieved from a single experiment thanks to the possibility to simultaneously cultivate three independent scaffolds using the same device, under identical environmental conditions. These results indicated some bioreactor flexibility and our system allows to study the influence of several parameters on engineered tissue growth. Our system, characterized by controlled conditions in a wide range of allowable flow rate and pressure, permits to systematically study the influence of several parameters on engineered tissue growth, using viable staining and a standard fluorescence microscope.



Concept



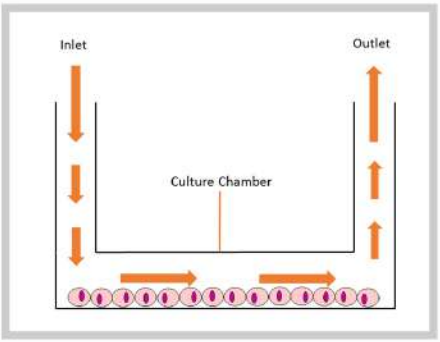
Inside the perfectly sealed miniaturized chamber MOAB guarantees highly controlled culture conditions thanks to the tissue-like **interstitial levels of flow** for very tuned cellular stimulation.

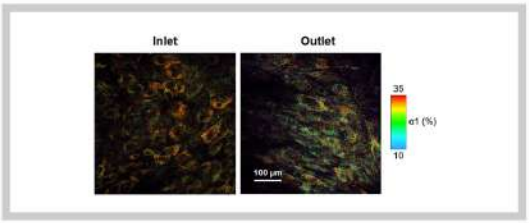


How It Works

MOAB can host several types of cell culture substrates

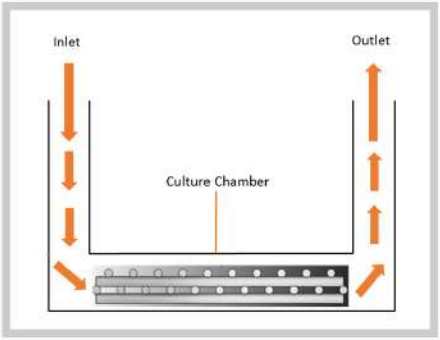
Cell monolayer

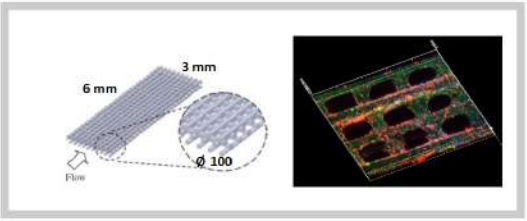




h-MSCs under dynamic perfusion expressing metabolic behaviour (α1) imaged, non invasively, by Fluorescence Lifetime Imaging Microscopy.

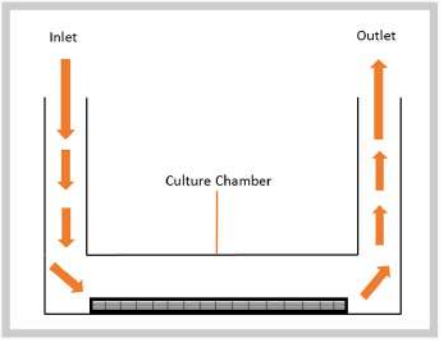
Macroscopic scaffold

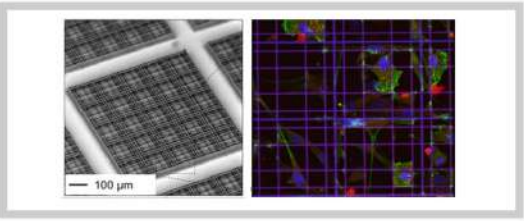




Left: polystyrene scaffolds from 3DBiotek® for interstitial perfusion. Right: confocal image showing antigen-specific T-cells adhering to dendritic cells while crawling on the scaffold.

Microscopic Nichoid TM scaffold



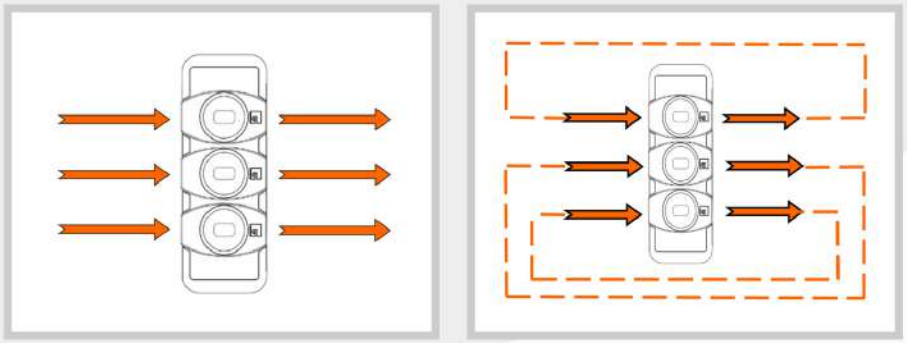


Left: SEM image of the NICHIOID architecture. Right: confocal image showing mesenchymal stem cells adhering inside the 3D microstructure

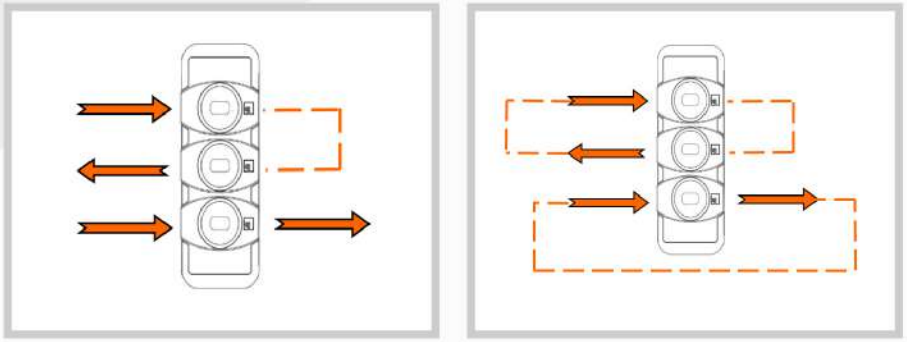
Perfusion circuits

MOAB can be configured in several perfusion circuits

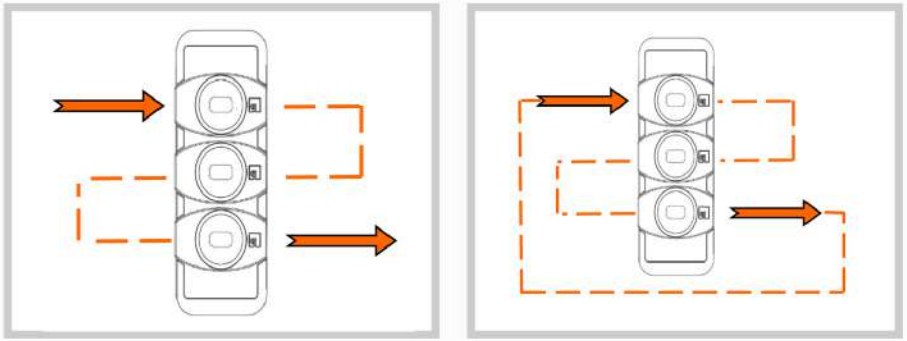
Standard parallel perfusion, with single passage or recirculation



Perfusion with single chamber bypass, with single passage or recirculation



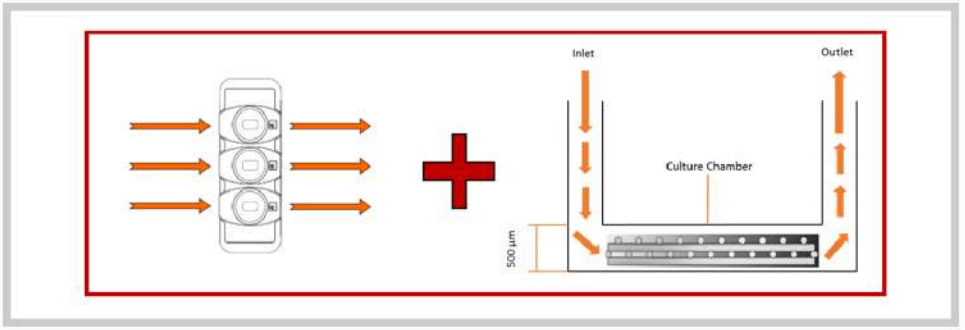
Perfusion with double chamber bypass, with single passage or recirculation

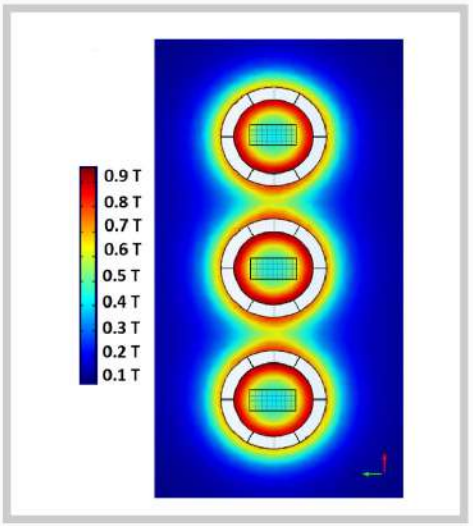


Applications

The Influence of static magnetic field on cell response

Adopted configuration





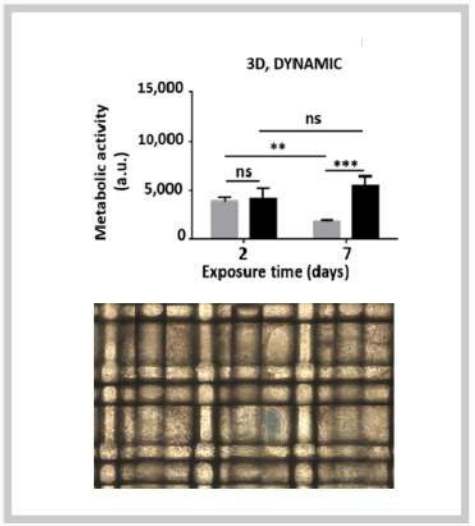


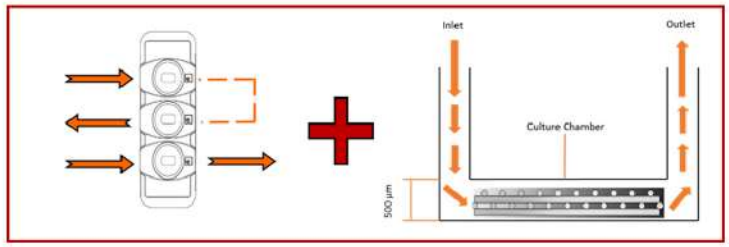
Figure: On the left, color map showing the distribution of the magnetic induction field around the magnets (top view). The black rectangles highlights the dimensions of polystyrene scaffolds. On the right, upper part, metabolic activity of SH-SY5Y cells with time in dynamic conditions; we showed the results from resazurin assay (3 replicates/group). We reported the results as mean ± SD. On the lower right, image from optical microscope of the populated polystyrene scaffold.

Reference: Izzo, L., Tunesi, M., Boeri, L., Laganà, M., Giordano, C., and Raimondi, M.T. (2019). Influence of the static magnetic field on cell response in a miniaturized optically accessible bioreactor for 3D cell culture. Biomed Microdevices. 21,29. DOI: 10.1007/s10544-019-0387-8

Applications

Model of fronto-temporal neurodegeneration

Adopted configuration



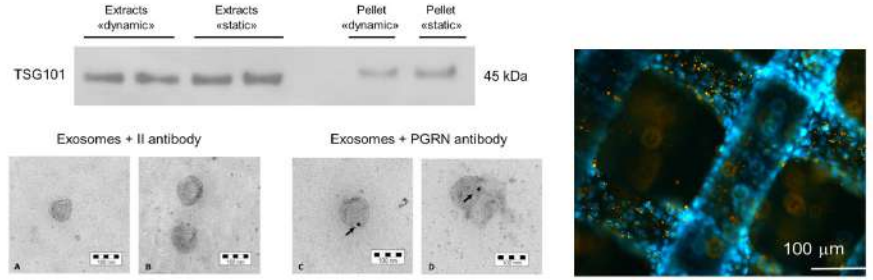


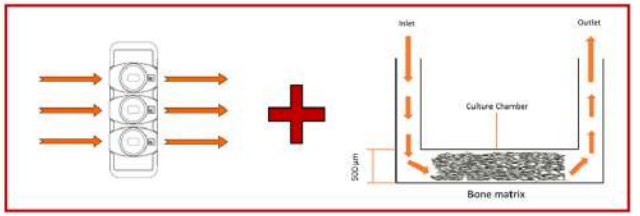
Figure: Left. Western blot and electron microscopy of exosomes collected from SH-SY5Y cells. Western blotting to detect the presence of the exosomal marker TSG101, both in the exosome fraction and in the whole cell protein lysate in dynamic vs static. The presence of the FTLD-related protein progranulin was observed in exosomes by electron microscpy. Right. Confocal microscopy of cell populated polystyren scaffold under perfusion inside MOAB.

Reference: Tunesi, M., Fusco, F., Fiordaliso, F., Corbelli, A., Biella, G. and Raimondi, M., 2016. Optimization of a 3D Dynamic Culturing System for In Vitro Modeling of Frontotemporal Neurodegeneration-Relevant Pathologic Features. Frontiers in Aging Neuroscience, 8, 146. https://doi.org/10.3389/fnagi.2016.00146

Applications

Human bone perivascular niche-on-a-chip

Adopted configuration



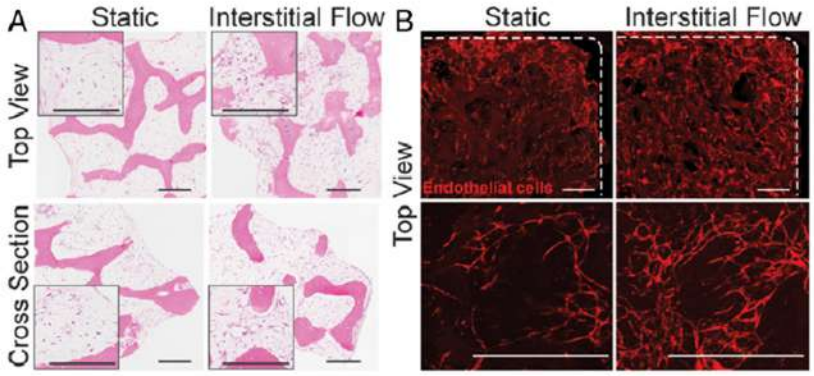


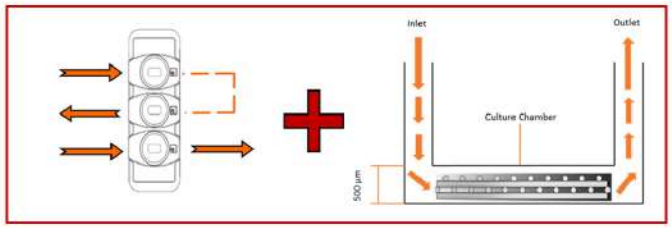
Figure: Flow promotes vasculogenesis in the BoPV niche-on-a-chip. (A) Top view and cross-section of hematoxylin and eosin staining, showing newly formed tissue and capillary-like structures filling the trabecular spaces in the BoPV niche cultured statically or with interstitial flow. (Scale bar: 250 µm). Composite images were obtained by capturing and stitching high-magnification images. (B) Live confocal images showing RFP-labeled ECs in the BoPV niche and the static controls, after 2 weeks of culture.

Reference: Marturano-Kruik, A., Nava, M., Yeager, K., Chramiec, A., Hao, L., Robinson, S., Guo, E., Raimondi, M. and Vunjak-Novakovic, G., 2018. Human bone perivascular niche-on-a-chip for studying metastatic colonization. Proceedings of the National Academy of Sciences, 115(6), pp.1256-1261.

Applications

Exosome tracking in vitro reproduce in vivo kinetics, timing of distribution and fusion to targeted tissues

Adopted configuration



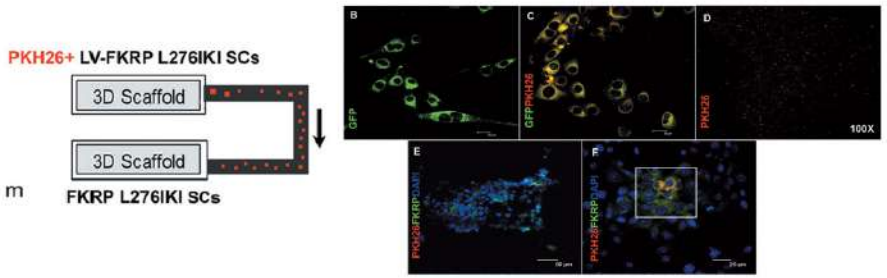


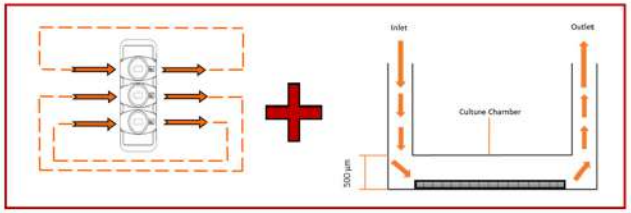
Figure: PKH26+FKRP L276IKI satellite cells were seeded on the first scaffold and secrete PKH26+exosomes, able to reach not-infected SCs seeded on the second scaffold, running through a microfluidic channel connecting the two chambers (A). IF analysis of infected FKRP L276IKI SCs; GFP signal was detected in the cell cytoplasm (B). PKH26- labelled FKRP L276IKI SCs cells (C). PKH26- exosomes secreted by PKH26- labelled FKRP L276IKI SCs cells (D). Images were taken at 100X magnification. Immunofluorescent analysis of not-infected SCs seeded on the second scaffold. PKH26þexosomes- (in red) targeted SCs recovered the expression of wildtype FKRP (in green). Nuclei were stained with Dapi (E, F).

Reference: Frattini, P., Villa, C., De Santis, F., Meregalli, M., Belicchi, M., Erratico, S., Bella, P., Raimondi, M., Lu, Q. and Torrente, Y., 2017. Autologous intramuscular transplantation of engineered satellite cells induces exosome-mediated systemic expression of Fukutin-related protein and rescues disease phenotype in a murine model of limb-girdle muscular dystrophy type 2I. Human Molecular Genetics, 26(19), pp.3682-3698.

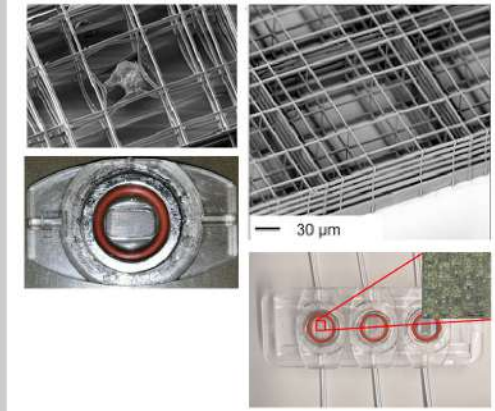
Project in progress

MOAB - NICHOID

Adopted configuration



One of the fundamental goals in regenerative medicine is to control key aspects of stem cell response, including migration, proliferation and differentiation. MOAB® - NICHOID allows to host and cultivate stem cells in synthetic niche engineering, which is based on building a closed container in which the cells are confined and maintain the stemness potential. We introduce the new perspective of building an open 3D microenvironment, structurally interacting with individual cells, to guide their spontaneous homing and proliferation of purely mechanical cues for guiding stem cell behavior.



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