Sample preparation in for high end imaging assays

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Sample preparation is a critical step in today's imaging. One the one hand samples need to be prepared in the most physiological relevant manner but the samples need to be perfectly ready for today's high end imaging systems as confocal imaging, super resolution or correlative light and electron microscopy (CLEM).

As a biophysical company we shape imaging chambers with well defined cavities in a way that we can address biological studies e.g. wound healing or angiogenesis assays or cell perfusion assays. That way we can either study cells in or on gels, hereby analyzing their biomechanical properties or we can produce extremely well-defined flow chambers, to address, simulate and analyze the shear stress effects to endothelia cells.

In the talk I will focus on two assays.

Angiogenesis where a tube formation is analyzed. In this study we used Human umbilical vein endothelial cell´s (HUVEC´s) to follow a tube formation in 2 D. Therefore individual HUVEC`s have been seeded onto a Matrigel cushion. In time, these cells start to form tubes and can be used as models for blood vessel formation. The formation of these tubes in time resolved microscopy assays is shown. The analysis of the data is done with a WIMASIS tool – that allows the read out of parameters like numbers of tubes, branching points and overall coverage of the surface.

A *perfusion assay* where HUVEC´s are cultivated at physiological flow conditions with a shear stress of 20 dyne/cm2. In such a perfusion assays micro fluidic structures are used to resemble a physiological blood flow. The channel dimensions are in a range of a few 100 µm. Here, the alignment of a HUVEC layer is shown in dependency of the applied flow conditions. Further on data on a rolling and adhesion assay as well as a transmigration of white blood cells to an endothelial layer in confocal scanning assays are presented.

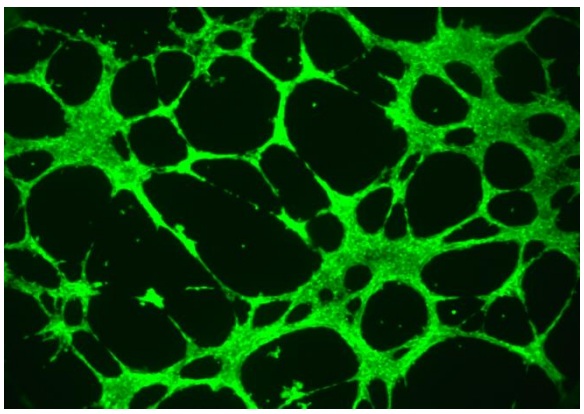

Fig. 1 Tube formation of endothelial cells

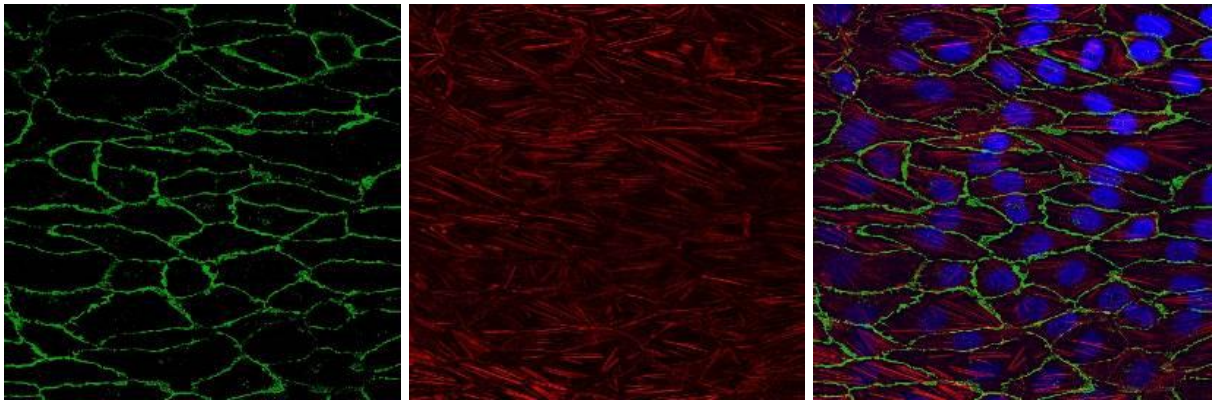

Fig. 2 Shows the alignment of proteins inside HUVEC´s under defined shear conditions