MULTIFOCAL MICROSCOPY APPLIED TO THE FLUID SHEAR STRESS ON CELLS

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Fluid shear stress (FSS) is a mechanical stimulation of cells that can elicit biophysical and biochemical responses. An example is the rapid increase in intracellular Ca^{2+} concentration in osteoblast cells, which are linked to morphological changes of the cell and other processes in bones [1]. However, despite the link to cell growth and maintenance, the exact mechanism driving these responses is poorly understood.

One way of conducting these FSS studies is by in vitro mechanotransduction experiments, in which a constant flow regime is assumed to produce a defined mechanical stimulus on cells. However, flow regimes found in perfusion chambers are not constant, meaning that shear stresses at cell level are position dependent [2] and local observations are necessary to properly connect mechanical forces and biochemical responses. Confocal micro-particle image velocimetry (μ-PIV) [3], a technique to generate 3D velocity and shear stress maps at sample level, is a powerful technique to explore these effects. Unfortunately, this method suffers from a limited acquisition speed.

In this work, a multifocal diffraction microscope (MUM) is used to recover the local shear stress generated by a fluid flowing on top of a fixed HeLa cell, with sub diffraction limited precision and high frame rate (faster than 10 frame/s). To reconstruct 3D velocity and shear stress maps, 1 μm beads within the flow are tracked with a three-plane MUM system, whose planes are spaced 640 nm apart [4]. Because the three planes are recorded simultaneously, this technique can record xyz position information from an 8 μm thick volume in a single shot, with an axial localisation precision (50 nm) smaller than one tenth of the axial diffraction limit. The excellent shear stress resolution given by MUM should clarify the mechanisms through which organisms develop, grow and adapt while undergoing FSS.

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