Freestyle Fluidics – A novel platform to study macrophage chemotaxis

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Background

Chemotaxis is defined as directed cell migration along chemoattractant gradient and is crucial in inflammation and development. Many chemotaxis assays have limited physiological relevance, no flexibility in experimental design and are hard to reproduce. This study aimed to develop a highly reproducible chemotaxis platform that addressed these challenges and to compare it to existing real-time modified Boyden chamber chemotaxis assay.

Material and methods

Freestyle fluidics (FF) is a novel robotic device that prints circuits in any shape using cell media and stabilises them using fluorocarbon oil FC40. Current protocol involves printing three parallel chambers connected to the middle one by 3 channels. Bone marrow-derived macrophages are plated in one of the chambers and after 24h rest 10nmol/L chemoattractant complement C5a is added to one of the other chambers while media is added to the other. A diffusion gradient is established and cells are imaged in real time for 48h using IncuCyte Zoom.

Results

Cell migration was quantified with custom-made algorithms, which created histograms of cell distribution throughout the circuit over time and informed on the kinetics of the population and individual cells. FF allowed both population studies and single cell tracking of haptotactic movement over ECM-coated plate, while existing system only showed population migration through non-physiological 8 µm pores. FF circuits is adjusted in size to fit different population sizes and to control the diffusion rate based on the chemoattractant. FF circuits allowed to test multiple conditions simultaneously on the same cell population.

Conclusion

Freestyle Fluidics provides a novel alternative to current real-time chemotaxis systems, allowing multiplicity of designs and real-time imaging of migration under more physiological conditions, all impossible in other systems. We plan to extend our protocols to competition assays between different chemoattractants or cell types by adding additional compartments to the circuits.

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