TWO-PHOTON MICROSCOPY AND ACOUSTIC TRAPPING FOR THE ANALYSIS OF CELL OSMOSIS

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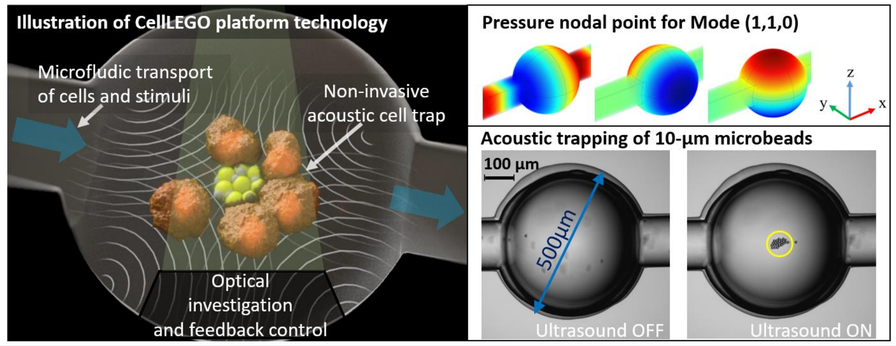
Abstract

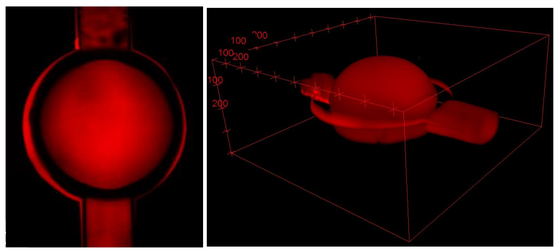
Acoustic trapping of red blood cells (RBCs) and lung cells (A549) in a spherical microchamber (SMC) are demonstrated for the manipulation and analysis of cellular microaggregates under microscopy methods brightfield and two-photon excited fluorescence (TPEF). We envision a platform technology [1] for non-invasive accumulating of cells in 3D without a wall contact in microfluidic flow conditions and compare both microscopy methods for optical analysis of cell functions and cell interactions using osmosis in RBCs as example. Future work on our system combined with TPEF will be directed towards cell function diagnostics covering a diagnostic gap of in-vitro cell testing in 3D for biomedical research.



Chip Design and Platform Technology

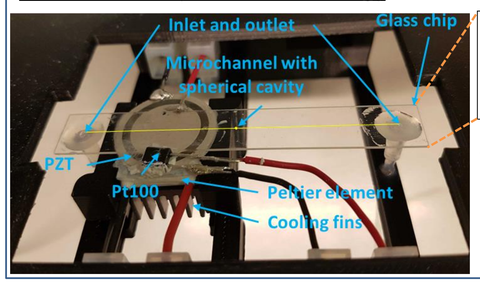
We applied the (1,1,0)-mode for cell manipulation via acoustic radiation force in a spherical microchamber [2].

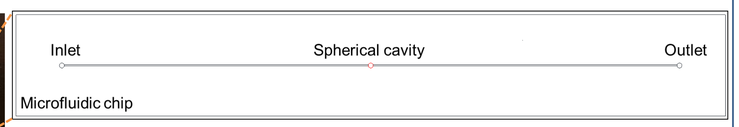
3D scan of the fabricated near-spherical microchamber under the TPEF microscope



TPEF microscopy technology

Acoustofluidics platform technology



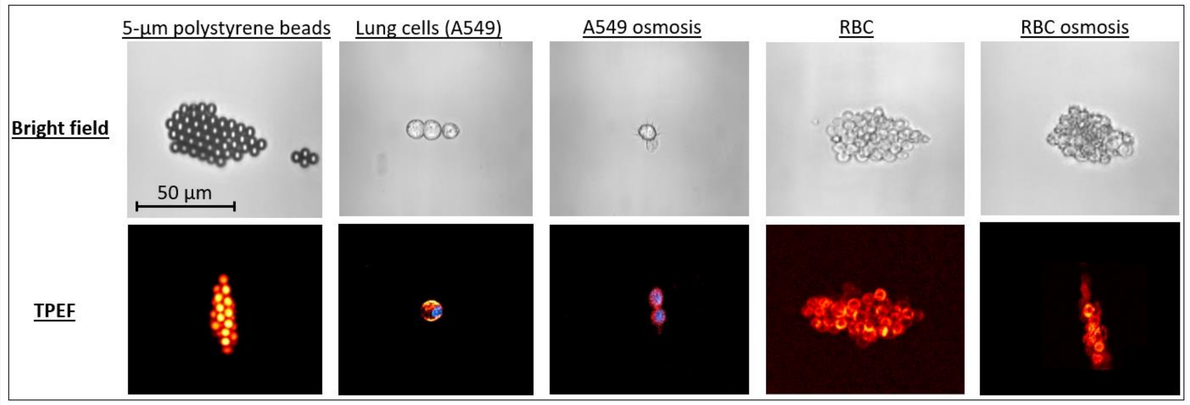


Combination of

1) Acoustic cell trapping
2) Microfluidic flow control
3) 3D real-time optical inspection and control
4) Stable body temperature environment in real-time performance

Acoustic Particle Trapping and Cell Osmosis

Comparison of bright field and TPEF microscopy method in acoustic particle/cell trapping using osmosis



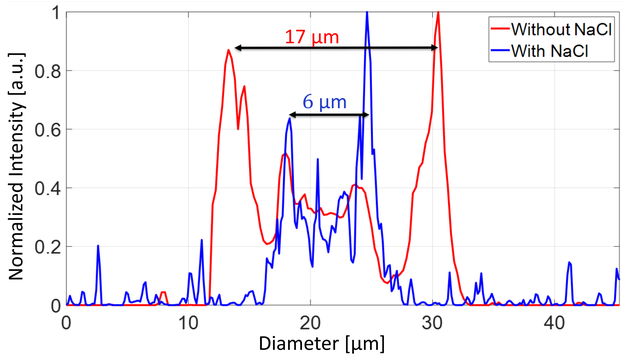
Acoustic trapping parameters: $f_{\mathrm{modulated},(1,1,0)} = 1.750 - 1.900$ MHz; $U_{\mathrm{modulated},(1,1,0)} = 15\ \mathrm{V_{pp}}$

TPEF: Femtosecond laser with $\lambda_{\mathrm{Exc.}} = 1040$ nm; $f_{\mathrm{Repetition}} = 100$ MHz

Cell labeling: Flipper-TR for cell membrane staining; Hoechst for cell nucleus

→ 3D views of the trapped cell constructs in pressure nodal point at the chamber center

Intensity profile (diameter) of lung cells without and with NaCl (osmosis)



→ Intensity profile of A549 lung cells without and with NaCl

$d_{\mathrm{withoutNaCl}} = 17\ \mu\mathrm{m}$; $d_{\mathrm{withNaCl}} = 6\ \mu\mathrm{m}$

$\Delta d = 65\ \%$

→ Equal results for diameter of RBCs without and with NaCl

$d_{\mathrm{withoutNaCl}} = 8\ \mu\mathrm{m}$; $d_{\mathrm{withNaCl}} = 5\ \mu\mathrm{m}$

$\Delta d = 37\ \%$

Outlook

Our further research will be the expansion in analysis of different cell types towards cell function diagnostics in combination with TPEF in field of biomedical research covering a diagnostic gap of in-vitro cell testing in 3D.

Conclusion

In this work we have showed acoustic cell trapping with the example of osmosis on RBCs and lung cells (A549) in a spherical microchamber under two microscopy methods for potential biomedical research applications – brightfield and TPEF.

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References

[1] B. Sailer, et al., MicroTAS 2020, 04.-09. Oktober 2020, Virtual Conference, Abstract No. 3029
[2] D. A. Russell, American Journal of Physics, 78, 549-554 (2010).

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