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Fast sorting of microfluidic droplets by content type with combined bright field and fluorescence detection

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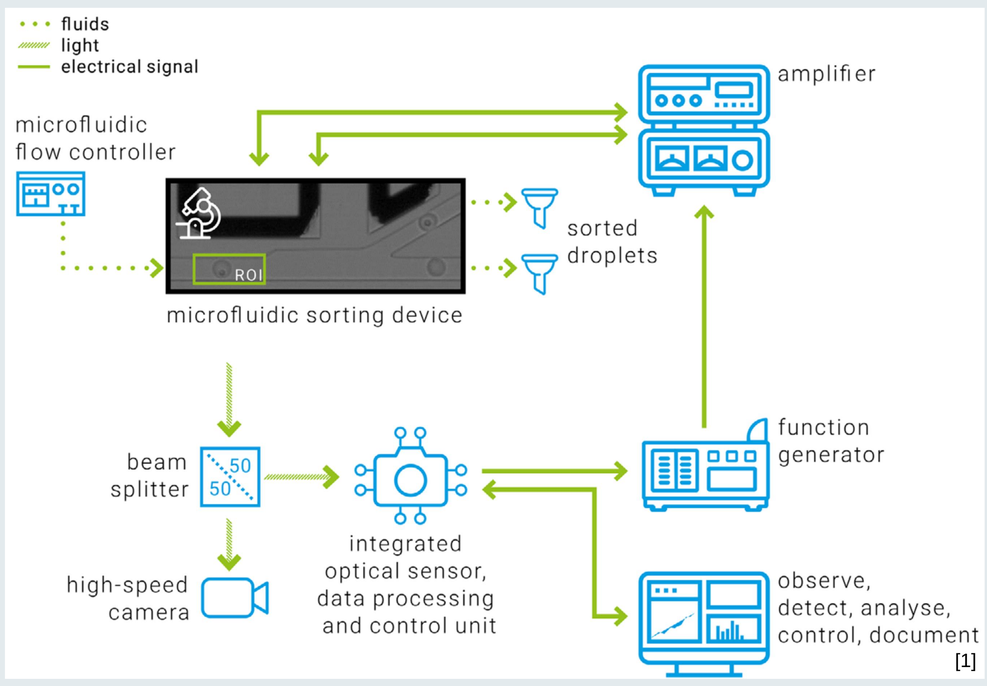
Abstract

Droplet-based microfluidics in context of fluorescent imaging can be used for a multitude of applications in biophysics, medicine, and lab-on-a-chip. One remaining issue in the encapsulation process of particle-like objects is that the number of encapsulated objects is Poisson or Poisson-like distributed. A sorting step immediately after the encapsulation reduces the number of falsely-laden droplets.

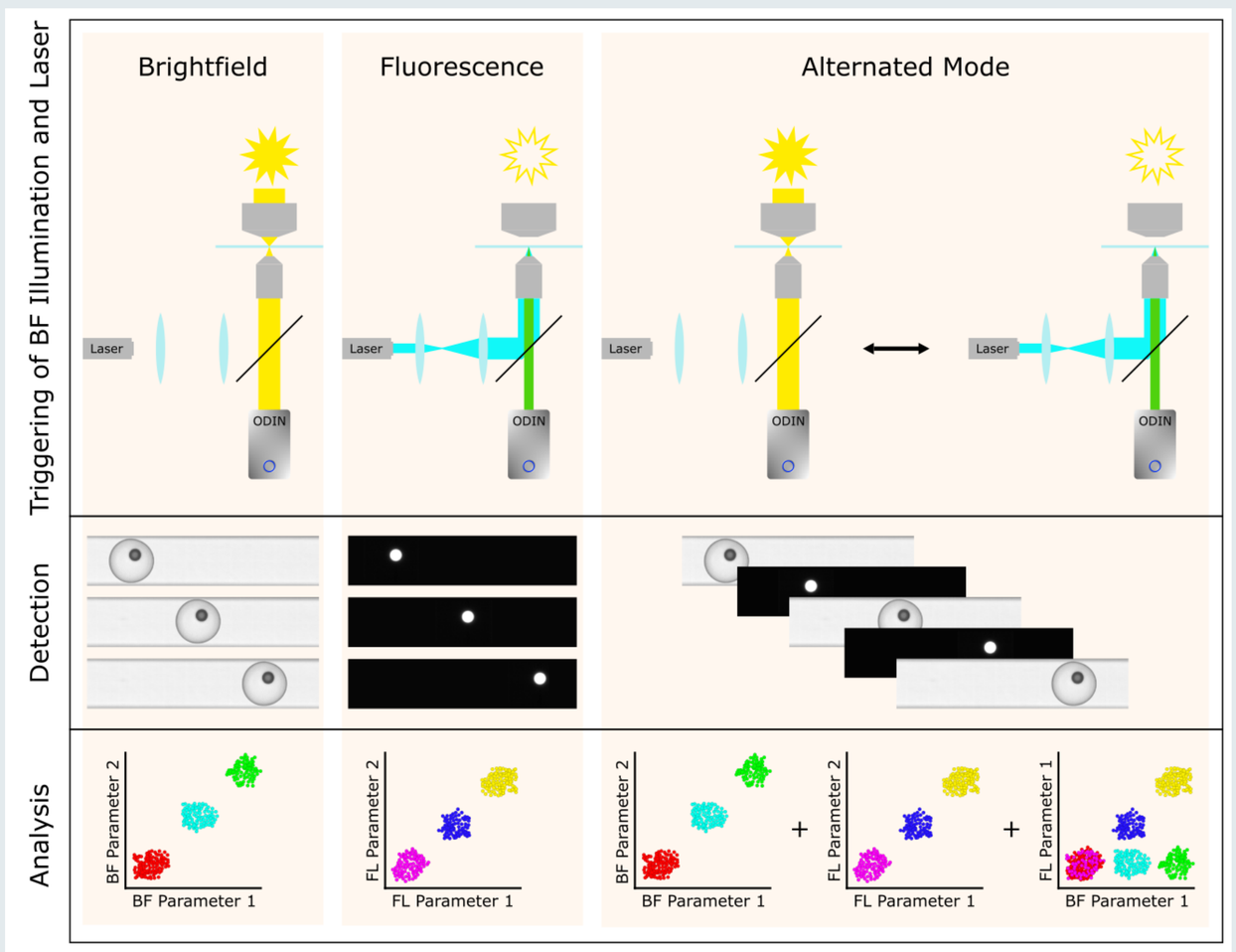
Here, we present results of the detection of beads with different diameters and different fluorescent signals. Therefore, we encapsulate 10 µm blank beads and 15 µm fluorescent beads and detect them using a time multiplexed imaging approach to simultaneously detect the population for each bead type. Thereby, we show that it is possible to achieve a user-defined, homogeneous configuration of fluorescent and non fluorescent particles in droplets.

Hardware setup

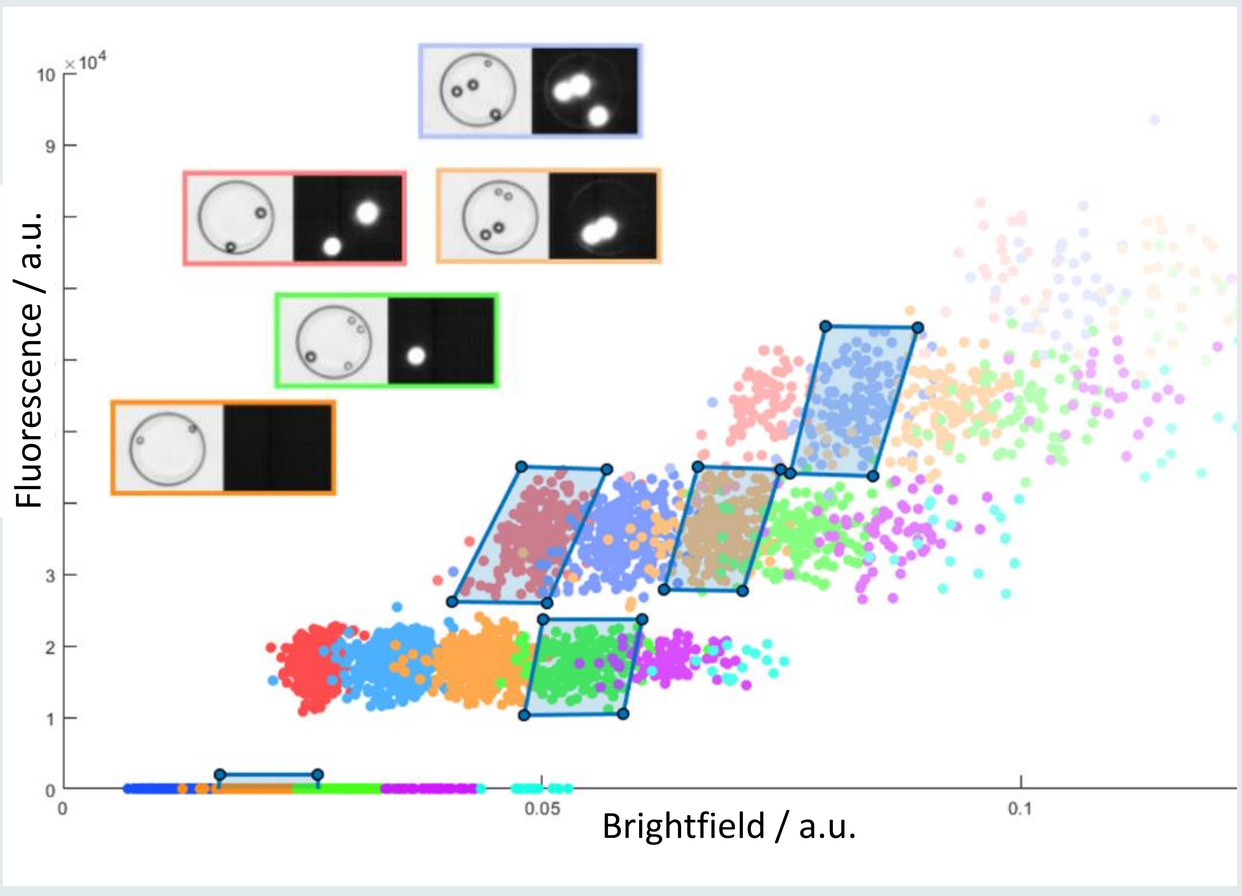


Time multiplexed setup

- Alternating between different illumination settings
- Pulsed illumination
  - Enhancing contrast
  - Eliminating motion blur
  - Minimizing bleaching
  - Minimizing light damage
- Can be extended to up to four different settings
- 10 kHz capture rate
- 3000 droplets per second
- 500 Hz sorting rate



Detection results



- User defined parameter combinations
- Freely chooseable sorting polygons
- Enrichtment of arbitrary combinations can reach up to 25
- Very high specificity of > 99 %

n=4033	2BF/2FL	2BF/0FL	0BF/2FL	3BF/1FL	1BF/3FL
Specificity [%]	99.23	99.52	99.68	99.42	99.51
Sensitivity [%]	82.88	97.32	96.02	91.74	86.67
Rel. Abundance [%]	6.37	7.39	5.60	5.41	3.35
Rel. Abundance in Polygon [%]	88.02	94.16	94.76	90.09	86.03
Factor of Enrichment	13.81	12.74	16.91	16.67	25.70

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Conclusion

With this experiments we validated the fast detection of complex droplets in microfluidic systems with high specificity, high sensitivity and a large enrichment factor.

Poster

Sorting video

Literature

[1] Frey C, Pfeil J,Neckernuss T, et al. Label-free monitoring and manipulation of microfluidic water-in-oil droplets. VIEW. 2020;1:20200101.https://doi.org/10.1002/VIW.20200101