

## Single-Layer Planar On-Chip Flow Cytometer Using Microfluidic Drifting Based Three-Dimensional (3D) Hydrodynamic Focusing

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**A real-time video of transition from the “non-focused” to “three-dimensionally focused” particle flow (Sideview, Flow Direction: Right to Left)**

### 0 to 5 sec:

The syringe pumps were turned off just prior to recording (at  $t = 0$  sec), after which particles were still shown to travel through the channel due to residual pressure at different heights and at relatively low velocities. Particle concentration was higher near the bottom of the channel due to gravity.

### 5 to 8 sec:

Injection of fluid was resumed at  $t = 5$  sec. Fluctuation of the particle stream width was observed between 5 to 6 sec, due to pressure fluctuation in the polystyrene tubing and the PDMS channel. However, the focused stream soon became stabilized, and a three-dimensionally focused particle flow was realized by  $t = 8$  sec.