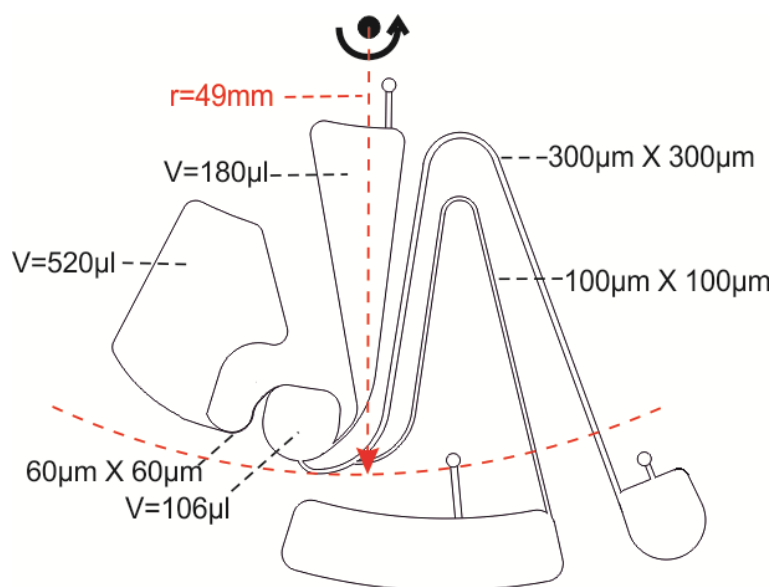


Centrifugo-pneumatic sedimentation, re-suspension and transport of microparticles – Electronic supplementary information

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5 Details of the microfluidic structure for handling of microparticles



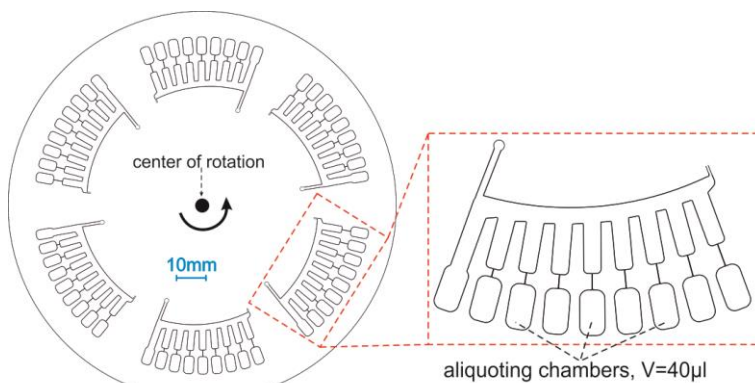
ESI Fig. 1: Detailed schematic illustration of the microfluidic structure with dimensions.

Design scenario

One challenge of handling of microparticles in microfluidics is that particles may be packed in the channel when the particles are large and the concentration is high. In particular for DNA extraction where DNA is bound to the particles which then form clusters. This may lead to blocking of the channel so that the particles restrict or prevent liquid flowing through the channel. To avoid such effect, the particle size and concentration, siphon dimensions, ratio between siphon cross-section and the cross-section of the first compression chamber were considered in the design. Using the studied configuration in ESI-Fig. 1 (particle diameter of 2.8 μm, ratio between the cross-section of the higher siphon and cross-section of the first compression chamber of 1:280) for a typical immunoassay,¹ no clogging of the channel was observed.

Calibration of particle concentration measurement

To calibrate the microparticle concentration measurement, LabDisks with aliquoting structures as used by Mark et al.² were fabricated in cyclic olefin polymer foils (COP ZF 14, Zeon Chemicals, USA) with micro thermoforming by soft lithography³ from the Hahn-Schickard Lab-on-a-Chip Design & Foundry Service. The aliquoting structure is depicted in ESI Fig. 2.



ESI Fig. 2: Schematic illustration of the aliquoting structures

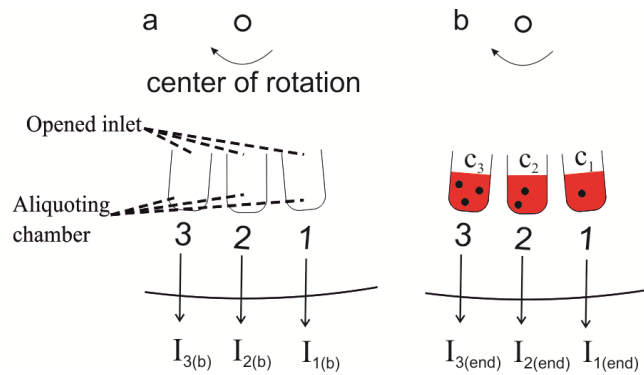
The disks were cleaned with isopropanol, rinsed with DI water and dried with nitrogen gas. Afterwards they were sealed with pressure sensitive polyolefin adhesive foils (900360, HJ-BIOANALYTIK GmbH). Each aliquoting chamber was opened at the radially inner position to facilitate loading of microparticle suspension (ESI Fig. 3-a). The background fluorescence signal ($I_{(b)}$) of each aliquoting chamber was measured in the LabDisk player (Qiagen Lake Constance GmbH). Afterwards a dilution series of microparticle suspension (concentrations are known) were prepared in 0.1M PBS containing 0.1% (w/v) BSA. The prepared microparticle suspensions are then pipetted into the aliquoting chamber and the fluorescence signals are measured ($I_{(end)}$). The fluorescence signal is owing to the autofluorescence property of the microparticles. Thus the fluorescence signal of microparticles in the i^{th} chamber (I_i) was:

$$I_i = I_{i(end)} - I_{i(b)} \quad (1)$$

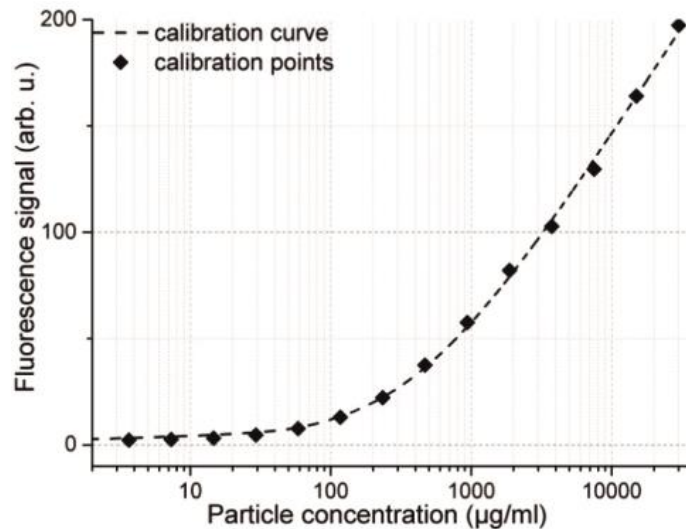
With the measured fluorescence signals (calibration points), a calibration curve was fitted (ESI Fig. 4). The corresponding function of the calibration curve is the calibration function.

$$I = F(c) \quad (2)$$

The fluorescence signal of the microparticle suspension (I) is a function of concentration (c). Thus, the concentration of any microparticle suspension with unknown concentration can be determined by first measuring the autofluorescence signal and afterwards calculation from the calibration function.



ESI Fig. 3: The method for calibration of microparticle concentration measurement. a) Schematic illustration of the aliquoting chambers after being opened at the radially inward position. The background fluorescence signal ($I_{(b)}$) of each aliquoting chamber was measured b) a dilution series of microparticle suspensions with known concentration was loaded into the chambers and the fluorescence signals are measured ($I_{(end)}$).

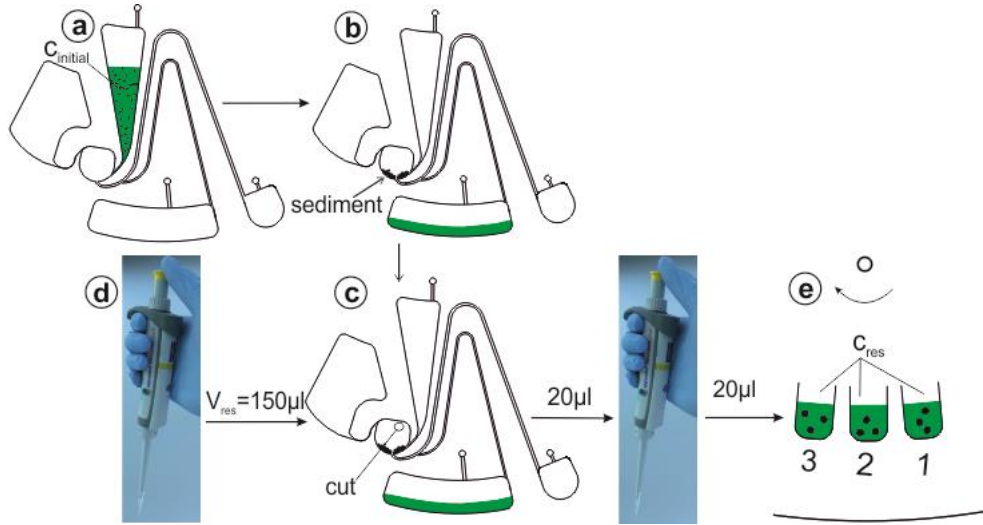


ESI Fig. 4: Plot of the calibration points and the fitted calibration curve

Quantification of particle loss after removal of supernatant

1. Measure the autofluorescence signal of initial microparticle suspension ($I_{initial}$) → calculate the initial concentration of microparticle suspension ($c_{initial}$) according to equation 2
2. Load 150µl ($V_{initial}$) of the initial suspension into the disk (ESI Fig. 4a) and perform sedimentation and remove supernatant (ESI Fig. 4b).
3. Cut a hole in compression chamber one (ESI Fig. 5c), re-suspend the sedimented microparticles manually with 150 µl (V_{res}) of 0.1M PBS containing 0.1% (w/v) BSA (ESI Fig. 5d).
4. As shown in ESI Fig. 5e, transfer 20µl of the re-suspended suspension into the aliquoting structure and measure the fluorescence signal (I_{res}). Calculate the concentration of the re-suspended particle suspension (c_{res}) according to equation 2.

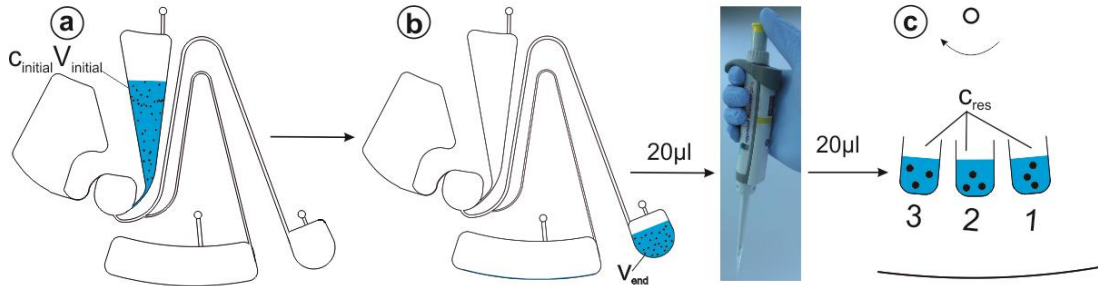
5. Calculate particle loss (PL) using equation: $PL_s = \left(1 - \frac{c_{res} \cdot V_{res}}{c_{initial} \cdot V_{initial}}\right)$



ESI Fig. 5: Schematic illustration of the process to quantify the particle loss during removal of supernatant

Quantification of particle loss after transport

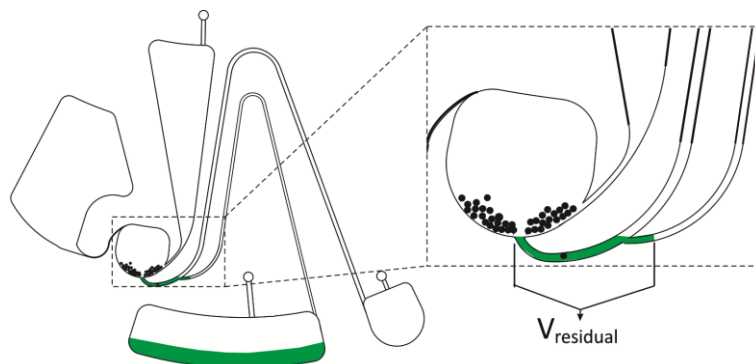
1. Measure the autofluorescence signal of initial microparticle suspension ($I_{initial}$) → calculate the initial concentration of microparticle suspension ($c_{initial}$) according to equation 2
2. Load 150 μl ($V_{initial}$) of the initial suspension into the disk (ESI Fig. 6a) and perform transport of microparticles (ESI Fig. 6b). Record real-time images.
3. Use the recorded real-time images to determine the liquid volume in the detection chamber (V_{end}) after transport of microparticles employing Solidworks 2014.
4. Manually transfer 20 μl of the microparticle suspension from the detection chamber into the aliquoting chamber and measure the fluorescence signal (ESI Fig. 6c). Calculate the concentration of the particle suspension transported to the detection chamber (c_{end}) according to equation 2
5. Calculate particle loss (PL) using equation: $PL_t = \left(1 - \frac{c_{end} \cdot V_{end}}{c_{initial} \cdot V_{initial}}\right)$



ESI Fig. 6: schematic illustration of the process to quantify the particle loss during transport

Quantification of supernatant removal efficiency (ESI Fig. 7)

1. Pipette 150 μl ($V_{initial}$) of microparticle suspension into the disk and perform sedimentation and remove supernatant. Record real-time images.
2. Use the recorded real-time images to determine the volume of residual liquid ($V_{residual}$) in the siphon channels using Solidworks 2014.
3. Calculate supernatant removal efficiency (E_s) using equation: $E_s = \frac{V_{waste}}{V_{initial}} = \left(\frac{V_{initial} - V_{res}}{V_{initial}}\right)$



ESI Fig. 7: Sedimented microparticles and residual liquid in the 1st compression chamber and siphons after removal of supernatant.

References

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