ABSTRACT
We present a novel technique for transverse mass transport and mixing in micro-channels. This behavior relies solely on the fluid inertia associated with rotating micro-particles at high flow rates. Simple channel geometry, ease of operation, biological applications and compatibility with high-throughput microsystems such as extreme throughput cytometry [1] are some of the features that distinguish this technique.

KEYWORDS: micromixing, inertial microfluidics, solution exchange, cell dipping

INTRODUCTION
Microfluidic systems have small length scales, such that the Reynolds number $Re$ in these systems is typically from $10^{-6}$ to $10$ ($Re=\rho U H/\mu$, with $\rho$ and $\mu$ the fluid density and dynamic viscosity, $U$ the flow speed and $H$ the hydraulic diameter) [2]. These low values for $Re$, representative of inertial to viscous forces in a fluidic system, place such flows in the laminar regime (i.e. predictable flowing streams), but also generally lead to association between microfluidic flows and Stokes flow, where $Re$ is assumed to be zero, inertial effects neglected and the flow thoroughly dominated by viscous forces. However, the relevance of inertial forces in small scales was recently revisited [3,4] and used for various applications, including particle focusing and separation [5].

Another consequence of flows with low Reynolds number is the absence of turbulence. For many biological applications involving mixing of different solutions, microfluidic approaches generally rely on either diffusion - which requires extremely low flow rates - or higher flow rate chaotic mixing - which requires introduction of more complex and hard-to-fabricate designs such as herringbones or curved channels [6].

Here, we show for the first time that fluid inertia associated with rotating particles at high flow rates tends to create an overall secondary flow inside micro-channels, resembling the recirculating Dean flow. This inertial effect becomes important enough to create local disturbances in the fluid flow and a drastic enhancement in cross-channel transport (Figure 1). This approach can lead to simplified mixing at high flow rates, especially in systems where particles are already present in the flow [3], with further applications in fast solution switching and reaction with bioparticles.

THEORY AND NUMERICAL RESULTS
In a shear flow without inertia, closed and symmetric streamlines are expected to surround a particle as it rotates due to a velocity differential across it. When inertia is important, it has been shown that randomly distributed particles in a channel flow inertially focus to preferred lateral equilibrium positions [4] approximately halfway between the channel centerline and wall. Focusing places particles in a condition similar to a shear flow. Subramanian and Koch [7] theoretically showed that finite inertia also alters the pattern of flow around a particle rotating in a shear flow without confinement. Addition of channel confinement, which occurs in microfluidic systems, is expected to further increase the complexity and the amount of the disturbance flow around the particle compared to the case of Stokes flow.

Figure 1: Numerical results of cross-streamline transport. (A) Comparison of the average y-velocity at the y=0 plane between Stokes and non-Stokes flow, averaged along the x-axis from -70μm to +70μm. (B) Net in-plane mass transfer, integrated along the x-axis from -70μm to +70μm: an overall secondary flow is created by the inertial effects around the spinning particle.
Numerical simulations revealed a unique recirculating disturbance flow around a confined rotating particle when inertia was considered. A single 10 μm particle stabilized at its lateral equilibrium position is considered in a Poiseuille flow within a straight rectangular channel. We observe an interesting difference between Stokes and finite-Re flows. First, by integrating the y-direction velocity in the central plane (y=0), we observe that no net mass transport occurs in Stokes flow, whereas in the inertial flow we observe a significant transverse component of velocity across the central plane (Figure 1A). Second, investigating the in-plane flow over more points within the channel cross-section demonstrates the presence of a unique secondary recirculating flow for finite inertia (Figure 1B). This secondary recirculating flow resembles Dean Flow in curved channels with the slight difference that here the inertial effect is created by the spinning particle instead of the centrifugal force of the turning fluid.

MATERIAL AND METHODS

Microfluidic devices were fabricated using standard polydimethylsiloxane (PDMS) technologies. Suspensions were injected with a syringe pump, for flow rates ranging from 5 to 300 μL/min, corresponding to a wide range of particles Reynolds numbers $R_p = \frac{Re a}{H^2}$, with $a$ the particle diameter and $H$ the hydraulic diameter) and Peclet numbers $Pe$ (defined as $Pe = \frac{L U}{D}$, with $L$ the critical channel dimension, $U$ the flow speed and $D$ the diffusion coefficient). Blood was collected from healthy volunteers and white cells were extracted after red blood cell lysis (eBiosciences). Fluorescent particles (10 μm, 1.05 g/ml, Duke Scientific) were diluted in DI water, white blood cells and HeLa cells in PBS, to achieve a desired length fraction $\phi$, with $\phi$ defined as the fraction of channel length covered by focused particles (i.e. $\phi=1$ indicates particles would all be in contact in a single line). Confocal and fluorescence images were captured with a Leica Inverted SP1 Confocal microscope and Nikon Eclipse Ti microscope. From the fluorescence intensity profile of cross-sections, the extent of transverse fluid transport can be characterized by the transport factor $TF$, $TF=2(\Delta H/\omega-0.5)$, with $\Delta H/\omega$ being the cross-section extent for intensity higher than a limiting intensity $LI = 0.2(I_{max}-I_{min}) + I_{min}$.

EXPERIMENTAL RESULTS AND DISCUSSION

To investigate the presence of cross-stream transport and the predicted recirculating flow, a simple two-inlet microchannel was fabricated. Experiments consisted of co-flowing two streams - one with fluorescent dye, one with focused particles - into one main channel (Figure 2.A, B and C). Fluorescence images were captured at 2.5 cm after the interfacial contact of the two streams. To identify the effect of particle concentration, various length fractions $\phi$ have also been considered (Figure 2.D and E).

Compared to flows without particles, the introduction of particles leads to increasing transverse transport as Peclet number increases. Extremely low flow rates are needed to obtain reasonable transverse transport due to diffusion alone. As flow rate and corresponding Peclet number increase, the extent of transverse transport drops considerably. Similar
Experiments are conducted with particles present in one of the flows. The extent of transport is again observed to be high at low flow rates and with increasing flow rate, extent of transport starts to decrease once more. However, for flow rates where inertia becomes important, an unexpected shift in the trend occurs and transport starts to increase with increasing flow rates: the width of the fluorescent region becomes narrower for $\varphi=0\%$ but widens again for $\varphi=25\%$ (Figure 2.E). A plot of $TF$ as a function of $\varphi$ also demonstrates the dependence of this transport on particle concentration (Figure 2.D). Indeed, increasing $\varphi$ intensifies the extent of transfer: while for $\varphi=10\%$ $TF$ reaches no higher than $\sim 0.6$, it is almost equal to $1$ for $\varphi=55\%$.

Deeper phenomenological understanding of this behavior can be obtained by looking at the cross-sectional shape of the flow via confocal microscopy (Figure 2.F). Without any beads, only a rectangular fluorescent shape occupies approximately half of the channel at high flow rates. On the contrary, when particles are present, the fluorescent fluid appears to be dragged from right to left as the flow rate increases beyond around $100 \mu$L/min. Increasing flow rate intensifies this deformation and finally, at $300 \mu$L/min, a fluorescent H-shape is easily discernable within the channel. Thus, confocal images clearly confirm the presence of transverse convection through secondary flow, as predicted by our numerical simulations.

Microfluidic solution switching and mixing, especially when bio-particles such as cells are already present within the system will benefit from this approach. We conducted similar experiments to those described above with HeLa cells and leukocytes, suspended in PBS. These particles are deformable and polydisperse in size ($\sim 7-30 \mu$m) but yield similar transport (Figure 3). These results are quite promising for biological applications, such as the continuous and fast staining of white blood cell subtypes before their detection in a miniaturized flow cytometer.

**CONCLUSION**

We have introduced a novel and unique physical phenomenon in which passively spinning particles flowing inside a straight channel induce a secondary flow that creates a net transverse mass transport. Numerous applications in lab-on-a-chip systems for this simple approach include fluid mixing for sample preparation, controlled cell staining and washing, analysis of protein folding, and selective blood lysis.

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