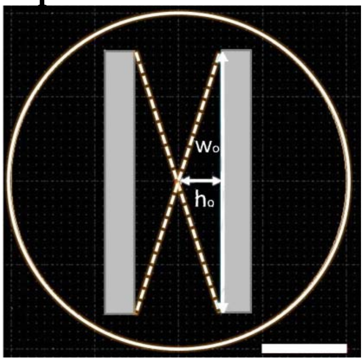
1. Footprint constraints and compactedness definition.



Suppl Fig1. Schematic representation of the space available for the microfluidic circuitry. The circle delimits the surface of the glass substrate and the two parallel rectangles represent the two microfluidic analysis chambers. We define the compactedness parameter C_o as: $C_o = h_o/w_o$ Here, $w_o = 30$mm, and $h_o = 5$mm. Scale bar: 1cm.

2. Theoretical Background of Fluid dynamics

Navier-Stokes Equation

Considering that the fluids are Newtonian, incompressible and homogeneous, fluid flow follows Navier-Stokes equation:

$$\frac{\partial \vec{v}}{\partial t} + \left(\vec{v}.\overrightarrow{grad}\right).\vec{v} = -\frac{\overrightarrow{grad}\,P}{\rho} + \overrightarrow{f_m} + \nu\Delta\vec{v} \quad (1)$$

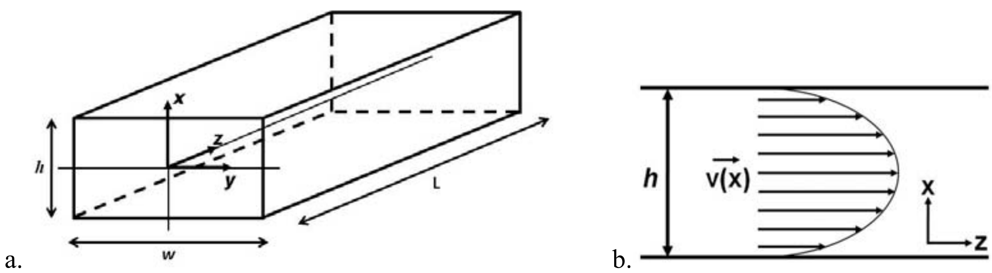
Where $\vec{v}$ (m.s^{-1}) is the fluid velocity, ρ ($kg.m^{-3}$) is the fluid density, P (Pa) is the pressure, f (N. kg^{-1}) represents body forces (per unit volume) acting on the fluid (such as gravity or centrifugal force.), ν (m^2. s^{-1}) is the cinematic viscosity.

When fluid flows in microfluidic channels, the Reynolds number (which compares the effect of momentum of a fluid to the effect of viscosity) generally becomes very low and flow is then laminar. This implies that:

$$\vec{v}(x, y, z, t) = \vec{v}(x, y, z)$$

Fluid flow in a rectangular channel

Generally, microfabricated channel are rectangular, as drawn in Fig1a. When a fluid flows in a rectangular shape channel where the length L of the channel is much longer than the other dimensions (h, the height and w the width of the channel), the Navier-Stokes equation can be simplified:



Suppl Fig2. Definition of coordinates and scale parameters of rectangular microchannels with a height *h*, a width *w* and a length *L*.

The coordinate axes are defined as shown in Suppl Fig 2a. Due to translation invariance along the *z* axis, the velocity field of a Newtonian fluid in a straight channel is parallel to the *z* axis, and takes the form $\vec{v}(x, y, z) = v(x, y).\overrightarrow{ez}$.

Consequently, the nonlinear term in the Navier-Stokes equation drops out:

$$\vec{v}.\overrightarrow{grad}(\vec{v}) = v(x, y).\frac{\partial v(x, y)}{\partial z} = 0 \quad (2)$$

In steady state, given the pressure gradient $\overrightarrow{grad}(p) = -\frac{\Delta p}{L}.\overrightarrow{ez}$ where $\Delta p = p_{out} - p_{in} < 0$ is the pressure drop between the input and the output of the channel, the velocity $\vec{v}(x,y)$ is thus given by the Stokes law (which is actually a Poisson equation) :

$$\left(\partial^2{}_x + \partial^2{}_y\right) v(x,y) = \frac{\Delta p}{\eta . L} \quad (3)$$

With η = ν/ρ, the dynamic viscosity (Pa.s)

This partial differential equation is generally not easy to solve. But in the case of a rectangular channel and with the velocity being subject to a no-slip condition at the boundary of the channel, it can be solved by using Fourier series[35-36]:

$$v(x,y) = \frac{\Delta p}{\eta . L} . \frac{4\, h^2}{\pi^3} . \sum_{n=1,3,5,\ldots\infty}^{\infty} \frac{1}{n^3\left(1 - \frac{\cosh\left(\frac{n\pi x}{h}\right)}{\cosh\left(\frac{n\pi w}{2h}\right)}\right)} \sin\left(\frac{n\pi y}{h}\right) \quad (4)$$

Here, the coordinate system is chosen so that $-w/2<y<w/2$ and $0<x<h$.
This solution is not easy to use analytically and to plot. This calculation, however, can be simplified if the channel has a rather high aspect ratio, and treating it as a mere slit (Hele-Shaw flow).

Hele-Shaw Flow
By assuming that the width of the microfluididic channel is much larger than its height (w>>h), the effect of the verticals walls can,be neglected and $\vec{v}(x,y,z) = v(x,y).\overrightarrow{ez}$. The Stokes law can then be written as:

$$\frac{d^2 v(y)}{dy^2} = \frac{\Delta p}{\eta . L} \quad (5)$$

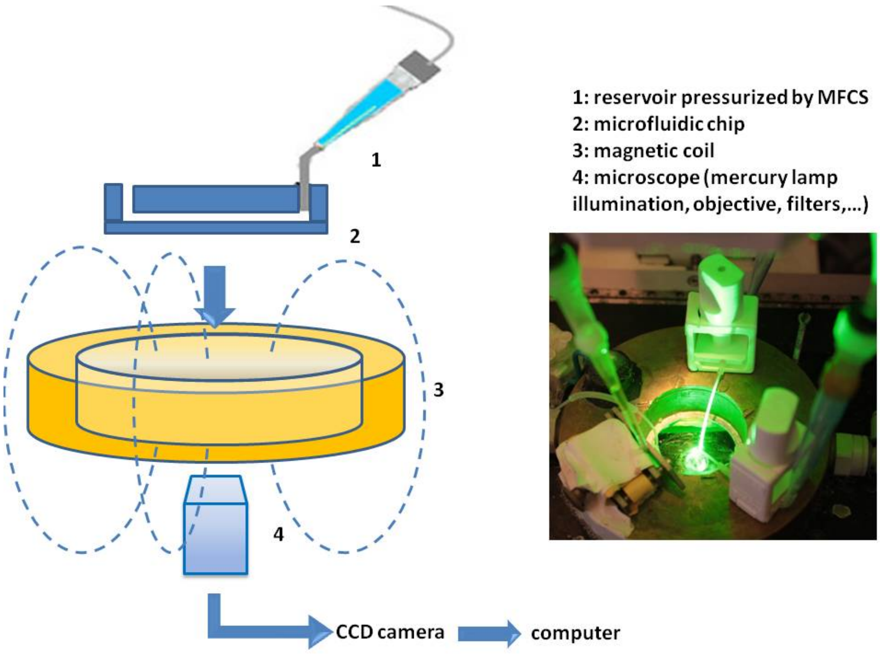
With the velocity being subject to a no-slip condition at the horizontal walls of the channel $\left(v\left(\frac{h}{2}\right) = v\left(-\frac{h}{2}\right) = 0\right)$, it follows the Poiseuille low (Fig 2b) which shows that:

$$v(x) = \frac{\Delta p}{2\,\eta . L} . \left(x^2 - \frac{h^2}{4}\right) = v_{max} . \left(1 - \frac{x^2}{\frac{h^2}{4}}\right) \quad (6)$$

Here, the coordinate system is chosen so that $-h/2<x<h/2$.

3. Experimental setup for immunomagnetic cell sorting

The microfluidic device developed in this issue was used to perform immunomagnetic cell sorting experiments (Section 5). The application of an external magnetic field was then necessary to form the array a magnetic micro columns inside the cell capture chamber. The magnetic field lines had to be oriented perpendicular to the fluid flow. We used a magnetic coil cooled by glycerol circulation. 35mT could be reached inside the chip by applying 3,5A. The space available inside the coil was of 4cm diameter. The experimental setup is depicted in Suppl Fig3. For this application, two constraints limit the maximal total footprint of the chip (flow throughput and maximum shear stress being kept constant). First, the electric energy needed to impose a given field strength increases with the square of the area. Thus the size, weight and power consumption of the power supply and cooling installation increase rapidly with the chip area, making the installation cumbersome. The above diameter of 4 cm allows the use of a low cost electric power supply. Second, in clinical applications the whole capture area has to be scanned by a high resolution imaging device, and both scanning time and memory space are directly proportional to the scanned area, so the smaller the chamber area, for a given flow rate, the more efficient the design.



Suppl Fig3: Schematic representation of experimental setup for microfluidic immunomagnetic cell sorting and photography of a microfluidic chip connected with tubing to reservoirs, introduced in the middle of the magnetic coil. Dotted lines represent some magnetic field lines produced by the coil.