

INTEGRATED PLATFORM FOR STEM CELLS SEPARATION/COUNTING

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Hematopoietic stem/progenitor cells (HSC) have vast potential for use in medical applications (such as bone marrow transplantations) and the separation of these cells from blood has an important role in the stem cells research [1]. However, the most usual techniques, (MACS™) and (FACS), have some drawbacks like being cell-loosing or very time-consuming.

This work is in the area of nanobioengineering, combining a cross-disciplinary approach of nanotechnology, bioseparation engineering and hematology with a common goal: the separation, purification and monitoring of HSC *in vitro*.

Until now a device based on magnetophoresis has been developed using microfluidics and in-situ magnetic fields (up to 6kA/m), generated by metallic lines, in order to separate stem cells from blood in a more efficient and easy way [2], [3], [4], [5]. In the same device Spin Valve sensors are used to measure the efficiency of the separation (by counting the cells as they are separated).

The geometry of the separator makes it possible to change only the path of specific stem cells, that are magnetically labeled with 50nm magnetic beads functionalized with monoclonal antibodies (MAbs), instead of separating all the magnetic elements in the fluid (which is vital to keep count of the stem cells).

The fluidic system consists in 2 micro-channels (150μm wide, 14μm thick and 40μm apart) joined over several gaps of 2.5mm and disposed in an “H”-type geometry (fig.1 and fig.2). The laminar flow is generated in the y-direction along both channels, from the inlet to the outlet. The fluidic system is bonded to the separator platform which consists in two successive lines (7μm wide and 500nm thick), deviated from the y-direction by an angle of 5 degrees, starting in one channel and ending in the other one. At the end of each metallic line there are 3 SV in each channel to count the cells that have been separated and those that might fail to be separated [6],[7].

To prove the concept and test the design, preliminary tests were made passing 2μm magnetic particles through the channels. These particles ($\chi=0.22$ and $\rho=1.1 \times 10^3 \text{ kg/m}^3$) feel a magnetic force of 9.5pN, when passing over the metallic lines (due to a magnetic field of 6kA/m and a gradient of $2.2 \times 10^6 \text{ kA/m}$, when applying 100mA), which force them to be deviated in the x-direction. With velocities flow rates around 30nL/min, particles were observed to follow the line path, moving from the beginning of the line (in the first channel) until the end of it (in the second channel) (fig.3). When any particle fail to feel the first line (due to agglomeration for e.g.) it can be separated when passing over the next line. These measurements were made for a concentration of 1×10^5 particles/μl to make the optical inspection easier. The real experiment will be made with human hematopoietic stem/progenitor cells (CD34⁺ enriched cells), from umbilical cord blood samples.

References:

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Figures:

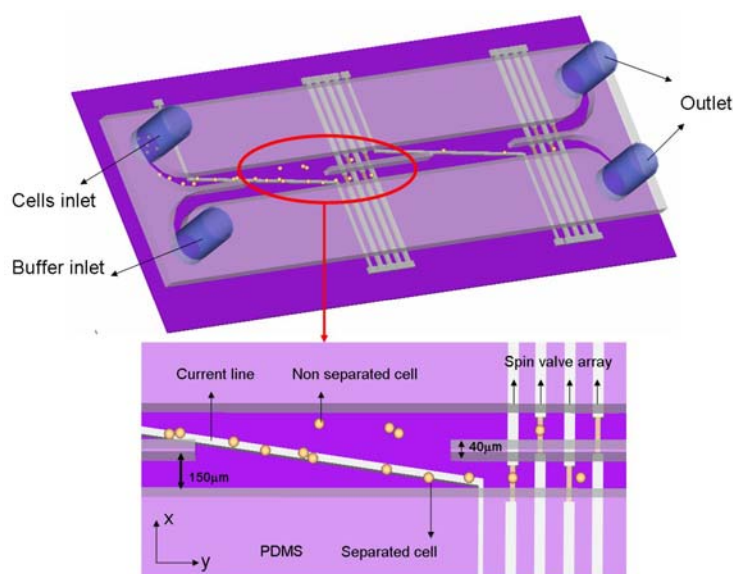


Fig1: “H-type” fluidic platform, allowing stem cells to be separated from one channel to the other due to the magnetic field created by the oblique current lines. Each channel has spin valves at the end to count the cells

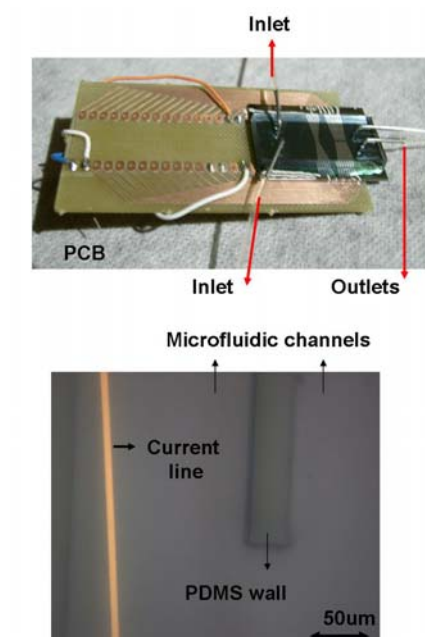


Fig2: Real chip mounted on a PCB to allow electrical measurements (top); Microscope image of the microfluidic system showing the current line near the gap between both channels

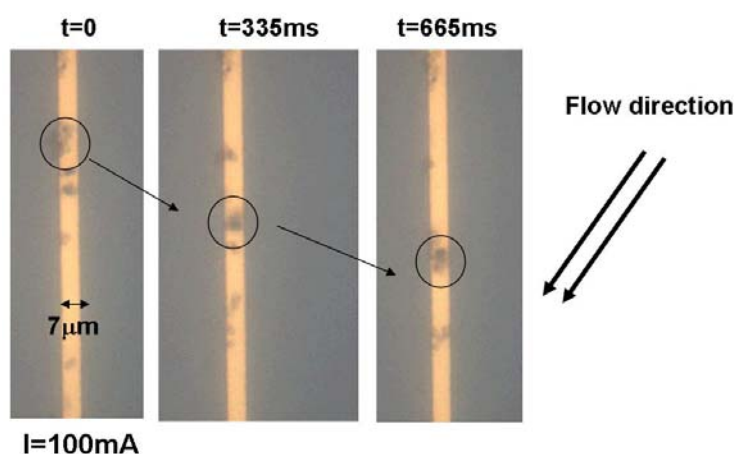


Fig3: Time evolution of the 2µm particles position due to the fluid velocity and the magnetic field created by the current line (when applying 100mA) during the separation. These particles were moving over the line with a velocity around 75µm/s.