A 3D microdevice for the in vivo trapping of cancer-associated circulating cells

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This work introduces a unique system for the capture of circulating biomarkers *in vivo*, relying, for the first time, on a 3D microdevice to be placed directly into the blood circulation, to selectively sort out cancer cells from other blood components based on their physical specificities. Circulating Tumor Cells (CTCs) are cancer cells that have detached from a tumor and have entered the blood circulation through an Epithelial-to-Mesenchymal Transition. Capturing CTCs can allow physicians to monitor cancer progression and treatment efficiency, without the need for invasive methods such as solid biopsies [1]. However, considering the rareness of CTCs in blood (<1cell/mL at early stages), capturing them *in vitro* is a true challenge. Microfiltration and microfluidic-based systems have provided robust alternatives to this challenge, showing short sample processing times and high sensitivity levels [2,3]. However, the volume of blood processed is constrained by blood draw, limiting representativeness and statistical confidence. By placing the trapping system *in vivo*, the amount of blood screened can be increased, thus reducing the bias induced by sampling. Our prototype combines the advantages of *in vivo* capture and physical trapping of CTCs (**Figure 1**). A polymeric net-like microdevice was fabricated in 3D onto a tailored metallic guidewire using a two-photon lithography process (Nanoscribe®) (**Figure 2**). To optimize and validate the prototype, we conducted computational fluid dynamic simulations and *in vitro* experimentation using a fluidic platform acting like an artificial vein in terms of blood pressure, dimensions and flow rate. We succeeded in capturing human prostate cancer cells (PC3) spiked into whole blood in just a few minutes, with no blood pre-processing required and with extremely low contamination levels (**Figure 3**). This was further confirmed *in vivo* through animal experimentation. The low pressure exerted on cells (<100 Pa), allows the preservation of cellular integrity and viability. Captured cells can then be easily collected for further analysis. This minimally invasive technology could offer high-quality information to physicians and a tool for personalized therapeutic follow-up in clinical routine. Its versatility should render it transposable to the capture of CTCs, individual or clustered, derived from various types of cancer and, by extension, to other rare circulating biomarkers.

[1]. de Bono, J. S. *et al.* Circulating tumor cells predict survival benefit from treatment in metastatic castration-resistant prostate cancer. *Clin. Cancer Res. Off. J. Am. Assoc. Cancer Res.* **14,** 6302–6309 (2008).
[2]. Vona, G. *et al.* Isolation by Size of Epithelial Tumor Cells. *Am. J. Pathol.* **156,** 57–63 (2000).
[3]. Ao, P. D., Zheng, Cote, M. D., FRCPath, FCAP, Richard J., Datar, M. P., Ph. D. ..Ram H. & Williams, P. D., Anthony. in *Circulating Tumor Cells* (ed. Fan, Z. H.) 173–182 (John Wiley & Sons, Inc, 2016).

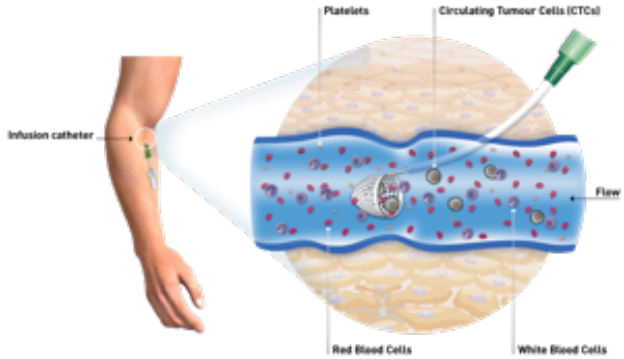

Figure 1: Illustration of the 3D microdevice fixed at the end tip of an insertion guidewire.

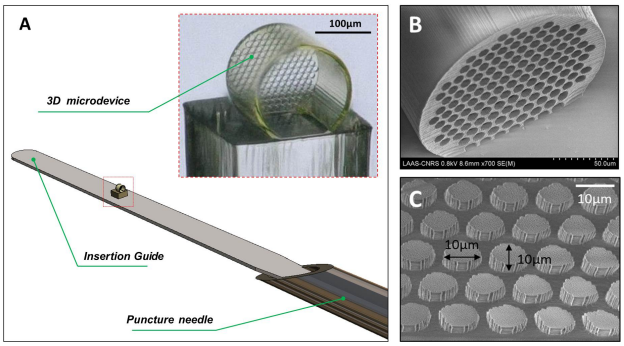

Figure 2: (A) Intravascular prototype. The 3D microdevice and a lift-base micropillar are fabricated onto a metallic strip, the latter used as an insertion guide. A commercially available and clinically used hypodermic needle (18G) is used as puncture needle. Optical bright field image (A) and corresponding electron micrograph (B and C) of the polymeric 3D microdevice fabricated using direct laser writing.

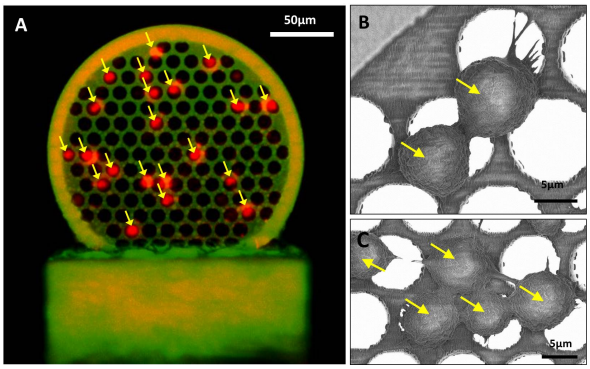

Figure 3: (A) Fluorescence image of captured cells using the intravascular prototype. (B) Scanning electron micrograph of captured cells. Arrows point out cells.