Supplementary material

1.Methods to prepare silicon nanowires

Lithographically patterned silicon nanowires were prepared by a standard photolithography and a chemical wet etching process. Firstly, polymethyl methacrylate (PMMA) A8 photoresist was spin coated on a silicon wafer (0.5 mm thick). After exposure of UV light, the silicon wafer was kept in etching solution of $NH_4F/AgNO_3$. Under the assist of Ag nanoparticle deposition, there formed a template on the surface of the silicon wafer. Then, H_2O_2/NH_4F was used to further etch the template for 10 minus and it was cleaned by H_2SO_4/H_2O_2 (70/30, v/v), deionized water and absolute ethanol. After being dried with nitrogen, the patterned silicon nanowires were obtained.

2.Preparation of chemical modified and biological functioned silicon nanowires

Firstly, using photolithography and wet etching to introduce vertically aligned silicon nanowires onto a silicon wafer. Then, the washed and dried silicon wafer was soaked in a freshly prepared toluene solution of 3-aminopropyl triethoxysilane (APTES, 1.0 wt%) for 2 hours. Afterwards, the silicon wafer (surface-bonded with mercaptopropylmethoxysilane (MPTMS)) was taken out and washed with absolute ethanol and dried with nitrogen. The modified silicon wafer with MPTMS were then soaked in a PBS solution (50 ng/mL, 200 mL) of NHS-PEG_n-Biotin (n = 1000) for 2 hours. Then the silicon wafer was taken out, washed with absolute ethanol and dried with nitrogen and stored under dry environment at 4 ^oC.

3.Fabrication of microfluidic chip

In this work, the microfluidic system used a microfluidic piping (polydimethylsiloxane (PDMS)) system. By adopting the sub-type structure, one entrance was gradually divided into eight parallel microfluidic channels (length of 20 mm, wide of 1 mm). There is a fishbone structure (with a width of 10 microns and a pitch of 10 μm) on the top of the microfluidic tubing. Height of the fishbone structure is 30 μm and the overall height of the microfluidic tubing is 100 μm.

PDMS chaotic mixers were fabricated based on a soft lithographic approach： 1). Designing and printing the masks of microfluidic channel structure and fishbone structure respectively. 2). The microfluidic channel structure was fabricated by a standard photolithographic procedure. A negative photoresist (SU8-2100, Micro Chem Corp., Newton, MA, USA) was spin-coated with a 70 μm thickness onto a 3 inches silicon wafer. After exposure to UV under the masks of microfluidic channel structure, the microfluidic channel structure was obtained. 3). The fishbone structure: another negative photoresist (100 μm, SU8-2025, Micro Chem Corp.) was spin-coated on the same wafer. Prior to UV irradiation, the mask of fishbone structure was aligned to get an accurate alignment between the prior pattern and the pattern to be imprinted. After exposure to UV under the masks of microfluidic channel structure, the fishbone structure was obtained. 4). The mold was then exposed to trimethylchlorosilane vapor for 2-3 min and then transferred to a Petri dish. 5). To prepare a 4.5 mm thick chip, a well-mixed PDMS prepolymer (GE Silicones, Waterford, NY, USA; RTV 615 A and B in 10 to 1 ration) was poured into the mold and kept in an oven at 80 ^{o}C for 48 h. The PDMS chaotic were then peeled off from the mold, and two through-holes were

punched at the fabric channel's ends for connection with the fluidic handler.

4.Assembly of microfluidic device

Prior to the blood sample test, the silicon wafer was washed with PBS and then incubated with 200 μL streptavidin solution (5.0 μM) for 1 hour. After being washed with PBS, the silicon wafer was co-incubated with 200 μL PBS that containing p-EpCAM and CKAAKN polypeptide for 1 hour and then it was washed and connected to the inlet and outlet Tygon fluid tubing to the 1 mL syringe (already installed on the syringe pump) for CTC capture.

5.Preparation of artificial CTC sample

Cell suspensions (accurate counts) with a density of 10000 / mL were prepared from BxPC3 culture dishes of non-small cell pancreatic cancer cell lines with a culture abundance of about 95%. Then, through gradient dilution by PBS, BxPC3 cell suspensions with a density of 200/20 μL could be obtained. Afterwards, 20 μL BxPC3 cell suspensions (200 cells) was added into 180 μL WBCs suspension (containing 2×10^5 cells) which was set as 200 μL artificial CTC sample.

6.Procedure for CTC capture of artificial CTC sample

After complete the set-up of the microfluidic device, 200 μL artificial sample (prepared freshly within 2 h) was injected into the system at different flow rate. As the completion of sample processing in the microfluidic chip, 100 μL paraformaldehyde solution (2 wt%) was injected into the microfluidic chip (flow rate: 1.0 mL/h) to fix the captured cells. After disassembling the chips, the silicon nanowire substrate slide was removed and slightly wash with PBS. Then, 200 μL mixing solution of fluorophore labeled

pancreatic cancer specific markers of anti-CK and anti-CD45 was dropped on the silicon nanowire substrate slide. After 24h at 4 ºC, it was washed by PBS. Then secondary antibody staining was carried out for 30 minutes while the corresponding CK antibody is the Alex488 marker and the corresponding CD45 antibody is the Alex555 marker. After cleaning, surface residue should be removed and then the silicon nanowire substrate slide was encapsulated by 80 μL Hoechest solution (stained nuclear reagents). Fluorescence imaging can be performed by fluorescent microscope. The CK+/CD45-/DAPI+ cells were defined as CTC candidates; thus, the corresponding capture efficiency could be calculated (compared with initially added 200 cancer cells).

7.Procedure for CTC of clinical pancreatic cancer patients

Taking 4 mL whole blood as an example, in this work, the method of gradient density centrifugation was adopted to preliminarily purify the blood of cancer patients. Firstly, 4 mL PBS was added into 4 mL whole blood solution and mixed well. Then, the diluted blood sample was slowly added to a centrifuge tube (15 mL) while 4 mL gradient density centrifugation solution (1077) had been added into the tube already. Centrifugation was performed using 300 g for 40 minutes. Afterwards, serum was removed and about 2-4 mL of peripheral blood mononuclear cells (PBMCs) was collected. Then, the collected PBMCs was further centrifuged at 400 g for 5 minutes. Afterwards, the supernatant was removed and the PBMCs were washed with 2 mL PBS. After the supernatant was removed by centrifugation, a mixture solution (400 μL) of freshly prepared anti-CK (5 μM), Vimentin (5 μM) and anti-CD45 (5 μM) was added. PBMCs were incubated for 45 minutes. After cleaning, they were set at 400 μL PBS.

Then the CTC capture procedure and staining method were performed following procedure of artificial CTC sample. Here, the CK+/CD45-/DAPI+ cells and Vimentin+/CD45-/DAPI+ cells were defined as CTC candidates to calculate corresponding capture efficiency.

8.Cell viability assay for released cells

With the advantages of the high releasing efficiency presented by this method, the cell viability could be conveniently detected by the Vi-CELLTM XR (Cell Viability Analyzer, Beckman Coulter). Following the capture procedure described above, the substrate-modified cancer cells are transfer into a 4 ^{0}C refrigerator for 30 min. Then the substrate was gently rinsed with total 0.5 mL PBS for 3 times and the cells suspension solutions were collected. The released cancer cells on each silicon nanowires were finally prepared to a 1.0 mL-volume cell suspension, which was then carefully transferred to a sample container especially for the Vi-CELLTMXR. The cell viability was analysed as a triplicate.