Supporting information

A multilayer polymer-film inertial microfluidic device for high-throughput cell concentration

Nan Xiang, Rui Zhang, Yu Han and Zhonghua Ni

School of Mechanical Engineering, and Jiangsu Key Laboratory for Design and Manufacture of Micro-Nano Biomedical Instruments, Southeast University, Nanjing, 211189, China.

Table of Contents

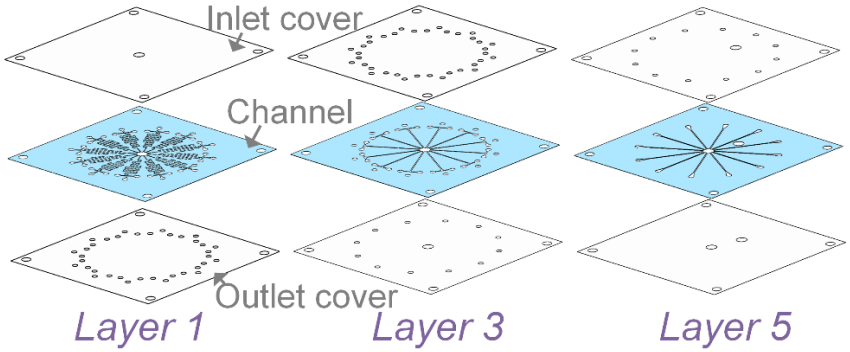

Figure S1. Three-sheet structure of channel layers (#1, #3, and #5) which is fabricated by enclosing the cut-through channels on the polymer film with the inlet and outlet covers.

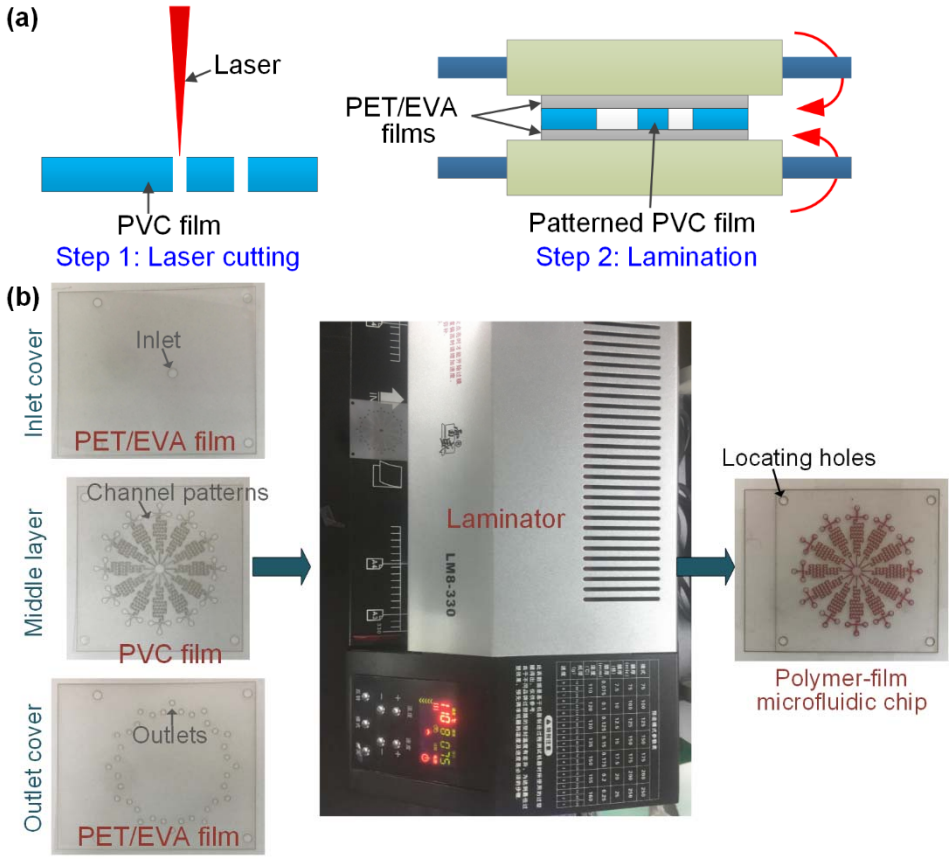

Figure S2. (a) Methods for fabricating the channel layers in our MPIM device. (b) Fabrication process for sealing the through channels on a polyvinyl chloride (PVC) film between two sheets of polyethylene terephthalate films with thermal sensitive ethylene-vinyl acetate copolymer (PET/EVA film) using the roll-to-roll lamination.

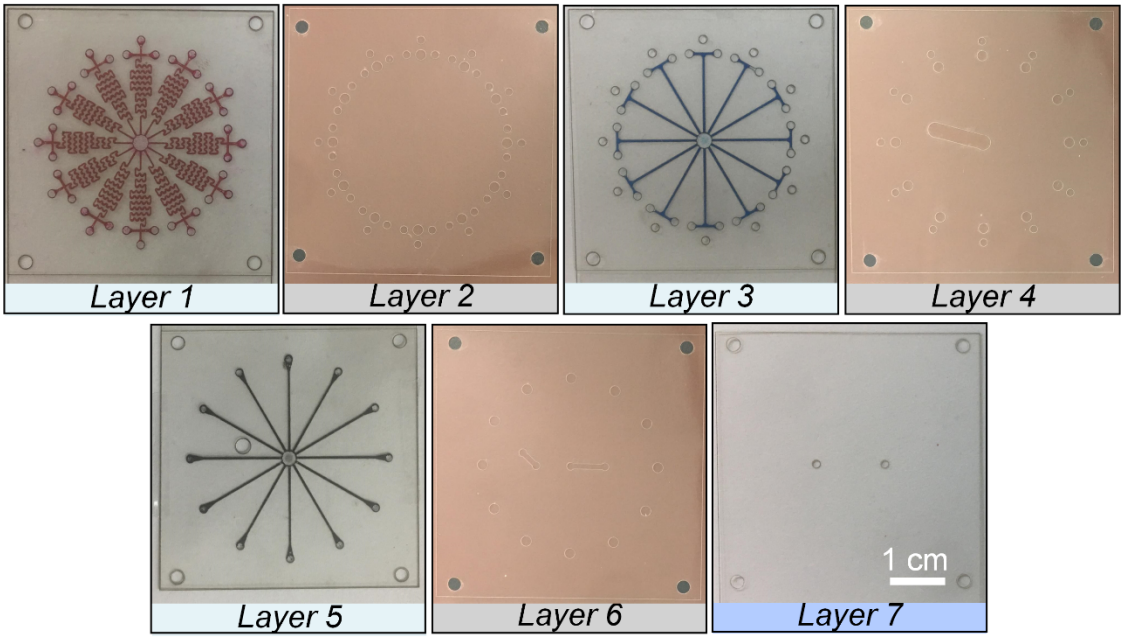

Figure S3. Photographs of the finished each layer. The channels were filled with ink for clear visualization. Before assembly, one side of the tape remained protected by the plastic backing (the brown film). It is worth noting that observation windows (cut-through holes) were designed on the adhesive layers at the positions before the cross-shaped outlet systems for better visualizing the cell migration in serpentine channels.

Figure S4. Assembly process of our MPIM device after fabricating each layer. The whole assembly process can be completed within three main steps.

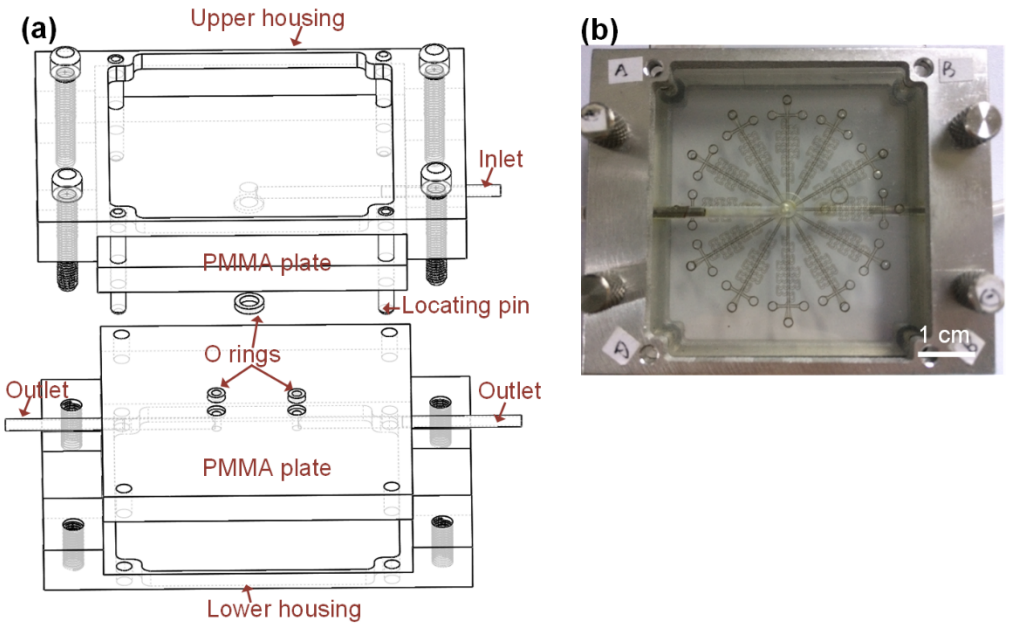

Figure S5. (a) Schematic diagram illustrating the structure of the housing for the chip-to-world interface. (b) Photograph of the custom housing equipped with our MPIM device.

Figure S6. Photograph of our MPIM device showing a good optical transparency although 13 layers have been stacked in vertical direction.

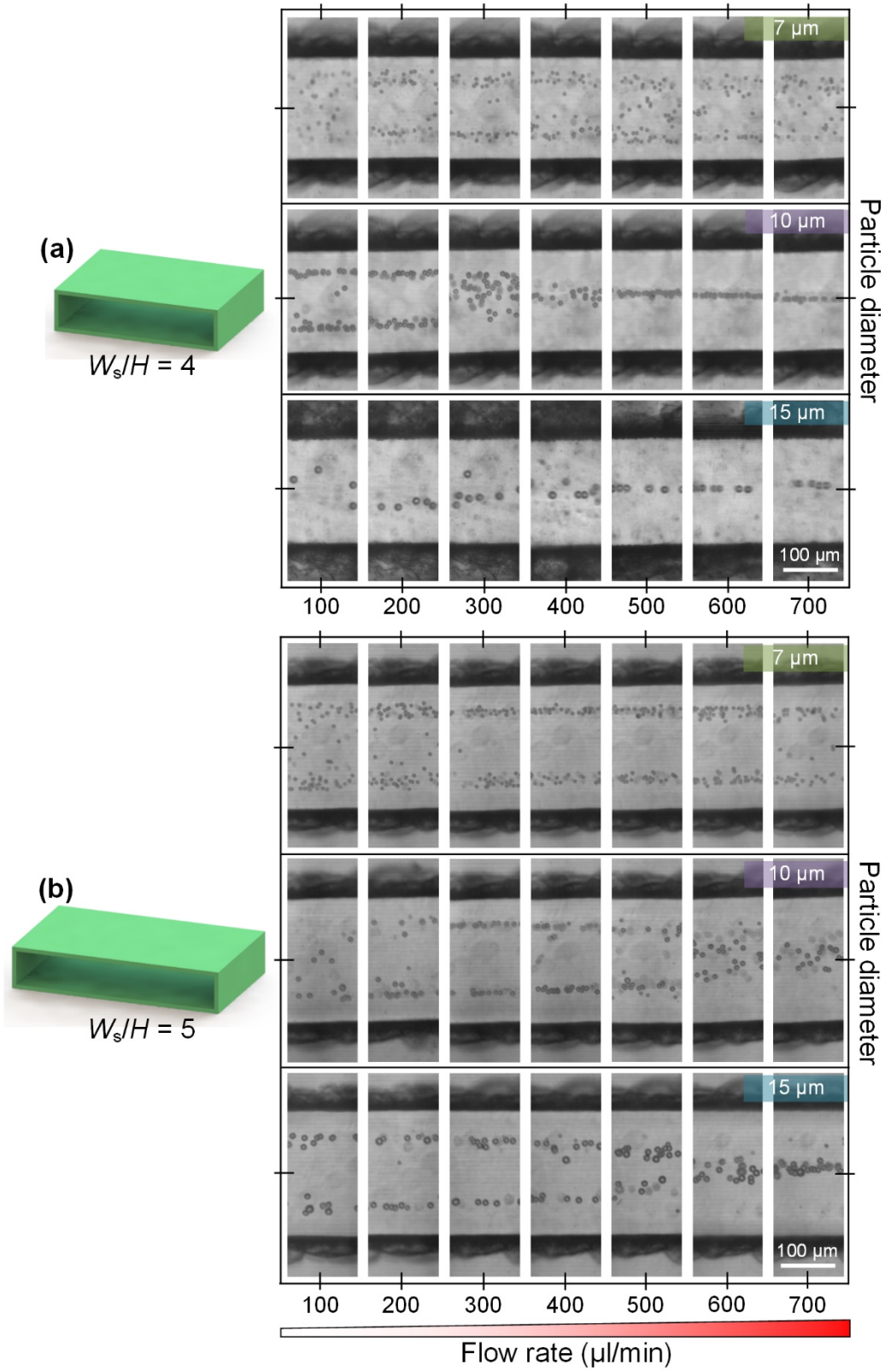

Figure S7. Focusing maps of 7 μm, 10 μm and 15 μm particles in serpentine channels with W_S/H of 4 (a) and 5 (b) at different flow rates ranging from 100 μl/min to 700 μl/min (with an interval of 100 μl/min). The particle focusing dynamics were captured near the cross-shaped outlet.

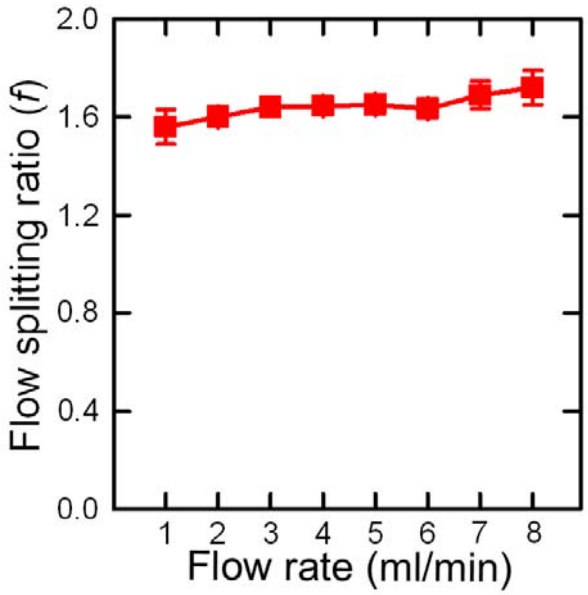

Figure S8. Flow splitting ratios (f=v_{blank}/v_{target}) of our MPIM device at different flow rates of 1~8 ml/min. The v_{blank} and v_{target} are respectively the volumes of the particle-free blank fluids and the volumes of target concentrated samples.

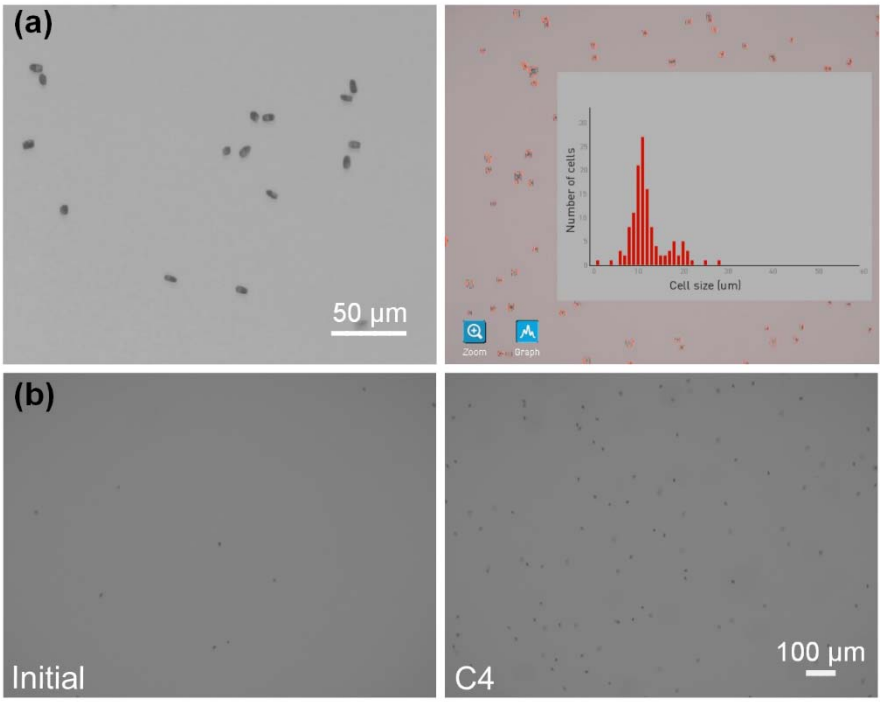

Figure S9. (a) Photograph and the measured size distribution of the green microalgae (GY-H1 Platymonas helgolandica tsingtaoensis). The average circularity of the green microalgae was measured to be ~0.5. (b) The microscopic images of the initial sample and the target sample collected after the C4 concentration step.

Table S1. Detailed dimensions of the employed serpentine channels

Channel types	R_S (mm)	W_S (mm)	R_L (mm)	W_L (mm)	W_S/H	W_L/W_S
I	0.1	0.15	0.36	0.30	3	2
II	0.1	0.20	0.50	0.40	4	2
III	0.1	0.25	0.66	0.50	5	2

Section S-1. Design guideline for our serpentine channels.

For the three channel designs in Table S1, the ratio of W_L to W_S and the innermost radius R_S of the small turn were respectively fixed at 2 and 0.1 mm. The using of small innermost radius R_S = 0.1 mm could induce strong Dean flow for speeding up the inertial focusing process as the Dean number for quantifying the Dean flow strength is proportional to $R^{-1/2}$.[1] The innermost radius R_L of the large turn was designed to make the large turn smoothly connect with the small turn. For each channel design, the W_S is constant at a specific value while the large turn smoothly connects with the small turn and has variable widths (the largest width of the large turn was designed to be W_L). To increase the throughput of our MPIM device, the W_S were designed to be 0.15 mm, 0.20 mm and 0.25 mm, respectively.

To realize the focusing of the flowing particles/cells, the channel height H needs to satisfy the criterion $H \leq a_p/0.07$ (a_p is the particle diameter)[1] For the current work, the channel height was fixed to be ~50 μm so that the particles/cells with their diameters larger than 3.5 μm can be successfully focused. For more comprehensive guidelines on how to design a curving channel, the readers can refer to the topic review on inertial microfluidics.[2]

Section S-2. Detailed descriptions on the process for fabricating the MPIM device.

The channel geometries were first patterned by cutting a through groove within the polyvinyl chloride (PVC) film with a thickness of 65 μm. After that, the patterned PVC film was sandwiched between two sheets of polyethylene terephthalate films with thermal sensitive ethylene-vinyl acetate copolymer (PET/EVA, a total thickness of 100 μm), and sealed using a desktop laminator (LM8-330, Rayson) under the determined lamination temperature and rolling speed. The inlets and outlets for these layers were respectively cut on the upper and lower PET/EVA films.

A single sheet of PVC film patterned with outlet holes was employed as the outlet layer. For the adhesive layers, the pressure sensitive PET double-sided tape with a thickness of 90 μm was used. The patterns on all these layers were cut using a laser machine (TH-UV200A, Tianhong) equipped with an UV laser (AWAVE 355-10 W-30 K, Advanced Optowave Corporation). More details about the process parameters concerning the fabrication method can refer to our previous work.[3]

All the employed materials are inexpensive and can be directly from the local market. The custom fixture for assembling each layer was manufactured in polymethyl methacrylate (PMMA) using a machining system. Four small stainless steel pins together with the locating holes in each layer were employed for aligning and stacking different layers.

Section S-3. Detailed descriptions on the assembly process of our MPIM device after fabricating each layer.

We realized the quick assembly of our MPIM device within three main steps by respectively layering different channel layers and adhesive double-sided tape layers in vertical direction (see Figure S4). In step one, we first loaded the patterned double-sided tape (layer #2) with its bottom face protected by the plastic backing into the fixture and then attached the layer #1 to the top of the above double-sided tape. The attached two layers (layers #1 and #2) were turned over and the plastic backing on the double-sided tape was peeled. Then, we attached the layer #3 to the bottom face of the double-sided tape (layer #2) to form the upper part of our MPIM device. Repeating the above procedure, we further assembled the layers #5, #6 and #7 to form the lower part of our MPIM device (step two). Finally, the upper and lower parts are adhered using the adhesive layer #4. After finishing the above assembly procedure, the entire device was compressed by rolling over 3 times using a plastic rolling pin on a bench top to make sure that the tight bonding of every layer. Through using our locating holes and corresponding custom fixture, an alignment accuracy as high as 10 μm can be achieved.

Supplementary Video S1. Particle migration in the MPIM device.

References

(1) Di Carlo, D.; Irimia, D.; Tompkins, R. G.; Toner, M. Continuous inertial focusing, ordering, and separation of particles in microchannels. *Proceedings of the National Academy of Sciences* **2007**, *104*, 18892-18897.

(2) Di Carlo, D. Inertial microfluidics. *Lab on a Chip* **2009**, *9*, 3038-3046.

(3) Zhang, X.; Huang, D.; Tang, W.; Jiang, D.; Chen, K.; Yi, H.; Xiang, N.; Ni, Z. A low cost and quasi-commercial polymer film chip for high-throughput inertial cell isolation. *RSC Advances* **2016**, *6*, 9734-9742.