

Electronic Supplementary information for
Size-selective filtration of extracellular vesicles with a movable-layer device

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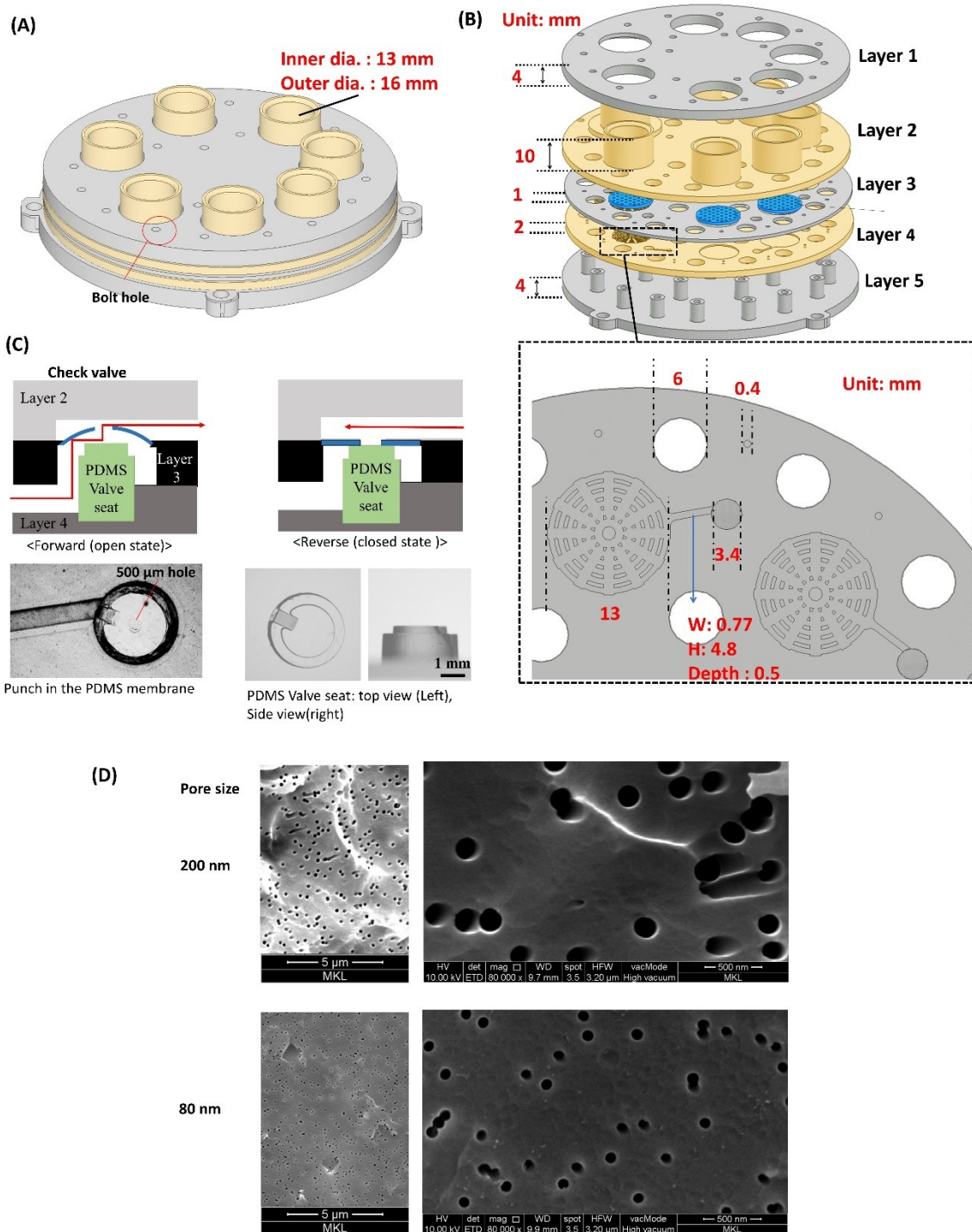


Fig. S1. (A) Schematic of a chip. (B) Exploded view of the chip that shows multi-layers. Layers 1, 2, 4, and 5 are 3D-printed materials. Layer 3 is a silicone gasket with an elastomeric membrane. Porous nanofilter membranes are positioned between layers 2 and 3. To guide their alignment, layer 5 had relatively large-size post structures (outer diameter: 6 mm) and layers 2–4 had holes. These posts and holes facilitated the alignment of the layers and also were used to assemble/disassemble the layers using screws. The layers of the device were reused and the filter membranes were disposable. (C). Open and closed states of a check valve and photos that show elastomeric membrane and PDMS valve seat. (D) SEM images of porous nanofilter membranes

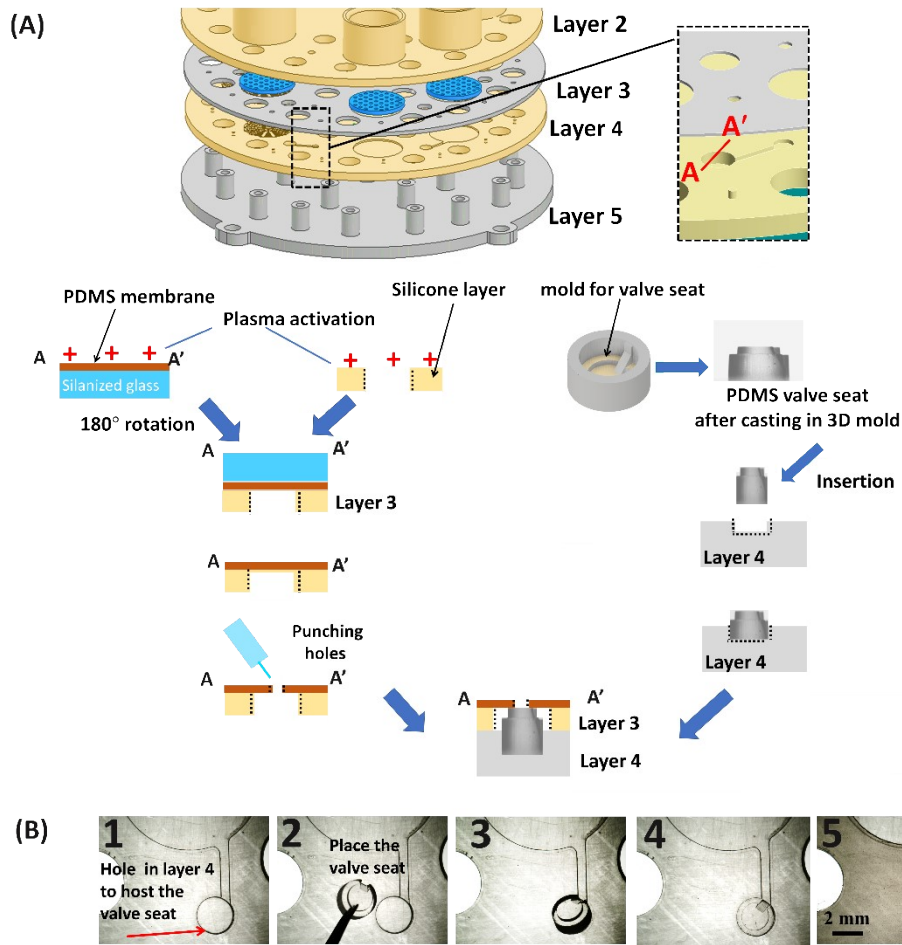


Fig. S2. Schematic illustration for the fabrication and assembly of the PDMS membrane and check valve. (A) PDMS membrane fabrication and assembly with the silicone layer. The PDMS membrane was spin-coated on a silanized (trichloro (1H,1H,2H,2H-perfluorooctyl) silane, Model 448931, Sigma Aldrich) glass plate with a thickness of 50 μm , cured at 120 $^{\circ}\text{C}$ for 20 min, and plasma-treated at 100 W for 30 s (Cute-1MP, FemtoScience) followed by bonding with a silicone gasket (LS005-3TS-005, VisionLab Science). The hole was punched on the PDMS membrane using a 500- μm -diameter biopsy punch (Harris Uni-Core-0.5). A valve seat, which was made of a PDMS material cast by a 3D printed mold, was inserted into layer 4. (B) Loading process of the PDMS valve seat and assembly of the layers 3 and 4. In steps 1–4, the valve seat was manually loaded on the layer 4. In step 5, the layer 3 is placed on the layer 4.

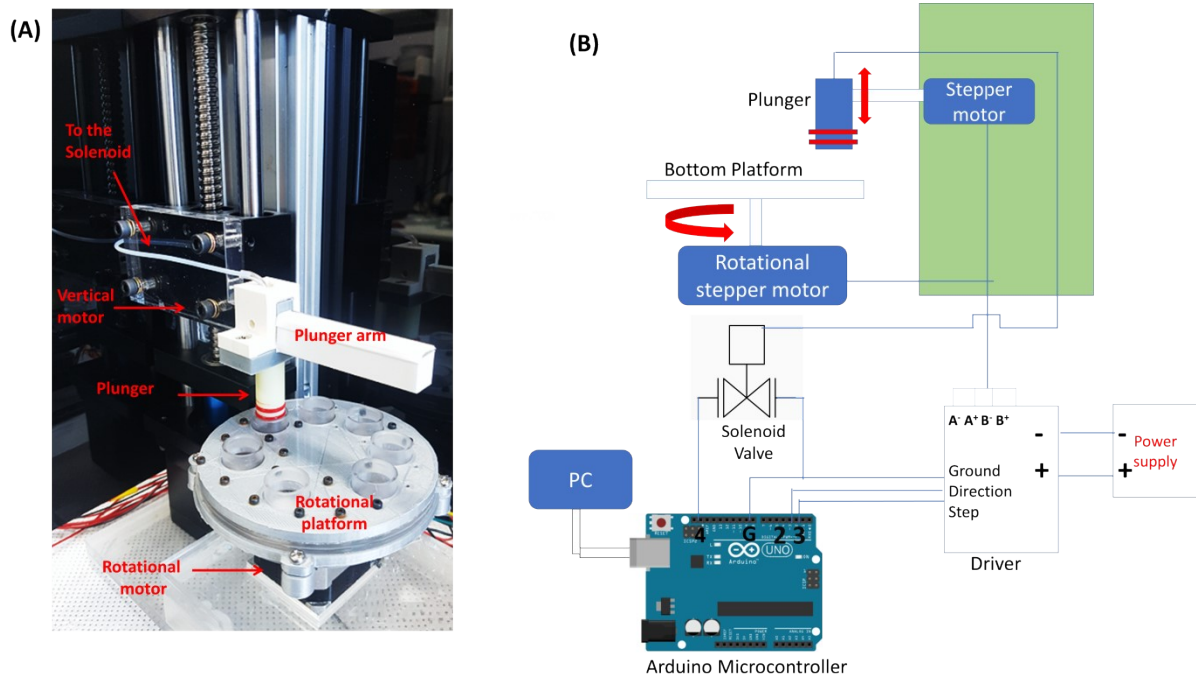


Fig. S3. (A) Photograph that shows the device and its peripheral setup. (B) Block diagram that shows control units for microcontroller, a solenoid valve, and stepper motors. The motors are directly connected to a driver, which amplifies the low-current to drive the motors. To regulate the motor's motion, the driver is connected to the microcontroller through 3 ports for direction, steps, and ground. Direction and step port is connected to 2 and 3 digital ports of the microcontroller. The solenoid valve is connected to gate 4 of the microcontroller. Then, a code is uploaded into the microcontroller, which is connected to a PC. The driver is powered by an external power supply and the microcontroller is powered through its USB connection from the PC.

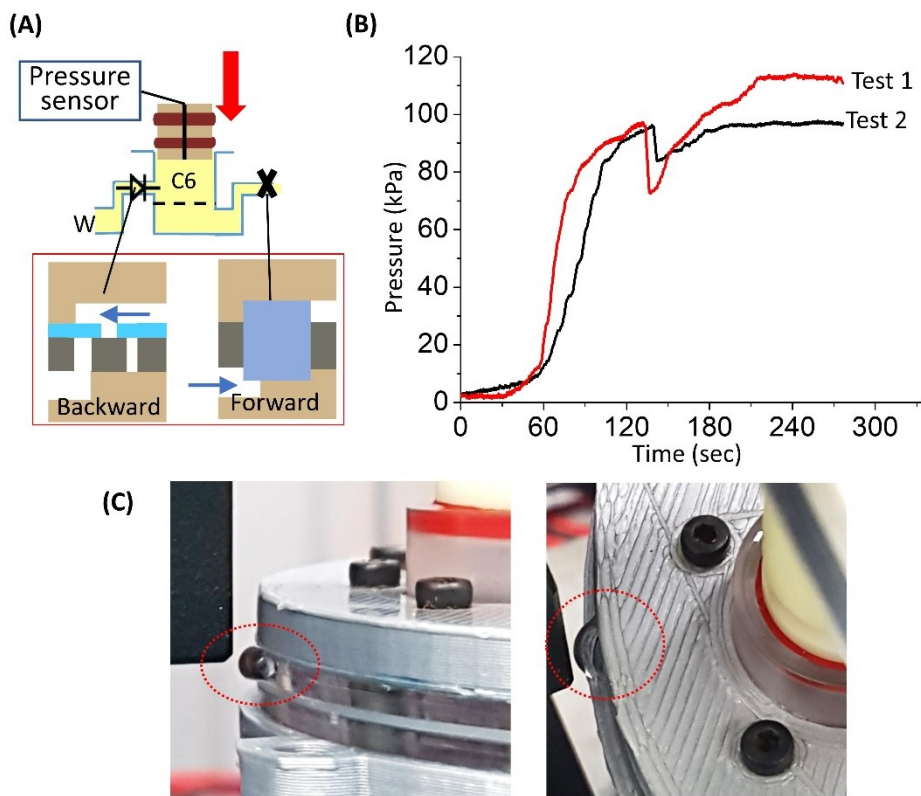


Fig. S4. Upper limit of pressure of the device/check valve prior to leakage. (A) Schematic that shows the experimental set-up. Chamber C6 is pressurized. The upstream of C6 (backward direction) is connected by a check valve and the downstream of C6 (forward direction) is blocked by a PDMS structure. (B) Pressure profiles in C6. Plunger with a constant speed of $500 \mu\text{m/s}$ pressurizes the chamber for 270 s. The leakage occurs between the layers when the pressure reaches at $\sim 95 \text{ kPa}$. The pressure decreases abruptly at the moment of leakage ($\sim 130 \text{ s}$) and then increases due to the continuous movement of the plunger. No breakage of the check valve and filter membrane is observed. (C) Photographs of the device that shows the leakage.