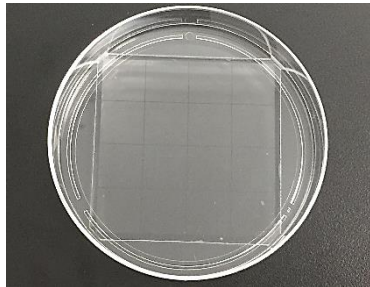


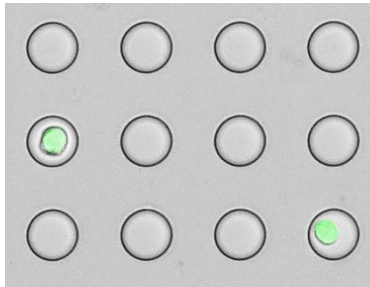
Quick-Start Protocol for Million Microwell Device (MMD)

Preparation



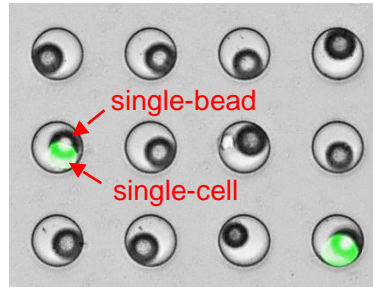
- Transfer one MMD to a 100 mm diameter Petri Dish with the microwell side facing up (the microwell openings face down in the packaging)
- Treat the surface of MMD with plasma for 1-2 min for hydrophilicity

Single-Cell Capture



- Pipette 2 mL of 30mg/mL BSA/PBS solution onto MMD; wash away unabsorbed BSA after 1 h of incubation
- Add cell suspension (10^5 /mL); settle down for 10 min (~10% microwells will capture cells); wash away uncaptured cells

Bead Capture



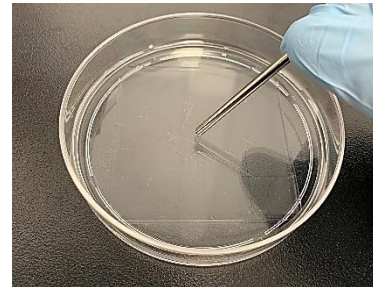
- Place a magnet at the bottom of the MMD
- Add the barcoded magnetic bead suspension ($\geq 10^6$ /mL); settle down for 10 min (most microwells will capture beads); wash away uncaptured beads

Single-Cell Lysis



- Add cold cell lysis buffer onto the MMD surface; incubate for 10 min; add fresh lysis buffer (optional)
- The magnet at the bottom of MMD will increase the stability of magnetic beads within the microwells

Bead Collection



- Remove the bottom magnet
- Place a magnetic bar on the surface of MMD; move the magnetic bar across the entire surface of MMD to collect the magnetic beads with captured mRNA

Bead Transfer



- Place a PCR tube containing PBS on the top of a strong magnet
- Insert the magnetic bar containing magnetic beads into the PBS; strongly stir up and down to transfer beads to the PCR tube

Applications

- Single-cell RNA-sequencing
- Single-cell capture