iBiochips

Protocol for

PicoWells In 6-Well Plate

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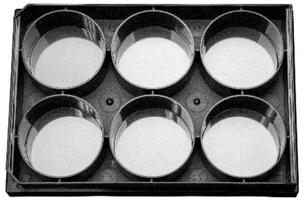
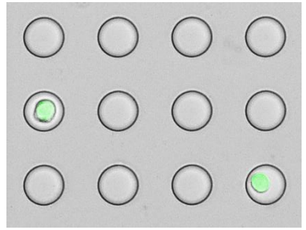
Plasma Treatment



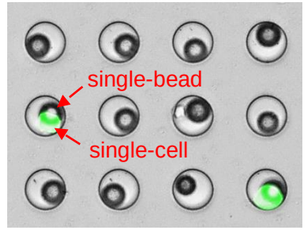
Take the lid off and put the *PicoWells In 6-Well Plate* in a plasma cleaner. Treat the surface of picowells with plasma for 1-2 min to change the surface properties of PDMS picowells from being hydrophobic to hydrophilic.

Single-Cell Capture



Pipette 1-2 mL of 30 mg/mL BSA/PBS solution into each well; wash away unabsorbed BSA after 1 h of incubation; add cell suspension (10^5/mL); settle down for 10 min (~10% picowells will have single cells); wash away uncaptured cells.

Single-Bead Capture

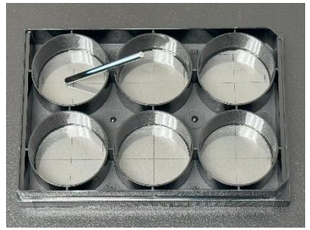


Place a Disk Magnet at the bottom of the picowells. Add the barcoded magnetic bead suspension (≥ 10^6/mL) and settle down for 10 min to allow most picowells to capture single beads. Gently wash away the uncaptured beads.

Single-Cell Lysis

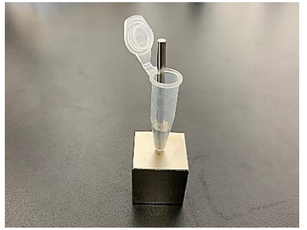
Add cold cell lysis buffer onto the surface of picowells; incubate for 10 min; add fresh lysis buffer (optional). The Disk Magnet at the bottom will increase the stability of magnetic beads within the picowells.

Beads Collection



Remove the Disk Magnet. Place a Cylinder Magnet on the surface of picowells, and move the Cylinder Magnet across the entire surface of picowells to collect the magnetic beads with captured mRNA.

Beads Transfer



Place a PCR tube containing PBS on top of a strong Cube Magnet. Insert the Cylinder Magnet containing magnetic beads with captured mRNA into the PBS; strongly stir up and down to transfer beads to the PCR tube.

Applications

- Single-Cell/Bead Isolation
- Single-Cell/Bead Imaging
- Single-Cell RNA-Sequencing

The Disk Magnet, Cylinder Magnet, and Cube Magnet need to be ordered separately.

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