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Fabrication of alginic acid sodium salt -based MWC-chip V.1

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We use this protocol and it's working

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Abstract

PDMS micromolds for MTS generation and microfabrication of the MWC-Chip.

The creation of PDMS male mold

- 1 The alginic acid sodium salt-based MWC-chips were fabricated using a micromolding method. Specifically, the layout of the chips was initially designed using AutoCAD software. A polymethyl methacrylate (PMMA) female mold was fabricated on a PMMA plate (CREROMEM, 200 mm × 300 mm × 2 mm) using a precision micromilling machine (JINGYAN) equipped with a parabolic spiral-flute bit (WeiTol, $\Phi 0.5$ mm). Subsequently, a polydimethylsiloxane (PDMS) male mold was created by pouring PDMS prepolymer (ratio 1:10, cross-linking agent: elastomer) into PMMA female mold and solidifying the PDMS prepolymer at 80 °C for 2 hours.

The generation of MWC-Chip

- 2 The released PDMS mold was then covered with an alginic acid sodium salt solution (2% w/v) (Sigma). The alginic acid sodium salt-based MWC-chips were formed by gelation in the CaCl_2 solution (5% w/v) (Sinopharm Chemical Reagent) at 25 °C for 1 minutes and subsequently released from the PDMS mold. The MWC-chips were gently placed into a 3.5 cm² plate. Each MWC-chip with a hydrogel substrate measured 2 × 2 cm² and contained approximately 15,000 microwells with dimensions of diameter: 80 μm , height: 40 μm and pitch: 80 μm . The resulting MWC-chips were sterilized thoroughly in the purified PBS supplemented with penicillin-streptomycin at a concentration of 1% w/v, followed by exposure to ultraviolet light for over 30 minutes. Finally, these sterilized MWC-chips were stored at 4 °C for subsequent generation of micro-tumorspheres (MTSs).

Protocol references

Brandenberg, N. *et al.* High-throughput automated organoid culture via stem-cell aggregation in microcavity arrays. *Nat Biomed Eng* **4**, 863-874 (2020).