Microfluidics: vertical chip

iGEM Pasteur Paris team - Institut Pasteur

Axelos Etienne Dejoux Alice Dumont Claire Ehret Antoine
Fyrillas Andreas Ghaneme Aymen Guerassimoff Léa
Jaoui Samuel Picaud-Lucet Emma Porte Sarah
Redheuil Ellyn Richard Charlotte Starck Thomas
Thomas Florian Trang Kelly Madelénat Manon
Paillarès Éléa Naccache Jonathan Sachet Gabriela

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Introduction

The vertical chip was one of the designs for the support of the final experiment, that would serve as a proof of concept. The idea is to isolate the modified bacteria from the neurons using a membrane. To simplify the membrane's integration in a PDMS chip, a vertical design instead of a microchannel horizontal one was proposed.

I. Mold

The mold was made of aluminium according to the following plan (Figure 1).

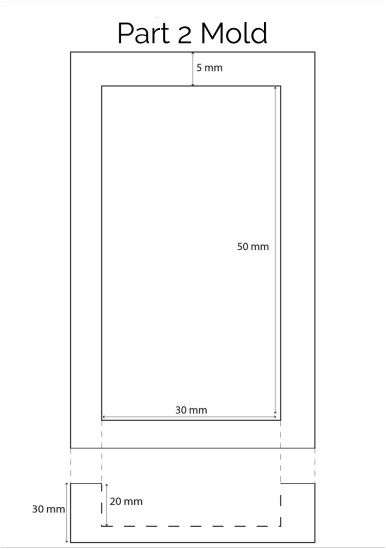

Figure 1 – *Vertical chip mold plan*

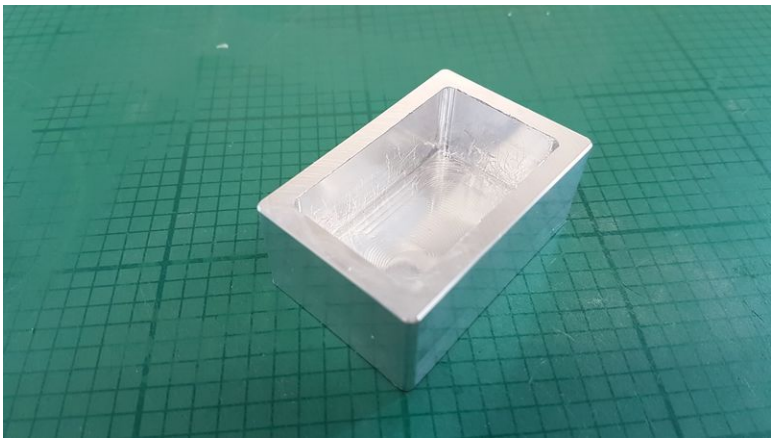

Figure 2 – *Vertical chip mold*

II. PDMS Vertical Chip Fabrication

II.1. Materials

- Molds
- Polycarbonate gold-coated membrane filters, 0.4 micron, 13mm diameter (Sterlitech) or polymerized membrane (see Microfluidics : Membranes, **link to protocol**)
- Biopsy puncher (Kai biopsy puncher 2 mm and 4 mm)
- Conductive silver paste (MG Chemicals 8330S-21G)
- Tissue culture dish (TPP 93060, 53 mm internal diameter)

Refer to Microfluidics : General Protocols (**Link**) for further materials.

II.2. Procedure

Step 1 : Prepare 15 g of PDMS monomer using section 1 of Microfluidics : General Protocols (**Link**).
Step 2 : Demold the PDMS layer following section 2 of Microfluidics : General Protocols (**Link**). Ignore step 2.
Step 3 : Cut PDMS layer in two halves.
Step 4 : Drill a hole in the center of each half with a 4 mm biopsy puncher. Drill holes in one of the two layer with a 2 mm biopsy puncher (see figure 3).

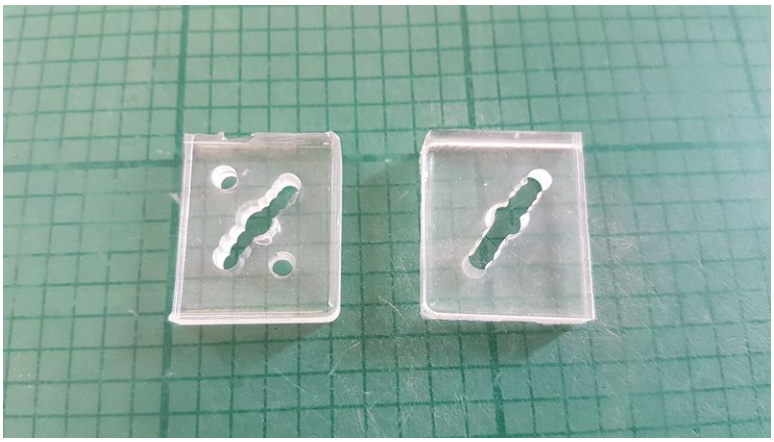

Figure 3 – *Two halves of PDMS layer. Left : upper layer. Right : bottom layer*

Step 5 : Prepare 2 mL of conductive silver paste following the manufacturer's instructions (Preheat parts A and B in stove at 70 degrees Celsius, put 0.5 mL of part A and 0.5 mL part B in a Petri dish, mix with the toothpick).
Step 6 : Apply a path of silver paste on bottom layer starting from the center hole and going outwards of the layer (figure 4). Deposit one half of a membrane filter on center hole. Put in stove at 70 degrees Celsius for 2 hours.

Figure 4 – *Assembled vertical chip with gold-coated membrane filter*

Step 7 : Bond the two layers together (figure 4) according to section 3 of Microfluidics : General Protocols (**Link**).

Step 8 : Take the bottom of a tissue culture dish and deposit the other half of the membrane filter in the center. Apply silver paste and put in stove at 70 degrees Celsius for 2 hours (figure 5).

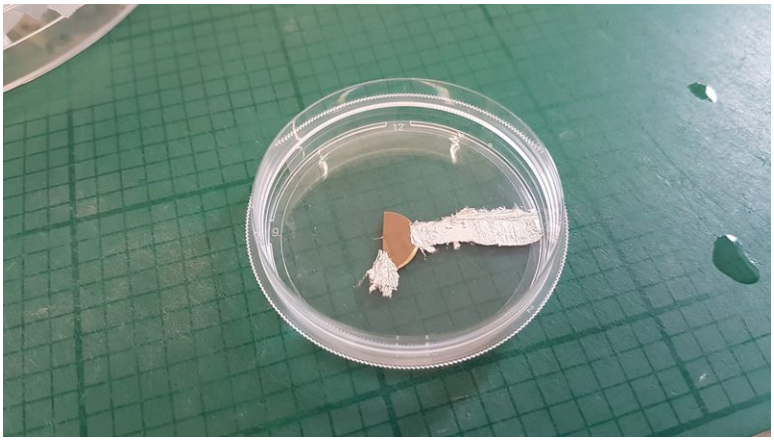

Figure 5 – *Tissue culture dish with membrane and silver paste*

Step 9 : Bond prepared tissue culture dish with product of step 7 (figure 6), bottom layer (figure 3) facing the dish, refering to section 3 of Microfluidics : General Protocols (**Link**).

Figure 6 – *Bonded vertical chip*

III. Chip Sterilization

Unwanted living organisms in microfluidic chips can be a big deal, especially when these chips have to stay for 3 days filled with cultur medium in an incubator. The chips need to be exposed to UV rays in order to eliminate these unwanted organisms. We took extra securitiy measures, because we also needed to transport our chips from Institut Curie's lab at IPGG to Institut Pasteur.

III.1. Materials

- Bonded chip, product of section 2.
- Big Petri dish (150 mm diameter)

- Gloves (Kimtech PFE)
- UV curing unit (DWS)
- Wrapfilm for food use (Ecopla France film pro)
- Parafilm (Bemis parafilm "M")
- Fridge

III.2. Procedure

Step 1 : Open dishes containing bonded chips and put them in the UV curing unit with their corresponding lid.

Figure 7 – *Dishes and their lid in UV curing unit*

Step 2 : Expose to UV rays for 20 minutes.
Step 3 : With gloves, put exposed dishes in a big Petri dish.
Step 4 : Seal Petri dish with parafilm.
Step 5 : Cover Petri dish with 3 layers of wrapfilm.
Step 6 : Expose 15 minutes to UV rays.
Step 7 : Cover Petri dish with 2 additional layers of wrapfilm.
Step 8 : Store in fridge.