

Materials List for:

Anatomically Inspired Three-dimensional Micro-tissue Engineered Neural Networks for Nervous System Reconstruction, Modulation, and Modeling

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Materials

Name	Company	Catalog Number	Comments
Laser cutter	Universal Laser Systems	PLS4.75	Used to fabricate the laser-cut micro-channel mold.
Laser-cut micro-column fabrication device	Custom-made	-----	Contact our research group if interested. Dimensions and blueprints provided in the manuscript.
Screws	-----	-----	#4-40 with a thread diameter of 3.05 mm
Nuts	-----	-----	#4-40 with a thread diameter of 3.05 mm
Acupuncture needle (180 µm diameter)	Lhasa Medical	sj.16X40	The diameter may be varied according to the desired size for the micro-column lumen.
Petri dish	Fisher	08772B	
Dulbecco's phosphate buffered saline (DPBS)	Invitrogen	14200075	
Polystyrene disposable serological pipet	Fisher	13-678-11D	
Agarose	Sigma	A9539-50G	
Capillary tube (398 µm diameter)	Fisher	21170D	The diameter may be varied according to the desired size for the micro-column shell.
Hot plate	Fisher	SP88857200	
Magnetic bar	Fisher	1451352	
Micropipette	Sigma	Z683884-1EA	
25 mm gauge needle	Fisher	14-826-49	
Microscalpel	Roboz Surgical	RS-6270	
Scissors	Fine Science Tools	14081-09	
Forceps	World Precision Instruments	501985	
Hot bead sterilizer	Sigma	Z378550-1EA	
Stereoscope	Nikon	SMZ800N	Used for all dissection steps and for micro-TENN fabrication.
Rat tail type I collagen	Corning	354236	Maintain at 4 °C and remove only when needed. Use ice to preserve its temperature when in use.

Microcentrifuge tube	Fisher	02-681-256	
Mouse laminin	Corning	354232	Maintain at 4 °C and remove only when needed. Use ice to preserve its temperature when in use.
Neurobasal medium	Invitrogen	21103049	Basal medium for the culture of pre-natal and embryonic neuronal cells. Store at 4°C and warm at 37 °C before use.
Sodium hydroxide (NaOH)	Fisher	SS2661	
Hydrochloric acid (HCl)	Fisher	SA48-1	
Litmus paper	Fisher	09-876-18	
Hank's balanced salt solution (HBSS)	Invitrogen	14170112	Store at 4 °C.
0.25% Trypsin-EDTA	Invitrogen	25200056	Store at -20 °C and warm at 37 °C before use.
Bovine pancreatic deoxyribonuclease (DNase) I	Sigma	10104159001	Store at -20 °C and warm at 37 °C before use.
B-27 Supplement	Invitrogen	12587010	Supplement added to Neurobasal medium for the culture of hippocampal and cortical neurons. Store at -20 °C and warm at 37 °C before use.
L-glutamine	Invitrogen	35050061	Store at -20 °C and warm at 37 °C before use.
Sprague Dawley embryonic day 18 rats	Charles River	Strain 001	
Pasteur pipette	Fisher	22-042816	
15 mL centrifuge tube	EMESCO	1194-352099	
Vortex	Fisher	02-215-414	
Centrifuge	Fisher	05-413-115	
Hemocytometer	Fisher	02-671-6	
Objet30 3D-Printer	Stratasys	-----	Used to fabricate the pyramidal micro-well molds.
3D-printed pyramidal well mold	Custom-made	-----	Contact our research group if interested. Dimensions and blueprints provided in the manuscript.
Polydimethylsiloxane (PDMS) and curing agent	Fisher	NC9285739	Comes as kit with elastomer and curing agent. Use inside a chemical fume hood.
Funnel	Fisher	10-348C	
1 ml pipette bulb	Sigma	Z509035	
Micro-spatula	Fisher	S50821	
12-well culture plate	EMESCO	1194-353043	
Oven	Fisher	11-475-154	
Incubator	Fisher	13 998 076	
AAV1.Syn.GCaMP6f.WPRE.SV40	UPenn Vector Core	36373	Store at -80°C. Commercially available adeno-associated virus (AAV) with the GCaMP6f calcium indicator.
Formaldehyde 40%	Fisher	F77P-4	Formaldehyde is a toxic compound known to be carcinogenic, and must be disposed of in a separate container.

Triton X-100	Sigma	T8787	Non-ionic surfactant used to permeabilize cell membranes.
Horse serum	Gibco	16050-122	
Mouse anti-Tuj-1/beta-III tubulin primary antibody	Sigma	T8578-200UL	Store at -20°C.
Rabbit anti-synapsin 1 primary antibody	Synaptic Systems	106-001	Store at -20°C.
Donkey anti-mouse 568 secondary antibody	Invitrogen	A10037	Store at 4°C.
Donkey anti-rabbit 488 secondary antibody	Invitrogen	A21206	Store at 4°C.
Hoechst 33342, Trihydrochloride	Invitrogen	H3570	Store at 4°C. Hoechst is a known mutagen that should be treated as a carcinogen. Therefore, it must be disposed of in a separate container.
A1RSI Laser Scanning Confocal Microscope	Nikon	-----	Used for taking the confocal reconstructions of immunolabeled constructs.
Eclipse Ti-S Microscope	Nikon	-----	Used for taking the phase-contrast images. With digital image acquisition using a QIClick camera interfaced with Nikon Elements Basic Research software (4.10.01).
High-speed Fluorescence Microscope	Nikon	-----	Nikon Eclipse Ti microscope paired with an ANDOR Neo/Zyla camera for calcium imaging.
NIS Elements AR 4.50.00 Software	Nikon Instruments	-----	Used to identify calcium transients from the recordings taken with the high-speed fluorescence microscope.