

# 3D Bioprinting of Murine Cortical Astrocytes for Engineering Neural-Like Tissue

 Bruna A. G. de Melo<sup>1</sup>, Elisa M. Cruz<sup>1</sup>, Taís N. Ribeiro<sup>1</sup>, Mayara V. Mundim<sup>1</sup>, Marimelia A. Porcionatto<sup>1</sup>
<sup>1</sup>Department of Biochemistry, Escola Paulista de Medicina, Universidade Federal de São Paulo

## Corresponding Author

 Bruna A. G. de Melo  
 g.melo.bruna@gmail.com

## Citation

 de Melo, B.A.G., Cruz, E.M., Ribeiro, T.N., Mundim, M.V., Porcionatto, M.A. 3D Bioprinting of Murine Cortical Astrocytes for Engineering Neural-Like Tissue. *J. Vis. Exp.* (173), e62691, doi:10.3791/62691 (2021).

## Date Published

July 16, 2021

## DOI

10.3791/62691

## URL

jove.com/video/62691

## Materials

Name	Company	Catalog Number	Comments
3D Bioprinter	3D Biotechnology Solutions		Extrusion-based bioprinter
Blunt-tip forceps	Integra Miltex	6--30	Forceps for brain dissection previously sterilized
Bovine serum albumin	Sigma-Aldrich	9048-46-8	Protease free, fatty acid free, essentially globulin free
CaCl <sub>2</sub>	Sigma-Aldrich	10043-52-4	
Cell culture flask	Fisher Scientific	156340	Culture flask T25
Cell strainer	Corning Incorporated	352340	Cell strainer 40 µm
Confocal microscope	Leica		Confocal TCS SP8 microscopy coupled with an Olympus FluoView 300 confocal system
Conical tubes	Thermo Scientific	339651, 339652	Sterile tubes of 15 mL and 50 mL
DAPI	Abcam	ab224589	DAPI staining solution
DMEM/F12	Gibco; Life Technologies Corporation	12500062	DMEM/F-12 50/50, 1X (Dulbecco's Mod. Of Eagle's Medium/Ham's F12 50/50 Mix) with L-glutamine
Dyalysis tubing	Sigma-Aldrich	D9527	Molecular weight cut-off = 14 kDa
Ethanol	Fisher Scientific	64-15-5	Reagent grade
Fetal Bovine Serum	Gibco; Life Technologies Corporation	12657011	Research Grade
Fibrinogen	Sigma-Aldrich	9001-32-5	Fibrinogen crystalline powder from bovine plasma
Gelatin	Sigma-Aldrich	9000-70-8	Gelatin powder from porcine skin
Glycine	Sigma-Aldrich	56-40-6	Glycine powder
Hanks Buffered Salt Solution (HBSS)	Gibco; Life Technologies Corporation	14175095	No calcium, no magnesium, no phenol red
L-Glutamine	Sigma-Aldrich	56-85-9	L-Glutamine crystalline powder
Laminin	Sigma-Aldrich	114956-81-9	Laminin 1-2 mg/mL L in 50 mM Tris-HCl
Live dead kit cell imaging kit	Thermo Scientific	R37601	Green fluorescence in live cells (ex/em 488 nm/515 nm). Red

			fluorescence in dead cells (ex/em 570 nm/602 nm)
Methacrylic anhydride	Sigma-Aldrich	760-93-0	For GelMA preparation
Microtubes	Corning Incorporated	MCT-150-C	Microtubes of 1,5 mL
NaCl	Sigma-Aldrich	7647-14-5	
Needle 22G	Fisher Scientific	NC1362045	Sterile blunt needle
Operating scissor	Integra Miltex	05--02	Sharp scissor for brain dissection previously sterilized
Paraformaldehyde	Sigma-Aldrich	30525-89-4	Paraformaldehyde powder
Penicillin/Streptomycin	Gibco; Life Technologies Corporation	15070063	Pen Strep (5,000 Units/ mL Penicillin; 5,000 ug/mL Streptomycin)
Petri dish	Corning Incorporated	430591, 430588	Sterile petri dishes of 35 and 100 mm
Phalloidin	Abcam	ab176753	iFluor 488 reagent
Photoinitiator	Sigma-Aldrich	106797-53-9	2-Hydroxy-4'-(2-hydroxyethoxy)-2-methylpropiophenone
Phosphate buffer saline (PBS)	Gibco; Life Technologies Corporation	10010023	PBS 1 x, culture grade, no calcium, no magnesium
Poly-L-lysine	Sigma-Aldrich	25988-63-0	Poly-L-lysine hydrobromide mol wt 30,000-70,000
Primary antibody	Abcam	ab4674	Chicken polyclonal to GFAP
Secondary antibody	Abcam	ab150176	Alexa fluor 594 anti-chicken
Spatula	Miltex	V973-70	Number 24 cement spatula previously sterilized
Stereomicroscope	Fisherbrand	3000038	Microscope for brain dissection
Syringe 5 mL	BD	1222C84	Sterile syringe
Syringe filter 2 µm	Fisher Scientific	09-740-105	Polypropylene filter for sterilization
Thrombin	Sigma-Aldrich	9002--04-4	Thrombin crystalline powder from bovine plasma
Triton X-100	Sigma-Aldrich	9002-93-1	Laboratory grade
Trypsin-EDTA	Gibco; Life Technologies Corporation	15400054	Trypsin no phenol red 1 x diluted in PBS
Versene solution	Gibco; Life Technologies Corporation	15040066	Versene Solution (0.48 mM) formulated as 0.2 g EDTA(Na4) per liter of PBS
Well plate	Thermo Scientific	144530	Sterile 24-well plate