

Materials List for:

# A High Output Method to Isolate Cerebral Pericytes from Mouse

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## Materials

Name	Company	Catalog Number	Comments
Amino acids BME	Sigma	B-6766	Store at 4 °C.
Basal DMEM media	Invitrogen	316000083	Store at 4 °C.
Basic fibroblast growth factor	Sigma	F-0291	Store at -20 °C.
BSA	Sigma	A-8412	Store at 4 °C.
Collagenase dispase	Sigma	10269638001	Prepare a 10x stock solution in sterile PBS-CMF. Filter the solution with a 0.22 µm syringe filter and store at -20 °C. Note: For the enzyme digestion step of the protocol, for every set of 10 mice for extraction, 300 µL of 10x collagenase dispase is required.
Dextran	Sigma	31398	
DNase I	Sigma	11284932001	Prepare a 1000X stock solution by dissolving 100 mg in 10 ml sterile water and store at -20°C.
Gelatin	Sigma	G-2500	Prepare the working coating by making a 0.2% gelatin solution in sterile PBS-CMF (8 g/L NaCl, 0.2 g/L KCl, 0.2 g/L KH <sub>2</sub> PO <sub>4</sub> , 2.86 g/L NaHPO <sub>4</sub> (12 H <sub>2</sub> O), pH 7.4). Autoclave the solution for minimum 20 minutes at 120 °C and store at room temperature. Culture dishes to be coated for at least 4 hours at 4 °C.
Gentamycin	Biochrom AG	A-2712	Store at 4 °C.
Glutamine	Merck	I.00289	Store at -20 °C.
HBSS	Sigma	H-8264	Store at 4 °C.
HEPES	Sigma	H-0887	Store at 4 °C.
Matrigel	BD Biocoat	354230	Prepare a working coating solution of Matrigel by diluting stock in cold DMEM at 1:48 ratio with its final concentration to be 85 µg/cm <sup>2</sup> . Cell culture dishes should be coated at least for 1 hour at room temperature.
Pericyte Medium-mouse	Sciencell research laboratories	1231	Store at 4 °C.
Tosyl Lysin Chloromethyl Ketone	Sigma	T-7254	Prepare a 1000X stock solution in WBA by dissolving 16 mg in 10.88 mL of WBA to make a 4 mM solution and store at 4 °C.
Vitamins	Sigma	B-6891	Store at -20 °C.

<b>Equipment Requirements</b>			
Filtration tools	Sefar, Nylon mesh, 60-micron porosity		
Laboratory equipment	Swing bucket rotor centrifuge		
Water bath with agitator			
Laminar Flow Hood : BSL2			
Glassware (all components to be heat sterilized)	Dounce Tissue Grinder With Glass Pestle		
Pestle I: 0.0035 - 0.0065 inches			
Pestle II: 0.0010 - 0.0030 inches			
Vacuum filter assembly with coarse porosity fritted glass filter support base			
Surgical dissection tools (all components to be heat sterilized)	Forceps, scissors, Bunsen burner, cotton swabs, gauge		