

Establishing Mixed Neuronal and Glial Cell Cultures from Embryonic Mouse Brains to Study Infection and Innate Immunity

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Materials

Name	Company	Catalog Number	Comments
10x Trypsin	Sigma	T4549-100ML	To digest tissue
140 mm TC Dish	Fisher	11339283	Put 8 35 mm dishes per 1 140 mm dish
15 mL Falcon	Sarstedt	62554502	To collect cells into pellet for resuspension in plating media
18 G needle	Henke Sass Wolf	4710012040	For trituration of sample
21 G needle	BD	304432	For trituration of sample
23 G needle	Henke Sass Wolf	4710006030	For trituration of sample
35 mm TC Dish	Corning	430165	Plate out 3 PLL coated coverslips per 1 35 mm dish
5 mL syringe	Fisher	15869152	For trituration of sample
6 well plate	Corning	3516	To plate out cells for RT-qPCR, and flow cytometry
7 mL Bijoux	Fisher	DIS080010R	To put brains in tp
96 well plate	Corning	3596	To plate out cells for high-throughput testing
ACSA-2 Antibody, anti-mouse, PE	Miltenyi	130-123-284	For flow cytometry staining of astrocytes
Angled forceps	Dumont	0108-5/45-PO	For dissection
Biotin	Sigma	B4501	For DM+/-
Boric Acid	Sigma	B6768-500G	For boric acid buffer
Brilliant Violet 421 anti-mouse CD24 Antibody, clone M1-69	Biolegend	101825	For flow cytometry staining of neurons and astrocytes
Brilliant Violet 605 anti-mouse CD45 Antibody, clone 30-F11	Biolegend	103139	For flow cytometry staining of microglia
Brilliant Violet 785 anti-mouse/human CD11b Antibody, clone M1/70	Biolegend	101243	For flow cytometry staining of microglia

BSA Fraction V	Sigma	A3059-10G	For SD Inhibitor
CNP	Abcam	AB6319	Mature oligodendrocytes
Coverslip	VWR	631-0149	To plate out cells for microscopy
Dissection Scissors	Sigma	S3146-1EA	For dissection
DMEM High glucose, sodium pyruvate, L-Glutamine	Gibco	21969-035	For DM+/-, and for plating media
DNase I	Thermofisher	18047019	For SD Inhibitor, can use this or the other Dnase from sigma
DNase I	Sigma	D4263	For SD Inhibitor, can use this or the other Dnase from thermofisher
eBioscience Fixable Viability Dye eFluor 780	Thermofisher	65-0865-14	Live / Dead stain
Fine forceps	Dumont	0102-SS135-PO	For dissection
GFAP	Invitrogen	13-0300	Astrocytes
HBSS w Ca Mg	Sigma	H9269-500ML	For plating media
HBSS w/o Ca Mg	Sigma	H9394-500ML	For brains to be added to
Horse Serum	Gibco	26050-070	For plating media
Hydrocortisone	Sigma	H0396	For DM+/-
Iba1	Alpha-Laboratories	019-1971	Microglia
Insulin	Sigma	I1882	For DM+
Leibovitz L-15	Gibco	11415-049	For SD Inhibitor
MBP	Bio-Rad	MCA409S	Myelin
Mouse CCL5/RANTES DuoSet ELISA Kit	BioTechne	DY478-05	ELISA kit for quantifying concentration of CCL5 in supernatants of 96 well plate
N1 media supplement	Sigma	N6530-5ML	For DM+/-
Nestin	Merck	MAB353	Neuronal stem/progenitor cells
NeuN	Thermofisher	PA578499	Neuronal cell body
NG2	Sigma	AB5320	Immature oligodendrocytes
O4 Antibody, anti-human/mouse/rat, APC	Miltenyi	130-119-155	For flow cytometry staining of oligodendrocytes
Pen/Strep	Sigma	P0781-100ML	For DM+/-, and for plating media
Poly-L-Lysinehydrobromide	Sigma	P1274	For Boric acid / poly-L-lysine solution to coat coverslips
SMI31	BioLegend	801601	Axons
Sodium Tetraborate	Sigma	221732-100G	For boric acid buffer
Trizol	Thermofisher	15596026	For lysing cells for RT-qPCR
Trypsin inhibitor from soybean	Sigma	T9003-100MG	For SD Inhibitor