

Materials List for:

A Cell Culture Model of Resistance Arteries

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Materials

Name	Company	Catalog Number	Comments
24 mm diameter, 0.4 µm Transwell Polyester membrane filter insert	Corning	3450	This is one 6 well plate of inserts
225 cm flask	Corning	431082	
6 well plates (without filter inserts)	Corning	3506	
Primary human coronary artery endothelial cells	Lonza	CC-2585	
Primary human coronary artery smooth muscle cells	Lonza	CC-2583	
EBM-2 EC Basal media	Lonza	CC-3156	
EC growth factors (EGM2 MV Single quotes)	Lonza	CC-4147	Add to basal media before use
SmBM SMC basal media	Lonza	CC-3181	
SMC growth factors (SmGM-2 SingleQuote)	Lonza	CC-4149	Add to basal media before use
0.5% Trypsin-EDTA 10X	Gibco	15400-054	Dilute to 2X with sterile dPBS
Hemocytometer	VWR	15170-208	
large petri dishes for SMC Plating (150mm)	Corning	353025	
Bovine gelatin	Sigma	G-9382	
Human fibronectin	Corning	356008	5µg/ml of fibronectin and store at -20
10x Hanks' Buffered salt solution (HBSS)	Gibco	14065-056	Dilute to 1x with sterile water
10x Dulbecco's Phosphate buffered saline (dPBS)	Gibco	14200-075	Dilute to 1x with sterile water
Disposable Curved Scalpel (30mm cutting edge) with handle	Amazon	MDS15210	
Forceps	Fisher	17-467-201	
Cell lifter	Corning	3008	
Cell scraper	BD Falcon	353085	
50 mL conical tube	Corning	352070	
10 mm petri dishes	Falcon	35-1008	
Rneasy mini kit	Qiagen	74104	
RNA lysis reagent	Qiagen	79306	
Paraformaldehyde	Sigma	158127	Make 4% solution with dPBS
70% ethanol	Fisher	04-355-305	

Cavicide disinfectant	Fisher	131000	
RIPA protein lysis buffer	Sigma	R0278	
Sonic dismembrator	Fisher	Model 50	
1x PBS for harvesting EC/SMC and immunocytochemistry	Gibco	10010023	does not need to be sterile
Name	Company	Catalog Number	Comments
Embedding/Sectioning/Staining			
Paraffin	Fisher	P31-500	
Automated tissue processor	Excelsior	ES	
Embedding station + cold plate	Leica	HistoCore Arcadia	
Tissue Tek Accu-Edge Microtome blade	VWR	25608-961 or 25608-964	
Microscope slides	Thermo Fisher	22-230-900	
Coverslips	Thermo Fisher	12-548-5E	
Prolong Gold mounting medium	Thermo Fisher	P36931	
Name	Company	Catalog Number	Comments
Other Solutions			**Perform all steps in sterile conditions if using the solution on live cells
Sterile dPBS			Autoclave water to sterilize. Using the sterile water make DPBS (450mL sterile water: 50mL 10X DPBS), mix and filter sterilize (0.2µm filter). Final concentration: 1X
sterile HBSS			Autoclave water to sterilize. Using the sterile water make HBSS (450mL sterile water: 50mL 10X HBSS), mix and filter sterilize (0.2µm filter). Final concentration: 1X
Fibronectin stock solution			Use 50mL of sterile water to fibronectin to make stock of 1mL aliquots, then freeze until needed Final concentration: 100µg/mL
Fibronectin to coat SMC side of VCCC			thaw one aliquot and add 1mL of fibronectin (100µg/mL) to 19mLs of 1X HBSS to reach a final concentration of 5µg/mL. Final concentration: 5µg/mL
2x Trypsin EDTA solution			Dilute the 10X 0.5% Trypsin-EDTA to 2X with 1X dPBS (40mL DPBS: 10mL 10X Trypsin) . Final concentration: 2X
Blocking/antibody solution			(9mL 1X PBS, 25mL Triton X100, 50mg bovine serum albumin, 500mL normal serum)
Bovine gelatin to coat EC VCCC			Mix 0.5g bovine gelatin into 100mL distilled water. Autoclave prior to first use and store at room temperature. Keep in sterile conditions. Final concentration: 0.50%
1x PBS for harvesting cells			does not need to be sterile