

Materials List for

Directed Differentiation of Hemogenic Endothelial Cells from Human Pluripotent Stem Cells

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Citation

Nelson, E.A., Qiu, J., Chavkin, N.W., Hirschi, K.K. Directed Differentiation of Hemogenic Endothelial Cells from Human Pluripotent Stem Cells. *J. Vis. Exp.* (169), e62391, doi:10.3791/62391 (2021).

Date Published

March 31, 2021

DOI

10.3791/62391

URL

jove.com/video/62391

Materials

Name	Company	Catalog Number	Comments
15 cm dishes	Corning	430599	tissue culture treated
35 mm dishes	Corning	430165	tissue culture treated
6-well plates	Corning	3516	tissue culture treated
Antimicrobial reagent Brand Name: Normocin	Invitrogen	ant-nr-1	
bFGF	R&D systems	233-FB-025	use at 50 ng/mL
BMP4	BioLegend	595202	use at 25 ng/mL
Bovine Serum Albumin (BSA)	Fisher Scientific	BP1600-1	
Cell Detachment Solution Brand Name: Accutase	Stemcell Technologies	7920	
Dimethyl Sulfoxide (DMSO)	Sigma Aldrich	D2650-100mL	
Dispase	Stemcell Technologies	7913	
DLL4	R&D systems	1506-D4/CF	recombinant human; use at 10 µg/mL
DMEM:F12	Gibco	11320-033	
Dulbecco's Phosphate Buffered Saline (PBS)	Gibco	14190144	
Endothelial cell growth medium Brand Name: EGM-2 Endothelial Cell Growth Medium-2 BulletKit (EGM-2)	Lonza	CC-3162	
FACS tubes	Corning	352235	polystyrene round bottom with filter cap
Fetal Bovine Serum (FBS)	Gemini Bio	100-106	
Fibronectin	ThermoFisher Scientific	33016015	use at 4 µg/cm ²
GSK3i/CHIR99021	Stemgent	04-0004-02	10 mM stock; use at 5 µM
Hanks Balanced Salt Solution (HBSS)	Gibco	14175-095	
Hydrochloric Acid (HCl)	Fisher Scientific	A144S-500	
Matrix protein Brand Name: Matrigel	Corning	356230	Growth factor reduced. Refer to the Certificate of Analysis for the lot

			to determine the protein (Matrigel) concentration. This concentration is required to calculate the volume of Matrigel that contains 1 mg of protein.
Methylcellulose-based medium Brand Name: MethoCult H4435 Enriched	Stemcell Technologies	4435	
Pluripotent stem cell differentiation medium Brand Name: STEMdiff APEL 2	Stemcell Technologies	5270	
Pluripotent stem cells: H1, H9, H9-FUCCI	WiCell	WA09 (H9), WA01 (H1)	human; H9-FUCCI were obtained from Dr. Ludovic Vallier's lab at Cambridge Stem Cell Institute
Protein-Free Hybridoma Medium (PFMH)	Gibco	12040077	
Retinoic Acid	Sigma Aldrich	R2625-50mg	use at 0.5 μ M
Reverse transcription master mix Brand Name: iScript Reverse Transcription Supermix	BioRad	1708840	
RNA extraction kit Brand Name: RNeasy Mini Kit	Qiagen	74104	
Sodium Hydroxide (NaOH)	Fisher Scientific	SS255-1	
Stem cell growth medium Brand Name: mTeSR1	Stemcell Technologies	85850	
SYBR Green master mix Brand Name: iTaq Universal SYBR Green Master Mix	BioRad	1725121	
Trypsin-EDTA	Gibco	25299956	0.25%
VEGF ₁₆₅ (VEGF-A)	PeproTech	100-20	use at 50 ng/mL
α -CD31-FITC	BioLegend	303104	2 μ g/mL*
α -CD34-Pacific Blue	BioLegend	343512	2 μ g/mL*
α -CD45-APC/Cy7	BioLegend	304014	2 μ g/mL*
α -c-Kit-APC	BioLegend	313206	2 μ g/mL*
α -Flk-1-PE/Cy7	BioLegend	359911	2 μ g/mL*
α -VE-Cadherin-PE	BioLegend	348506	2 μ g/mL*
* Antibody fluorescent conjugates should be optimized based on the cell sorter used. Presented here are the final concentrations utilized in this study.			