

Materials List for:

Generation of Human Cardiomyocytes: A Differentiation Protocol from Feederfree Human Induced Pluripotent Stem Cells

Elisa Di Pasquale^{1,2}, Belle Song¹, Gianluigi Condorelli¹

Correspondence to: Elisa Di Pasquale at elisadipa@gmail.com

URL: https://www.jove.com/video/50429

DOI: doi:10.3791/50429

Materials

Name	Company	Catalog Number	Comments
Media and Reagents	'	•	
Human ESC-qualified Matrix	BD Biosciences	354277	Thaw the 5 ml bottle at 4 °C on ice O/N, aliquot in pre-chilled cryotubes on ice, according to the data sheet dilution instructions and store at -20 °C.
Fibronectin, from human plasma, 0.1% solution	Sigma	F0896-2MG	Working concentration: 5 μg/cm ²
Laminin	Sigma	L2020-1MG	Working concentration: 5 μg/cm ²
PBS (no Mg and Ca), 10X	Lonza	BE17-515F	
PBS (with Mg and Ca), 10X	Bio Sera	XC-S2067/500	
Dispase II, neutral protease, grade II	Roche	4942078001	Stock: 10 mg/ml in DMEM-F12 at -20 °C, working solution: 1 mg/ml in PBS (no Mg/Ca)
Trypsin/EDTA, 0.5%	Sigma	T3924	
Collagenase type 2	Worthington	4176	Dilute to 480 U/ml (e.g. 1.6 mg/ml) in PBS with Mg and Ca
DMEM-F12	Life Technologies	21331-020	
Knockout DMEM	Life Technologies	10829-018	
Knockout Serum Replacement (KSR)	Life Technologies	10828-028	Thaw at 4 °C, aliquot and store at -20 °C
F–tal Bovine Serum (origin South America)	Invitrogen	10270-106	Batches should be tested for their cardiogenic potential. Thaw at 4 °C, heat-inactivate at 56 °C for 30 min, aliquot and store at -20 °C
PenStrep, penicillin-Streptomycin, 100X	Life Technologies	15140-122	
GlutaMAX, 100X	Life Technologies	35050-038	
MEM NEAA, 100X	Invitrogen	11140-135	
N2 supplement, 100X	Life Technologies	17502-048	
B27 supplement, 50X	Life Technologies	12587-010	
2-mercapt-thanol	Invitrogen	31350-010	
Gelatin, from porcine skin	Sigma	G1890-100G	Make stock at 1% in water, store at -20 °C. Dilute to 0.1% in PBS and keep at 4 °C
mTeSR1	Stem Cell Technologies	5850	Combine Supplement 5X with the basic medium, aliquot and store at -20 °C. Do not keep complete

¹Humanitas Clinical and Research Center, Italy

²Institute of Genetic and Biomedical Research (IRGB), National Research Council (CNR)



			medium at 4 °C for more than 1 week
Nutristem hESC XF	Stemgent from Biological Industries	05-100-1A	Thaw at 4 °C O/N, aliquot and store at -20 °C until use. Aliquots may be kept at 4 °C for no more than a week.
mFreSR	Stem Cell Technologies	5853	
Ascorbic acid	Sigma	A4544-25G	Prepare stocks at 10 mg/ml in water. Store aliquots at -20 °C in the dark. Add to EBM at the final concentration (500X)
Plasticware	•		•
35 mm culture dishes	Becton Dickinson	353001	
Cell scraper	Corning Incorporated	3008	
12-well plates	Becton Dickinson	353043	
Permanox 2-well chamber slides	Thermo Fisher Scientific	177437	
Glass 2-well chamber slides	Thermo Fisher Scientific	177380	
6-well plates, ultra-low-attachment surface	Corning Incorporated	3471	
18G needle	Becton Dickinson	304622	
Glass pasteur pipettes, 230 mm	VWR International	612-1702	
2.5 ml syringe	Becton Dickinson	300188	
Equipments	•		•
IVF Workstation	KSystem, by Nikon		
Nikon SMZ1500 Stereomicroscope	Nikon		