

Materials List for:

Isolation and Genetic Manipulation of Adult Cardiac Myocytes for Confocal Imaging

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Materials

Name	Company	Catalog Number	Comments
Anesthesia solution	Serumwerk Bernburg GmbH (Ursotamin), Bayer Health Care (Rompun)		Mixture of 1 ml Ursotamin (100 mg/ml Ketaminhydrochlorid) and 240 µl Rompun (2% Xylazinhydrochlorid)
Citrate solution			117,64 mg sodium citrate in 10 ml 0.9% NaCl solution
Solution A			134 mM NaCl, 4 mM KCl, 11 mM glucose, 1.2 mM MgSO ₄ , 1.2 mM Na ₂ HPO ₄ , 10 mM HEPES, pH adjusted to 7.35 using 10 M NaOH, sterile filtered
Solution A+			Content of solution A plus 0.2 mM EGTA, pH adjusted to 7.35 using 10 M NaOH, sterile filtered
Liberase solution	F. Hoffmann# La Roche Ltd.	11988476 001	500 µl stock solution in 15 ml solution A (stock solution:10 mg/ml Liberase Blendzyme 4 in aqua dest.)
Solution B			Content of solution A plus 200 µM CaCl ₂ and 0.1% DNase solution
Solution B _{1/2}			1:1 mixture of solution A and solution B
DNase solution	Sigma-Aldrich	D4527	Deoxyribonuclease I, Type 2 from bovine pancreas, 40 KU (15 mg) are dissolved in 5 ml of 10 mM Tris#HCl buffer, adjusted to pH=7.35, additional content: 50 mM NaCl, 10 mM MgCl ₂ , 1 mM dithi-rythritol; this solution is mixed with 5 ml glycerol
Extracellular matrix protein (ECM) solution	Extracellular matrix protein (ECM) solution	E1270	1 ml stock solution as purchased is diluted with 6 mLmedium M199
Culture medium M199	PAA Laboratories	E15#834	Medium M199 supplemented with 1 µl/ml ITS solution and 20 µl/ml penicillin#streptomycin solution (5000 units/ml)
ITS solution			25 mg Insulin, 25 mg Transferrin and 50 µl Selenite stock solution are dissolved in 5 ml aqua dest. (stock solution: 0.5 mg/ml Sodiumselenite in aqua dest.). Add a few drops of 1 M HCl until one should get a clearsolution. Aliquots can be frozen.

Tyrode			135 mM NaCl, 5.4 mM KCl, 1.8 mM CaCl ₂ , 2mM MgCl ₂ , 10 mM glucose, 10 mM HEPES, pH adjusted to 7.35 using 10 M NaOH, sterile filtered
Master mix			5 µl 10×PCR#Buffer, 5 µl MgCl ₂ (25 mM), 2 µl forward primer, 2 µl reverse primer, 1 µl dNTP's (10 mM each), 33 µl H ₂ O, 1 µl DMSO, 1 µl Taq#DNA polymerase for each sample
LB#Medium			10 g/l Tryptone, 5 g/l Yeast extract, 5 g/l NaCl (and 15 g/l Agar if used as solid medium), pH 7.0
Pac I	New England Biolabs	R0547L	preparation: 50 µg DNA, 5 µl 10×buffer, 0.5 µl 100×BSA, 0.75 µl Pac I, fill with H ₂ O up to final volume of 50µl