Materials List for:

Adult and Embryonic Skeletal Muscle Microexplant Culture and Isolation of Skeletal Muscle Stem Cells

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URL: http://www.jove.com/video/2051/

Materials

Name	Company	Catalog Number	Comments
DMEM/F12 1:1 mix	Sigma-Aldrich		Liquid medium: (Dulbecco's Modified Eagles's medium and Ham's F12 medium, 1:1 v/v)
100Ã Glutamine (200mM)	Sigma-Aldrich		Diluted in medium to a 1Ã concentration of 2 mM (Glutamine HYBRI-MAX R)
Fetal calf serum (FCS)	From a number of different companies		Batch tested on primary cultures and skeletal muscle cell lines. 10-20% supplement to liquid media
Heraeus Labofuge 300	Heraeus Instruments		Lab centrifuge capable of reaching 1,000 rpm
15 ml Falcon centrifuge tubes	Fisher Scientific		Must fit lab centrifuge
Tissue culture plasticware (25, 75 or 175 mm2 tissue culture vessels; 96-well tissue culture plates. 60 mm Petri dishes).	Nalge Nunc international		
Humidified CO_2 incubator (Heraeus)	Heraeus Instruments		Ours is copper lined, recommended for reducing contamination
Sterile hood with laminar air flow (Heraeus)	Heraeus Instruments		Ours is a Class II hood suitable for use with Human tissues
Water Bath	Grant Equipment		Maintained always at 37°C
Inverted microscope	Leica Microsystems		
70% Ethanol			For sterilization (animals, dissection instruments) and swabbing benches, hood, etc.
Calcium- and magnesium-free phosphate-buffered saline (PBS)	Sigma-Aldrich		Cell culture-tested PBS (Dulbecco's formula) is purchased as a ready-mixed powder or in tablet form and made up with doubledistilled water before sterilization by autoclave. PBS consists of 2.68 mM potassium chloride (KCl); 1.47 mM potassium phosphate monobasic (KH2PO4); 0.137 M sodium chloride (NaCl); and 8.1 mM sodium phosphatedibasic (Na_2HPO_4). PBS can be prepared from scratch as follows: 200 mg KCl, 200 mg KH_2PO_4, 8 g NaCl and 1.15 g Na_2HPO_4/l of double-distilled water followed bysterilization by autoclave.
CryoTube vials	Nalge Nunc international		
A Neubauer haematocytometer and coverslips	Fisher Scientific		For estimating cell counting

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Hand counter	Fisher Scientific		
Dissection microscope, Zeiss Stemi 1000	Carl Zeiss, Inc.		For preparation of explants
Small sterile hood			
Sterile dissection instruments (including Jeweler's forceps)			Sterilised by autoclave
Sterile plastic collecting vessels (7 ml bijou tubes or 20ml universals)	Nalge Nunc international		
Warm PECM	Sigma-Aldrich		Made up in the sterile hood and warmed to 37°C in the tissue culture waterbath. DMEM:F-12 supplemented with 20% FCS, 1% glutamine and + 1% penicillin & streptomycin solution
Dispase (50 g/ml, equivalent to 6 units/mg)	Available from MP Biomedicals, UK		
10 g/ml 4_,6 Diamidino-2-phenylindole, dihydrochloride (DAPI).	Sigma-Aldrich		For microscopic visualization of apoptosis and mitosis
Vectashield fluorescent mounting fluid	Vector Laboratories		
Fluorescent upright microscope with ultraviolet filter (we use a Nikon Eclipse E600)	Nikon Instruments		
Digital camera and imaging software (we use a Nikon Coolpix 995 camera; A Nikon D3 camera	Nikon Instruments		
OpenLab4.0a software	PerkinElmer, Inc.		
Photoshop CS4	Adobe		

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