

Materials List for:

# Quantification of Monocyte Transmigration and Foam Cell Formation from Individuals with Chronic Inflammatory Conditions

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## Materials

Name	Company	Catalog Number	Comments
<b>Gel preparation reagents</b>			
NaOH	Sigma-Aldrich	221465-500G	0.1 M NaOH diluted in H <sub>2</sub> O
10x M199	Sigma-Aldrich	M0650	
AcCOOH	Sigma-Aldrich	695092-100ML	20 mM Acetic acid diluted in H <sub>2</sub> O
Cultrex Bovine Collagen I	R&D Systems	3442-050-01	Type I Fibrous Collagen
Name	Company	Catalog Number	Comments
<b>Cell culture</b>			
M199	Life Technologies	11150-059	M199 media containing Earle's salts, L-glutamine and 2.2 g/L Sodium Bicarbonate. Media supplemented with 100 µg/mL L-glutamine and 100 U/mL penicillin/streptomycin
M20			Supplemented M199 containing 20% heat-inactivated pooled or donor serum. Individual lipid species such as LDL can be added to M199.
HUVEC			Primary human umbilical cord endothelial cells (HUVEC) can be isolated from umbilical cords donated with informed consent and ethics approval. Isolated HUVEC may also be purchased commercially.
Human coronary artery endothelial cells			Primary human coronary artery endothelial cells can be isolated from arteries donated with informed consent and ethics approval. Isolated cells may also be purchased commercially.
EDTA	BDH Merck	10093.5V	Ethylenediaminetetraacetic acid (EDTA) - 0.5 M, pH 8.0
EGTA	Sigma-Aldrich	E3889-500G	Ethylene glycol-bis(β-aminoethyl ether)-N,N,N',N'-tetraacetic acid (EGTA) - 1 mM, pH 8.0
0.05% trypsin EDTA	Gibco	25300-054	0.05% trypsin/0.53 EDTA (1X)
L-glutamine	Gibco	25030-081	L-glutamine (200 mM)

Penicillin/streptomycin	Gibco	15140-122	Penicillin/streptomycin (10,000 units/mL Penicillin and 10,000 µg/mL Streptomycin)
New born calf serum	Gibco	16010-142	New born calf serum: New Zealand origin
Fibronectin	Sigma-Aldrich	F1056-1MG	Fibronectin - 50 µg/mL aliquots prepared and stored
PBS (1x)	Gibco	14200-075	Dulbecco's Phosphate Buffered Saline (10x): Dilute to 1x with sterile H <sub>2</sub> O
0.1% TNF	Gibco	PHC3015	Recombinant human TNF - Reconstituted in H <sub>2</sub> O and stored in 10 µg/mL aliquots
Low-density lipoprotein	Merck Millipore	LP2-2MG	Low-density lipoprotein (LDL)
<b>Name</b>	<b>Company</b>	<b>Catalog Number</b>	<b>Comments</b>
<b>Microscopy</b>			
1 or 2% formaldehyde	Polysciences	4018	1 or 2% formaldehyde diluted with sterile H <sub>2</sub> O
50% and 78% methanol	Ajax Finechem	318-2.5L GL	50% or 78% v/v methanol, diluted with H <sub>2</sub> O
Oil Red O stain	Sigma-Aldrich	O0625-25G	Dilute to 2 mg/mL in 22% 1M NaOH and 78% (v/v) methanol
Microscope slides	Mikro-Glass	S41104AMK	Twin frosted 45 degree ground edge microscope slides (25 X 76 mm)
Cover slips	Menzel-Gläser	MENCS224015GP	22 x 40 mm #1.5 size glass cover slips
Double-sided tape	3M Scotch	4011	Super strength exterior mounting tape (25.4 mm x 1.51 m)
Giemsa stain	Merck Millipore	1.09204.0500	Giemsa's azur eosin methylene blue solution (dilute stock 1:10 in H <sub>2</sub> O)
Hole punch			Hand-held single hole punch (6.35 mm punch)
<b>Name</b>	<b>Company</b>	<b>Catalog Number</b>	<b>Comments</b>
<b>Flow cytometry</b>			
Collagenase D	Roche Diagnostics	11088858001	Collagenase D diluted in M199 media to 1 mg/mL
35 µm nylon mesh capped polystyrene FACS tubes	BD Biosciences	352235	35 µm nylon mesh capped polystyrene FACS tubes
Live/Dead Fixable Yellow Dead Cell stain	Life Technologies	L34959	Live/Dead Fixable Yellow Dead Cell stain
FACS wash			Prepare by mixing 1 X PBS <sup>-</sup> , 2 mM EDTA and 1% New born calf serum
<b>Name</b>	<b>Company</b>	<b>Catalog Number</b>	<b>Comments</b>
<b>Plasticware</b>			
96 well plate	Nunc	167008	Delta surface flat-bottomed 96 well plate
10 cm Petri Dish	TPP	93100	Sterile 10 cm Petri dish
1.5 mL Eppendorf tubes	Eppendorf	0030 125.150	Eppendorf tubes
Transfer pipette	Samco Scientific	222-20S	Sterile transfer pipette (1 mL, large bulb)