

Materials List for:

Biological Compatibility Profile on Biomaterials for Bone Regeneration

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URL: <https://www.jove.com/video/58077>

DOI: [doi:10.3791/58077](https://doi.org/10.3791/58077)

Materials

Name	Company	Catalog Number	Comments
Biomaterials			
β-tricalcium phosphate disks (β-TCP, 14 mm)			Disks were sintered in a muffle furnace at a temperature of 1150 °C. A detailed description of the production can be found in Zimmer et al (doi: 10.1002/jbm.a.34639). Samples were UV-irradiated (15 min for each side) before using in cell cultures.
Orthovita Vitoss foam (β-TCP/C foam)	Orthovita	2102-1405	
Mouse osteoblast culture			
Alizarin Red S	Sigma-Aldrich	A5533	
ALP assay buffer (2x) pH 10.4			Dissolve 200 mM glycine, 2 mM magnesium chloride, 2 mM zinc chloride in ultrapure water and adjust to pH 10.
Bone growth medium (BGM)			α-MEM + 10% heat-inactivated FBS + 1% penicillin/streptomycin
Cell lysis buffer: CyQuant cell lysis buffer 20x concentrate	Thermo Scientific	C7027	Dilute the concentrated cell lysis buffer stock 20-fold in ultrapure water.
Cell viability reagent: Presto Blue cell viability reagent	Invitrogen, Thermo Fisher Scientific	A13261	
Collagenase type IV from clostridium histolyticum	Sigma-Aldrich	C5138	
DC protein assay kit II	Bio Rad	5000112	DC protein assay kit contains reagent A, reagent B, reagent S and BSA for the reference standard. For reagent A', add 20 μL reagent S to 1000 μL reagent A.
Dimethylsulfoxide (DMSO)	Sigma-Aldrich	D8418	Steril-filter DMSO before usage.
Dispase II (neutral protease, grade II)	Roche	4942078001	
EnzCheck Phosphatase Assay Kit for alkaline phosphatase activity	Invitrogen, Thermo Fisher Scientific	E12020	EnzCheck Phosphatase Assay Kit contains 6,8-difluoro-4-methylumbelliferyl phosphate (DiFMUP), N,N-dimethylformamide and 6,8-difluoro-7-hydroxy-4-methylcoumarin (DiFMU).
Fetal bovine serum (FBS)	Sigma-Aldrich	F7524	
Formalin solution neutral buffered 10%	Sigma-Aldrich	HT501128	

Glycine	Sigma-Aldrich	G8898	
Hexadecylpyridinium (cetylpyridinium) chloride monohydrate	Sigma-Aldrich	C9002	
L-Ascorbic acid 2-phosphate sesquimagnesium salt hydrate	Sigma-Aldrich	A8960	
Magnesium chloride hexahydrate	Sigma-Aldrich	M2670	
MEM alpha medium (α -MEM)	Gibco Life Technologies	11900-073	
Osteogenic mineralization medium (MM)			α -MEM + 10% heat-inactivated FBS + 1% penicillin-streptomycin + 50 μ g/mL ascorbic acid + 5 mM β -glycerophosphate
Penicillin-streptomycin	Sigma-Aldrich	P4333	
Phosphate-buffered saline 1x (PBS) pH 7.4	Gibco, Thermo Fisher Scientific	10010023	
Sigmafast BCIP/NBT	Sigma-Aldrich	B5655	
Trypsin-EDTA (0.5%), no phenol red (10x)	Gibco, Thermo Fisher Scientific	15400054	Dilute the concentrated stock solution 10-fold in 1x PBS.
Tween20	Sigma-Aldrich	P1379	
Ultrapure water: Cell culture water pyrogen free	VWR	L0970-500	
Wash buffer			Add 0.05% Tween20 to 1x PBS.
Zinc chloride	Sigma-Aldrich	1.08816	
β -Glycerophosphate disodium salt hydrate	Sigma-Aldrich	G5422	
24-well suspension culture plate	Greiner bio-one	662102	
24-well tissue culture plate	Greiner bio-one	662160	
50 mL conical tube	Greiner bio-one	227261	
96-well black plate	Greiner bio-one	655076	
96-well transparent plate	Greiner bio-one	655001	
Centrifuge: VWR Mega Star 600R	VWR		
CO2-Incubator: HeraCell 240	Thermo Scientific		
Cryovial 2 mL	Greiner bio-one	122263	
Disposable filter unit FP30/0.2 CA-S	GE Healthcare Life Sciences Whatman	10462200	For sterile filtration of enzyme solution.
Flatbed scanner: Epson Perfection 1200 Photo	Epson		
Infinite M200 Pro microplate reader	Tecan		
Microcentrifuge tube: Eppendorf tube safe-lock 0.5 mL	VWR	20901-505	
Microcentrifuge tube: Eppendorf tube safe-lock 1.5 mL	VWR	21008-959	
Shaker Swip (orbital and horizontal)	Edmund Buehler		
Shaking incubator GFL3032	GFL	3032	
Single-use pipet: serum pipet	Greiner bio-one	612301	
Sterile instruments (scissors, tweezers, curved forceps)			
Tissue culture plate (10 cm)	Greiner bio-one	664960	
Mouse osteoblast-osteoclast co-culture			

1 α ,25-Dihydroxyvitamin D3	Sigma-Aldrich	17936	1000x concentrated stock (10 μ M in ethanol 100%)
Acetone	VWR Chemicals	22065.327	
Bone growth medium (BGM)			α -MEM + 10% heat-inactivated FBS + 1% penicillin-streptomycin
Distilled water	Carl Roth	3478.4	
Ethanol (100% vol/vol)	VWR Chemicals	20821.365	
Ethanol (70% vol/vol)	VWR Chemicals	93003.1006	
Fast red violet LB salt	Sigma-Aldrich	F3381	
Fetal bovine serum (FBS)	Sigma-Aldrich	F7524	
Formalin solution neutral buffered 10%	Sigma-Aldrich	HT501128	
MEM alpha Medium (α -MEM)	Gibco Life Technologies	11900-073	
N,N-Dimethylformamide	Sigma-Aldrich	D4551	
Naphthol AS-MX phosphate	Sigma-Aldrich	N4875	
Osteoclast differentiation medium (DM)			α -MEM + 10% heat-inactivated FBS + 1% penicillin-streptomycin + 1 nM 1,25-(OH) $_2$ -vitamin D3 + 1 μ M prostaglandin E2
Penicillin-streptomycin	Sigma-Aldrich	P4333	
Phosphate-buffered saline 1x (PBS) pH 7.4	Gibco, Thermo Fisher Scientific	10010023	
Prostaglandin E2 (PGE2)	Cayman Chemical Company	9003016	1000x concentrated stock (1 mM in ethanol 100%)
Sodium acetate trihydrate	Sigma-Aldrich	S7670	
Sodium tartrate dibasic dihydrate	Sigma-Aldrich	S8640	
Sterile instruments (scissors, forceps, scalpel)			
TRAP buffer pH 5.0			Dissolve 40 mM sodium acetate, 10 mM sodium tartrate in ultrapure water and adjust to pH 5.
Ultrapure water: Cell culture water pyrogen free	VWR	L0970-500	
24-well suspension culture plate	Greiner bio-one	662102	
24-well tissue culture plate	Greiner bio-one	662160	
50 mL conical tube	Greiner bio-one	227261	
Centrifuge: VWR Mega Star 600R	VWR		
CO2-Incubator: HeraCell 240	Thermo Scientific		
Diamond wafering blade (10.2 cm x 0.3 cm)	Buehler		
Isomet low-speed saw	Buehler		
Petri dish (6 cm)	Greiner bio-one	628,161	
Screw cap glass bottle 20 mL	Carl Roth	EXY4.1	
Single-use sterile syringe 1 mL	Henry Schein Animal Health	9003016	
Single-use pipet: serum pipet	Greiner bio-one	612301	
Sterile needle: Hypodermic needle RW 27 G x 3/4"	Henry Schein Animal Health	9003340	
Mouse calvarial defect model			
Analgesia: Buprenorphine (Bupaq)	Richter Pharma AG		
Anesthesia: Ketamine (Ketamidol), Xylazine (Rompun)	Richter Pharma AG/Bayer HealthCare		

Cold sterilant: SafeSept Max	Henry Schein Animal Health	9882765	
Ethanol (70% vol/vol)	VWR Chemicals	93003.1006	
Eye ointment: VitA POS 5 g	Ursapharm		
Normal saline solution (0.9% sodium chloride solution)	Fresenius Kabi		
Phosphate-buffered saline 1x (PBS) pH 7.4	Gibco, Thermo Fisher Scientific	10010023	
Povidone iodine solution: Braunol 1000 mL	B. Braun	3864154	
Roti-Histofix 4.5% buffered formalin	Carl Roth	2213.6	
Tissue adhesive: Histoacryl 5 x 0.5 mL	B. Braun Surgical	1050060	
Personal protective equipment: surgical gloves, cap and mask, gown	Henry Schein Animal Health	1045073, 1026614, 9009062, 370406	
Sterile instruments (scalpel, scissors, forceps, needle holder)			
Sterile needles: Hypodermic needles RW 27 G x 3/4" and 30 G x 1/2"	Henry Schein Animal Health	9003340, 9003630	
Single-use sterile syringe 1 mL	Henry Schein Animal Health	9003016	
Heating plate: Physitemp	Rothacher Medical	TCAT-2LV	
Sterile gauze swabs	Henry Schein Animal Health	220192	
Sterile disposable scalpel (Figur 20)	Henry Schein Animal Health	9008957	
Surgical/dental drill: Implantmed SI-923	W&H Dentalwerk Bürmoos GmbH	16929000	
Handpiece type S-II	W&H Dentalwerk Bürmoos GmbH	30056000	
Trephine, diameter 4 mm	Hager&Meisinger GmbH	229040	
Irrigation tubing set 2.2 m	W&H Dentalwerk Bürmoos GmbH	4363600	
Periosteal elevator 2 mm / 3 mm	Henry Schein Animal Health	472683	
Non-resorbable suture: Polyester green, DS19, met. 1.5, USP 4/0 75 cm	Henry Schein Animal Health	300715	
<i>In vitro</i> immune responses			
Anesthesia for euthanasia: Ketamine (Ketamidol), Xylazine (Rompun)	Richter Pharma AG/Bayer HealthCare		
Cell Proliferation ELISA, BrdU (colorimetric) Assay Kit	Sigma-Aldrich	11647229001	The kit contains BrdU labeling solution, fixation solution (FixDenat), Anti-BrdU antibody solution, washing buffer and substrate solution.
Concanavalin A (ConA)	MP Biomedicals	150710	
Culture medium splenocytes			RPMI medium + 10% heat-inactivated FBS + 1% penicillin-streptomycin solution + 0.1% gentamicin + 0.2% β -mercaptoethanol + 1% non-essential amino acids
Ethanol (70% vol/vol)	VWR Chemicals	93003.1006	
Fetal bovine serum (FBS)	Gibco, Thermo Fisher Scientific	10500064	
Gentamicin	Gibco, Thermo Fisher Scientific	15750037	

Mouse IL-1 β ELISA Ready-SET-Go!	Invitrogen, Thermo Fisher Scientific	88-7013-88	
Mouse IL-2 ELISA MAX Standard	BioLegend	431001	
Mouse IL-4 ELISA MAX Standard	BioLegend	431101	
Mouse INF- γ ELISA MAX Standard	BioLegend	430801	
Non-essential amino acids solution	Gibco, Thermo Fisher Scientific	11140050	
Penicillin-streptomycin	Gibco, Thermo Fisher Scientific	15140122	
Phosphate-buffered saline 1x (PBS) pH 7.4	Gibco, Thermo Fisher Scientific	10010023	
Red blood cell (RBC) lysis buffer: BD Pharm Lyse	BD Bioscience	555899	
RPMI 1640 medium, HEPES	Gibco, Thermo Fisher Scientific	52400025	
β -Mercaptoethanol	Gibco, Thermo Fisher Scientific	31350010	
50 mL conical tube	Greiner bio-one	227261	
96-well tissue culture plate	Greiner bio-one	655180	
Centrifuge: VWR Mega Star 600R	VWR		
Cell strainer 40 μ m	BD Bioscience	352340	
Infinite M200 Pro microplate reader	Tecan		
Single-use sterile syringe 1 mL	Henry Schein Animal Health	9003016	
Sterile surgical instruments (forceps, glass slides)			
High throughput intraperitoneal model			
Anesthesia: Ketamine (Ketamidol), Xylazine (Rompun)	Richter Pharma AG/Bayer HealthCare		
Cold sterilant: SafeSept Max	Henry Schein Animal Health	9882765	
Ethanol (70% vol/vol)	VWR Chemicals	93003.1006	
Eye ointment: Vita POS 5 g	Ursapharm		
Mouse IL-1 β ELISA Ready-SET-Go!	Invitrogen, Thermo Fisher Scientific	88-7013-88	
Mouse IL-2 ELISA MAX Standard	BioLegend	431001	
Mouse IL-4 ELISA MAX Standard	BioLegend	431101	
Phosphate-buffered saline 1x (PBS) pH 7.4	Gibco, Thermo Fisher Scientific	10010023	
Povidone iodine solution: Braunol 1000 mL	B. Braun	3864154	
Shandon Rapid-Chrome Kwik-Diff Staining Kit	Thermo Scientific	9990700	For differential cell count.
Heating plate: Physitemp	Rothacher Medical	TCAT-2LV	
Hemocytometer: Neubauer chamber	Carl Roth	PC72.1	
Non-resorbable suture: Ethibond Excel 4-0	Ethicon	6683H	
Resorbable suture: Polysorb	Covidien	SL-5628	
Shandon centrifuge for cytopsin	Thermo Scientific		
Single-use sterile syringe 1 mL	Henry Schein Animal Health	9003016	
Sterile gauze swabs	Henry Schein Animal Health	220192	
Sterile needles: Hypodermic needles RW 27 G x 3/4" and 25 G	Henry Schein Animal Health	9003340, 420939	

Sterile surgical instruments (scalpel, scissors, forceps, needle holder)			
Trypan blue solution 0.4%	Gibco, Thermo Fisher Scientific	15250061	
Subchronic subcutaneous model			
Anesthesia: Ketamine (Ketamidol), Xylazine (Rompun)	Richter Pharma AG/Bayer HealthCare		
Cold sterilant: SafeSept Max	Henry Schein Animal Health	9882765	
Ethanol (70% vol/vol)	VWR Chemicals	93003.1006	
Eye ointment: VitA POS 5 g	Ursapharm		
Phosphate-buffered saline 1x (PBS) pH 7.4	Gibco, Thermo Fisher Scientific	10010023	
Povidone iodine solution: Braunol 1000 mL	B. Braun	3864154	
Roti-Histofix 4.5% buffered formalin	Carl Roth	2213.6	
Heating plate: Physitemp	Rothacher Medical	TCAT-2LV	
Non-resorbable suture: Ethibond Excel 4-0	Ethicon	6683H	
Personal protective equipment: surgical gloves, cap and mask, gown	Henry Schein Animal Health	1045073, 1026614, 9009062, 370406	
Single-use sterile syringe 1 mL	Henry Schein Animal Health	9003016	
Sterile gauze swabs	Henry Schein Animal Health	220192	
Sterile instruments (scalpel, scissors, forceps, needle holder)			
Sterile needle: Hypodermic needles RW 27 G x 3/4"	Henry Schein Animal Health	9003340, 420939	