

Materials List for

Fiber Type Identification of Human Skeletal Muscle

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Citation

Latchman, H.K., Wette, S.G., Ellul, D.J., Murphy, R.M., Frankenberg, N.T. Fiber Type Identification of Human Skeletal Muscle. *J. Vis. Exp.* (199), e65750, doi:10.3791/65750 (2023).

Date Published

September 22, 2023

DOI

10.3791/65750

URL

jove.com/video/65750

Materials

Name	Company	Catalog Number	Comments
1x Denaturing buffer	Make according to recipe	Constituents can be sourced from Sigma-Aldrich or other chemical distributing companies	1x denaturing buffer is made by diluting 3x denaturing buffer 1 in 3 v/v with 1X Tris-HCl (pH 6.8). Store at -20 °C.
3x Denaturing buffer	Make according to recipe	Constituents can be sourced from Sigma-Aldrich or other chemical distributors	3x denaturing buffer contains: 0.125M Tris-HCl, 10% glycerol, 4% SDS, 4 M urea, 10% 2-mercaptoethanol, and 0.001% bromophenol blue, pH 6.8. Store at -20 °C.
95% Ethanol	N/A	100% ethanol can be sourced from any company	Diluted to 95% with ultra-pure H ₂ O.
Actin rabbit polyclonal antibody	Sigma-Aldrich	A2066	Dilute 1 in 1,000 with BSA buffer.
Analytical scales	Mettler Toledo	Model number: MSZ042101	
Antibody enhancer	Thermo Fischer Scientific	32110	Product name is Miser Antibody Extender Solution NC.
Beaker (100 mL)	N/A	N/A	
Benchtop centrifuge	Eppendorf	5452	Model name: Mini Spin.
Blocking buffer: 5% Skim milk in Wash buffer.	Diploma	Store bought	
BSA buffer: 1 % BSA/PBST, 0.02 % NaN ₃	BSA: Sigma-Aldrich Bio-Rad Laboratories. NaN ₃ : Sigma-Aldrich	BSA: A6003-25G PBS: 1610780 S2002	10x NaN ₃ : Bovine serum albumin (BSA), Phosphate-buffered saline (PBS), and Sodium azide (NaN ₃). Store at 4 °C.
Cassette opening lever	Bio-Rad Laboratories	4560000	Used to open the precast gel cassettes.
Chemidoc MP Imager	Bio-Rad Laboratories	Model number: Universal hood III	Any imaging system with Stain-Free gel imaging capabilities.
Criterion blotter	Bio-Rad Laboratories	1704070	Includes ice pack, transfer tray, roller, 2 cassette holders, filter paper, foam pads and lid with cables.
Criterion Cell (Bio-Rad)	Bio-Rad Laboratories	1656001	
ECL (enhanced chemiluminescence)	Bio-Rad Laboratories	1705062	Product name: Clarity Max Western ECL Substrate.

Electrophoresis buffer 1x Tris Glycine SDS (TGS)	Bio-Rad Laboratories	1610772	Dilute 10x TGS 1 in 10 with ultra-pure H ₂ O.
Filter paper, 0.34 mm thick	Whatmann	3030917	Bulk size 3 MM, pack 100 sheets, 315 x 335 mm.
Fine tissue dissecting forceps	Dumont	F6521-1EA	Jeweller's forceps no. 5.
Flat plastic tray/lid	N/A	N/A	Large enough to place the membrane on. Ensure the surface is completely flat.
Freeze-drying System	Labconco	7750030	Freezone 4.5 L with glass chamber sample stage.
Freezer -80 °C	N/A	N/A	Any freezer with a constant temperature of -80 °C is suitable.
Gel releasers	1653320	Bio-Rad	Slide under the membrane to gather or move the membrane.
Grey lead pencil	N/A	N/A	
Image lab software	Bio-Rad Laboratories	N/A	Figures refers to software version 5.2.1 but other versions can used.
Incubator	Bio-Rad Laboratories	1660521	Any incubator that can be set to 37 °C would suffice.
Lamp	N/A	N/A	
Magnetic stirrer with flea	N/A	N/A	
Membrane roller	Bio-Rad Laboratories	1651279	Can be purchased in the Transfer bundle pack. However, if this product is not available, any smooth surface cylindrical tube long enough to roll over the membrane would suffice.
Microcentrifuge tubes (0.6 mL)	N/A	N/A	
Mouse IgG HRP secondary	Thermo Fisher Scientific	31430	Goat anti-Mouse IgG (H+L), RRID AB_228341. Dilute at 1 in 20,000 in blocking buffer.
Mouse IgM HRP secondary	Abcam	ab97230	Goat Anti-Mouse IgM mu chain. Use at the same dilution as mouse IgG.
Myosin Heavy Chain I (MHC I) primary antibody	DSHB	A4.840	Dilution range: 1 in 200 to 1 in 500 in BSA buffer.
Myosin Heavy Chain IIa (MHCIIa) primary antibody	DSHB	A4.74	Dilution range: 1 in 200 to 1 in 500 in BSA buffer.
Myosin Heavy Chain IIx (MHCIIx) primary antibody	DSHB	6H1	Dilution range: 1 in 200 to 1 in 500 in BSA buffer.
Nitrocellulose Membrane 0.45 µm	Bio-Rad Laboratories	1620115	For Western blotting.
Petri dish lid	N/A	N/A	
Plastic tweezers	N/A	N/A	
Power Pack	Bio-Rad Laboratories	164-5050	Product name: Basic power supply.
Protein ladder	Thermo Fisher Scientific	26616	PageRuler Prestained Protein Ladder, 10 to 180 kDa.
PVDF Membrane 0.2 µm	Bio-Rad Laboratories	1620177	
Rabbit HRP secondary	Thermo Fisher Scientific	31460	Goat anti-Rabbit IgG (H+L), RRID AB_228341. Dilution same as mouse secondary antibodies.
Rocker	N/A	N/A	
Ruler	N/A	N/A	
Scissors	N/A	N/A	
Stereomicroscope	Motic	SMZ-168	
Stripping buffer	Thermo Fisher Scientific	21059	Product name: Restore Western Blot Stripping Buffer.
Tissue (lint free)	Kimberly-Clark professional	34120	Product name: Kimwipe.

Transfer buffer (1x Tris Glycine buffer (TG), 20% Methanol)	TG: Bio-Rad Laboratories Merck	Methanol: TG buffer: 1610771 1.06018	Methanol: dilute 10x TG buffer with ultra-pure H ₂ O to 1x. Add 100% Methanol to a final concentration of 20% Methanol. Store at 4 °C.
Transfer tray	Bio-Rad Laboratories	1704089	
UV-activation precast gel	Bio-Rad Laboratories	5678085	Gel type: 4–15% Criterion TGX Stain-Free Protein Gel, 26 well, 15 µL.
Vortex	N/A	N/A	
Wash buffer (1x TBST)	10x TBS: Astral Scientific Sigma	Tween 20: BIOA0027-4L	1x TBST recipe: 10x Tris-buffered saline (TBS) is diluted down to 1x with ultra-pure H ₂ O, Tween 20 is added to a final concentration of 0.1%. Store buffer at 4 °C.
Wash containers	Sistema	Store bought	Any tupperware container, that suits the approximate dimensions of the membrane would suffice.