

# Isolation of Mitochondria from Mouse Skeletal Muscle for Respirometric Assays

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jove.com/video/63336

## Materials

Name	Company	Catalog Number	Comments
ADP	Sigma	A5285	Stock at -20 °C
AKT antibody	Cell Signaling Technology	C67E7	Rabbit (Host species)
anti-Goat HRP	Sigma	401504	Rabbit (Host species)
anti-Mouse HRP	Cell Signaling	#7076	Horse (Host species)
Antimycin A	Sigma	A8674	Stock at -20 °C
anti-Rabbit HRP	Cell Signaling	#7074	Goat (Host species)
Ascorbic acid	Sigma	A5960	Stock at RT
Bactin antibody	Sigma	MBS4-48085	Goat (Host species)
Bio-Rad Protein Assay Kit II	Bio-Rad	5000002	It includes 5x Bradford reagent and BSA of known concentration for the standard curve
BSA, fraction V, Fatty Acid-Free	Calbiochem	126575	Stock at 4 °C
C tube	Miltenyi Biotec	130-093-237	Purple lid
Calnexin antibody	ThermoFisher	MA3-027	Mouse (Host species)
D-mannitol	Sigma	M4125	Stock at RT
EDTA	BDH	280254D	Stock at 4 °C
EGTA	Sigma	E-4378	Stock at RT
FCCP	Sigma	C2920	Stock at -20 °C
gentleMACS Dissociator	Miltenyi Biotec	130-093-235	Homogenizer
HEPES	Sigma	H3375	Stock at RT
HSP70 antibody	Proteintech	10995-1-AP	Rabbit (Host species)
LDH-A antibody	Santa Cruz Biotechnology	SC27230	Goat (Host species)
Magnesium chloride	ChemCruz	sc-255260A	Stock at RT
Malic acid	Sigma	P1645	Stock at RT
Microplate spectrophotometer	BMG LABTECH GmbH	POLARstar OMEGA S/N 415-0292	Stock at RT
Milli-Q water	Millipore system	F7HA17757A	Ultrapure water
mtTFA antibody	Santa Cruz Biotechnology	SC23588	Goat (Host species)

Na <sup>+</sup> /K <sup>+</sup> -ATPase $\alpha$ 1 antibody	Novus Biologicals	NB300-14755	Mouse (Host species)
Oligomycin	Sigma	O4876	Stock at -20 °C
Palmitoyl-L-carnitine	Sigma	P1645	Stock at -20 °C
PBS tablets	Sigma	P4417-100TAB	1x stock at RT
Potassium dihydrogen phosphate	ChemCruz	sc-203211	Stock at RT
Potassium hydroxide	Sigma	60377	Stock at RT
Pyruvic acid	Sigma	107360	Stock at 4 °C
Rotenone	Sigma	R8875	Stock at -20 °C
Seahorse XF24 mitochondrial flux analyzer	Agilent Technologies	420179	XFe24 model
Seahorse XFe24 FluxPak mini	Agilent Technologies	102342-100	The kit includes cartridges, microplates, and calibrant solution
Succinate	Sigma	S7626	Stock at RT
Sucrose	Sigma	S9378	Stock at RT
TIMM23 antibody	Abcam	ab230253	Rabbit (Host species)
TMPD	Sigma	T7394	Stock at -20 °C
TOMM20 antibody	Abcam	ab56783	Mouse (Host species)
VDAC antibody	Abcam	ab15895	Rabbit (Host species)