Mouse PDGF Stimulated NIH/3T3 Whole Cell Lysate

Cat. No. NIH/3T3-065MCL **Lot. No.** (See product label)

SPECIFICATION

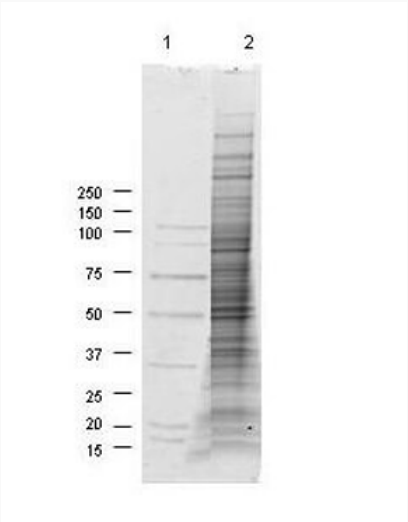
Product Overview	Ready-to-use whole cell lysates are derived from cell lines or tissues using highly refined extraction protocols to ensure exceptionally high quality, protein integrity and lot-to-lot reproducibility. All extracts are tested by SDS-PAGE using 4-20% gradient gels and immunoblot analysis using antibodies to key cell signaling components to confirm the presence of both high molecular weight and low molecular weight proteins.
Species	Mouse
Form	Liquid (sterile filtered)
Specificity	The cells were grown in medium supplemented with 10% fetal bovine serum. Cells were washed with PBS and then incubated on ice in modified RIPA buffer to lyse the cells. Protein integrity was ensured using a cocktail of protease inhibitors with broad specificity for the inhibition of aspartic, cysteine, and serine proteases as well as aminopeptidases (0.1 mM AEBSF HCl, 0.08 µM Aprotinin, 5 µM Bestatin, 1.5 µM E-64, 2 µM Leupeptin Hemisulfate, 1 µM Pepstatin A). Phosphatase inhibitors 1 mM NaF and 1 mM Na3VO4 were also added. Cell debris was removed by centrifugation. Protein concentration was determined by a modified Lowry assay using a commercially available kit. Protein concentration was adjusted to 2 mg/mL and then an equal volume of 2× SDS-PAGE sample buffer was added.
Applications	WB, SDS-PAGE

Tel: 1-631-559-9269 1-516-512-3133
Email: info@creative-biomart.com Fax: 1-631-938-8127
45-1 Ramsey Road, Shirley, NY 11967, USA

Cell Line	NIH Swiss Webster Mouse embryo
Lysate Fractionation	Whole Cell Lysate
Lysate Stimulation	PDGF
Culture Type	Tissue Culture
Induction	Platelet Derived Growth Factor (100 ng/mL)
Concentration	1.0 mg/mL by modified Lowry assay
Buffer	1× SDS-PAGE Sample Buffer (62.5 mM Tris HCl, 2% SDS, 10% Glycerol and 0.005% bromophenol blue, pH 6.8)
Preservative	None
Stabilizer	10% (v/v) Glycerol
Shipping	Dry Ice
Storage	Store vial at -70 centigrade or COLDER. For extended storage, aliquot contents to minimize freeze/thaw cycles.
Expiration	Expiration date is three months from date of receipt.

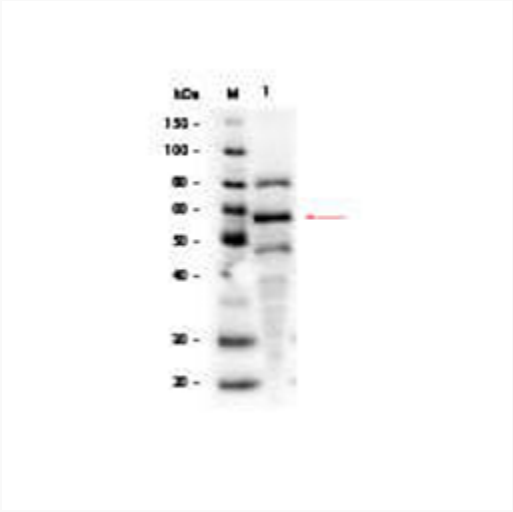
Tel: 1-631-559-9269 1-516-512-3133
Email: info@creative-biomart.com Fax: 1-631-938-8127
45-1 Ramsey Road, Shirley, NY 11967, USA

SDS-PAGE



Coomassie stained SDS-PAGE of 30 µg of Mouse Derived NIH/3T3 Whole Cell Lysate (Ready-to-Use) resolved on a 4-20% gradient gel under non-reducing conditions (lane 2). Molecular weight standards are shown in lane 1.

Western Blot



Western Blot of NIH/3T3 Whole Cell Lysate PDGF Stimulated.

Tel: 1-631-559-9269 1-516-512-3133
Email: info@creative-biomart.com Fax: 1-631-938-8127
45-1 Ramsey Road, Shirley, NY 11967, USA

Lane 1: NIH/3T3 Whole Cell Lysate PDGF Stimulated.

Load: 2 µg per lane.

Primary antibody: AKT1 pS473 Antibody at 1:1,000 overnight at 4 centigrade.

Secondary antibody: HRP mouse secondary antibody at 1:40,000 for 30 min at RT.

Block: Blocking Buffer for Fluorescent Western Blotting for 30 min at RT.

Predicted/Observed size: 55 kDa, 55 kDa for AKT1 pS473.

Tel: 1-631-559-9269 1-516-512-3133
Email: info@creative-biomart.com Fax: 1-631-938-8127
45-1 Ramsey Road, Shirley, NY 11967, USA