

Human MG-132 Treated HEK293 Whole Cell Lysate

Cat. No. HEK293-033HCL 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

Human

Form

Liquid

Specificity

HEK293 cells were grown in medium supplemented with 10% fetal bovine serum. Cells were treated O/N with 0.2 µg/mL of MG-132 in order to reduce degradation of ubiquitin-conjugated proteins by the proteasome. 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.

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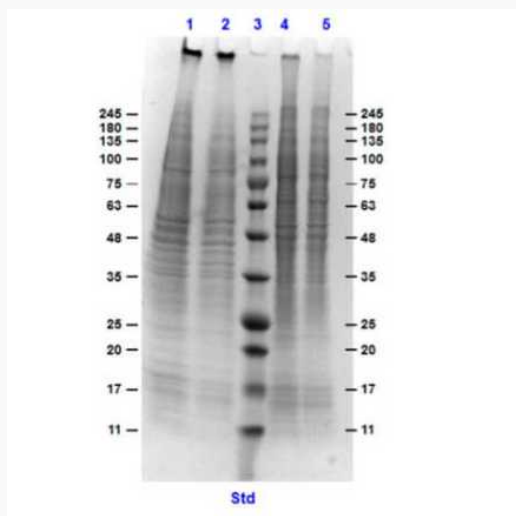
Applications	SDS-PAGE
Cell Line	Human Epithelial (Kidney)
Lysate Fractionation	Whole Cell Lysate
Lysate Stimulation	MG-132
Culture Type	Tissue Culture
Induction	MG-132
Concentration	1 mg/mL by modified Lowry assay
Buffer	1× SDS-PAGE Sample Buffer (62.5 mM Tris HCl, 2% SDS, 10% Glycerol and 0.005% bromophenol 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.

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SDS-PAGE



SDS-PAGE of HEK293 Whole Cell Lysate MG-132 Stimulated.

Lane 1: HEK293 unstimulated reduced.

Lane 2: HEK293 MG-132 stimulated reduced.

Lane 3: Molecular Weight Marker.

Lane 4: HEK293 unstimulated non-reduced.

Lane 5: HEK293 MG-132 stimulated non-reduced.

Load 10 ng.