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Hep2 Whole Cell Lysate



Image



Publication



Overview

Quantity: 500 µg **Protein Species:** Human Human Cells Species of Lysate: Application: Western Blotting (WB)

Product Details

Specificity:

Hep2 cells were grown in Dulbecco's medium supplemented with 10% fetal bovine serum. Cells were washed with PBS and then incubated on ice in modified RIPA buffer, containing 150 mM sodium chloride, 50 mM Tris HCl, pH 7.4, 1 mM EDTA, 1.0% NP-40, 0.5% sodium deoxycholic acid, 0.1% SDS and 0.01% (w/v) sodium azide 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 2X SDS-PAGE sample buffer was added.

Characteristics:

Cell Line: Human HEp2 (HeLa contaminant)

Induction: None (Control)

Lysate Fraction:

Whole Cell Lysate

Lysate Type:

Cell Lysate

Lysed Cells:

HepG2 Cells

Target Details

Background:

Hep2 Whole Cell Lysate Ready-to-use whole cell lysates produced 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.

Synonyms: Hep2, Lysate, Whole Cell Lysate, Hep2 Lysate

Application Details

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ADD	lication	notes.

Ready-to-use lysates are especially prepared as positive controls for separation by SDS-PAGE and subsequent western blot analysis. Lysates are prepared in denaturing buffer WITHOUT dissociating agents (i.e. no 2-mercaptoethanol or dithiothreitol has been added). Heat lysate to 95° C for 5 minutes and rapidly cool. If dissociating conditions are desired, add reducing agent prior to heating. The recommended loading volume per lane is 10-20 µl depending on the size format of your gel.

Comment:

Lysate Fractionation: Whole Cell Lysate
Lysate Stimulation: Not Stimulated
Lysate Tissue Culture: Tissue Culture

Restrictions:

For Research Use only

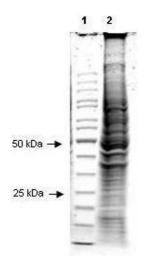
Handling

Format:	Liquid	
Concentration:	1.0 mg/mL	
Buffer:	1X SDS-PAGE Sample Buffer (62.5 mM Tris HCl, 2% SDS, 10% Glycerol and 0.005% bromophenol blue, pH 6.8)	
Handling Advice:	Ready-to-use lysates are especially prepared as positive controls for separation by SDS-PAGE and subsequent western blot analysis. Lysates are prepared in denaturing buffer WITHOUT dissociating agents (i.e. no 2-mercaptoethanol or dithiothreitol has been added). Heat lysate to 95 °C for 5 minutes and rapidly cool. If dissociating conditions are desired, add reducing agent prior to heating. The recommended loading volume per lane is 10-20 µl depending on the size format of your gel.	
Storage:	-80 °C	
Expiry Date:	3 months	

Product cited in:

Laffy, Dodev, Macpherson, Townsend, Lu, Dunn-Walters, Fraternali: "Promiscuous antibodies characterised by their physico-chemical properties: From sequence to structure and back." in: **Progress in biophysics and molecular biology**, Vol. 128, pp. 47-56, (2017) (PubMed).

Validation report #101185 for Enzyme Immunoassay (EIA)



SDS-PAGE

Image 1. Coommassie stained SDS-PAGE of 20 μ l of Human Derived Hep2 Whole Cell Lysate (Ready-to-Use) separated in a 4-20% gradient gel under reducing conditions (lane 2). Molecular weight standards are shown in lane 1.