

U-251 Whole Cell Lysate - W09-001-GY4

Code: W09-001-GY4

Size: 500 µg

Product Description: U-251 Whole Cell Lysate - W09-001-GY4

Concentration: 1.0 mg/ml by BCA assay

PhysicalState: Liquid (sterile filtered)

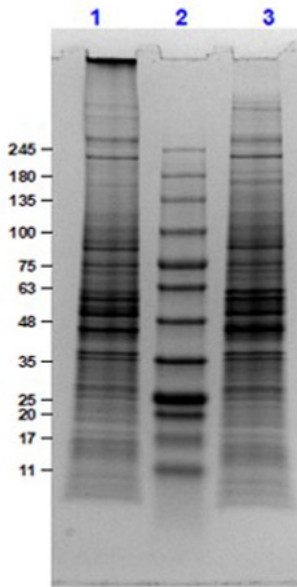
Label	Unconjugated
Buffer	1X SDS-PAGE Sample Buffer (62.5 mM Tris HCl, 2% SDS, 10% Glycerol and 0.005% bromophenol blue, pH 6.8)
Stabilizer	10% (v/v) Glycerol
Preservative	None
Storage Condition	Store vial at -70°C or COLDER. For extended storage, aliquot contents to minimize freeze/thaw cycles.
Synonyms	U251 Whole Cell Lysate, U-251 WCL, U-251 Lysate, U-251 MG Lysate, U-251-MG Lysate, U-251_MG Lysate, U251-MG Lysate, U251MG, U251N
Application Note	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.
Background	Ready-to-use whole cell lysates produced by Rockland Immunochemicals 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.
Purity And Specificity	U-251 cells were grown in Eagle's Minimum Essential 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 BCA assay using a commercially available kit. Protein concentration was adjusted to 1 mg/mL using 2X SDS-PAGE Sample buffer.
Western Blot	User Optimized
Expiration	Expiration date is three (3) months from date of opening.

Related Products

600-401-MN5	LC3B Antibody600-401-MN5
W09-000-360	MCF-7 Whole Cell Lysate - W09-000-360
MB-210-0500	Opal Prestained Protein Standard 10-245kDaMB-210-0500
600-401-MG4	Beclin 1 Antibody600-401-MG4

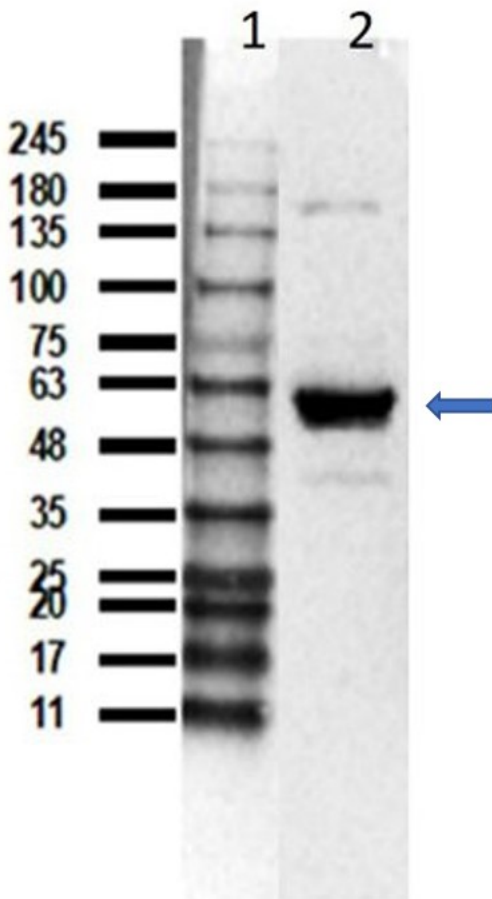
Images

1	SDS-PAGE Results of U-251 Whole Cell Lysate. Lane 1: U-251 Whole Cell Lysate (10µg) Reduced. Lane 2: Opal Prestained Molecular Weight Marker (p/n MB-210-0500). Lane 3: U-251 Whole Cell Lysate (10µg) Non-Reduced. Results show wide range of molecular weight bands with no sign of degradation.
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Western Blot results of Rabbit Anti-Beclin 1 Antibody using U-251 lysate. Lane 1: Opal Prestain Molecular Weight Marker (p/n MB-210-0500). Lane 2: U251-MG whole cell lysate (35ug) [+]. Primary Antibody: Anti-Beclin1 at 1:1000 overnight at 2-8°C. Secondary Antibody: Goat Anti-Rabbit IgG HRP (p/n 611-103-122) at 1:40,000 for 60min at RT. Block: 5% BLOTTO/TBST (p/n B501-0500). Expected MW: ~51kDa. Observed MW: ~53kDa.



Disclaimer

This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information. All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation. All properties listed are typical characteristics and are not specifications. All suggestions and data are offered in good faith but without guarantee as conditions and methods of use of our products are beyond our control. All claims must be made within 30 days following the date of delivery. The prospective user must determine the suitability of our materials before adopting them on a commercial scale. Suggested uses of our products are not recommendations to use our products in violation of any patent or as a license under any patent of Rockland Immunochemicals, Inc. If you require a commercial license to use this material and do not have one, then return this material, unopened to: Rockland Inc., P.O. BOX 5199, Limerick, Pennsylvania, USA.

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