

Product Datasheet

U-251 Whole Cell Lysate NBP3-06998

Unit Size: 500 ug

Store at -70C. Avoid freeze-thaw cycles.

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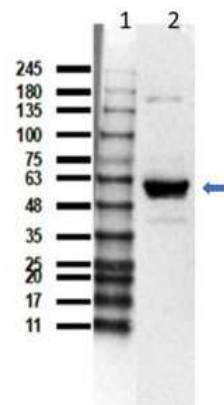


NBP3-06998**U-251 Whole Cell Lysate**

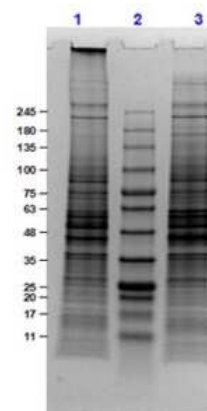
Product Information	
Unit Size	500 ug
Concentration	Please contact technical services for concentration.
Storage	Store at -70C. Avoid freeze-thaw cycles.
Preservative	No Preservative
Purity	Multi-step
Buffer	1X SDS-PAGE Sample Buffer (62.5 mM Tris HCl, 2% SDS, 10% Glycerol and 0.005% bromophenol blue, pH 6.8), 10% (v/v) Glycerol
Product Description	
Description	Store vial at -70C or COLDER. For extended storage, aliquot contents to minimize freeze/thaw cycles.
Species	Human
Preparation Method	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 uM Aprotinin, 5 uM Bestatin, 1.5 uM E-64, 2 uM Leupeptin Hemisulfate, 1 uM 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.
Lysate Type	Cell
Lysate Subcellular Fraction	Whole
Product Application Details	
Applications	Western Blot, SDS-Page
Recommended Dilutions	Western Blot, SDS-Page
Application Notes	This product has been tested by SDS-PAGE and western blot. 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 95C 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 uL depending on the size format of your gel.

Images

Western Blot: U-251 Whole Cell Lysate [NBP3-06998] - Western Blot results of Rabbit Anti-Beclin 1 Antibody using U-251 Whole Cell lysate. Lane 1: Opal Prestain Molecular Weight Marker. Lane 2: U251-MG whole cell lysate (35ug) [+]. Primary Antibody: Anti-Beclin1 at 1:1000 overnight at 2-8C. Secondary Antibody: Goat Anti-Rabbit IgG HRP at 1:40,000 for 60min at RT. Block: 5% BLOTTO/TBST. Expected MW: ~51kDa. Observed MW: ~53kDa.



SDS-Page: U-251 Whole Cell Lysate [NBP3-06998] - SDS-PAGE Results of U-251 Whole Cell Lysate. Lane 1: U-251 Whole Cell lysate (10ug) Reduced. Lane 2: Opal Prestained Molecular Weight Marker. Lane 3: U-251 Whole Cell lysate(10ug) Non-Reduced. Results show wide range of molecular weight bands with no sign of degradation.





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Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Lysates are guaranteed for 6 months from date of receipt.

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