Product Datasheet

OS9 Antibody - BSA Free BC100-520

Unit Size: 0.1 ml

Store at 4C. Do not freeze.

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BC100-520

OS9 Antibody - BSA Free

Product Information	
Unit Size	0.1 ml
Concentration	0.2 mg/ml
Storage	Store at 4C. Do not freeze.
Clonality	Polyclonal
Preservative	0.05% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	Tris-Glycine and 0.15M NaCl
Target Molecular Weight	75 kDa
Product Description	
Host	Rabbit
Gene ID	10956
Gene Symbol	OS9
Species	Human, Mouse, Rat
Immunogen	A C-terminal synthetic peptide made to isoform 1 of the human OS9 protein sequence (between residues 600-667). There are 3 isoforms of this protein. The immunogen used for this antibody is conserved in isoforms 1, 2, and 3 of the human OS9 protein.
Product Application Details	5
Applications	Western Blot, Simple Western, Immunocytochemistry/ Immunofluorescence
Recommended Dilutions	Western Blot 1:1000, Simple Western 1:100, Immunocytochemistry/ Immunofluorescence 1:500
Application Notes	 In Western Blot, a band can be seen at approx. 75 kDa. In Simple Western only 10 - 15 uL of the recommended dilution is used per data point. See <u>Simple Western Antibody Database</u> for Simple Western validation: Tested in HepG2 lysate 0.5 mg/mL, separated by Size, antibody dilution of 1:100, apparent MW was 25 kDa. Separated by Size-Wes, Sally Sue/Peggy Sue. The observed molecular weight of the protein may vary from the listed predicted molecular weight due to post translational modifications, post translation cleavages, relative charges, and other experimental factors.



Images

inages	
Western Blot: OS9 Antibody [BC100-520] - Initial characterization of the OS-9 protein. OS-9 expression in various human cell lines. Equal protein amounts of total cell lysates were used for SDS-PAGE and subsequent Western blotting. For each cell line, two independent samples are shown. Endogenous OS-9 was detected with a polyclonal antibody raised against a peptide corresponding to amino acids 600-667 of isoform 1 of OS-9. Image collected and cropped by CiteAb from the following publication (https://dx.plos.org/10.1371/journal.pone.0019151), licensed under a CC-BY license.	A MCF-7 Hep3B U2OS fibroblast HUVEC
Immunocytochemistry/Immunofluorescence: OS9 Antibody [BC100-520] - OS9 antibody was tested in HeLa cells with FITC (green). Nuclei and alpha-tubulin were counterstained with Dapi (blue) and Dylight 550 (red).	
Western Blot: OS9 Antibody [BC100-520] - WB analysis of OS9 in 1. A431 cell lysate, 2. HepG2 cell lysate, 3. MCF7 cell lysate and 4. 3T3 cell lysate.	250> 150> 100> 75> 50> 37> 25> 20> 15> 10> 15> 10> 15> 10> 15> 10> 15> 15> 10> 15> 15> 10> 10> 15> 15> 10> 15> 10> 15> 10> 15> 10> 15> 15> 15> 15> 15> 15> 15> 15
Simple Western: OS9 Antibody [BC100-520] - Simple Western lane view shows a specific band for OS9 in 0.5 mg/ml of HepG2 lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.	kDa 230- 180- 186- 40- 12-



Western Blot: OS9 Antibody [BC100-520] - Initial characterization of the C OS-9 protein.(A) OS-9 expression in various human cell lines. Equal protein amounts of total cell lysates were used for SDS-PAGE & subsequent Western blotting. For each cell line, two independent samples are shown. Endogenous OS-9 was detected with a polyclonal antibody raised against a peptide corresponding to amino acids 600–667 of isoform 1 of OS-9. (B) Protein stability assay of endogenous OS-9. U2OS cells were treated with the translational inhibitor cycloheximide (100 µM). At indicated time points, whole cell lysates were analysed by immunoblotting. (C) Effect of hypoxia on OS-9 expression. For hypoxia, UT-7 cells were exposed to 1% O2 for 24 h prior to Western blot analysis. To determine any influence of HIF-1a on OS-9 expression under normoxia, cells were incubated with the prolyl hydroxylase inhibitor DMOG (0.5 mM) for 24 h. (D) Protein interaction between OS-9 & PHD2 in vitro. For co-immunoprecipitation, U2OS cells were transiently co-transfected with the plasmids pOS-9-V5 & pPHD2-His, lysed in NP40 buffer, & subjected to immunoisolation with anti-V5 antibody recognizing OS-9 by its V5-tag. OS-9 & its associated proteins were separated by SDS-PAGE & analyzed by Western blot (lane 2). As controls, samples of untransfected (lane 1) cells or cells transfected with a single plasmid (lanes 3-4) were loaded. Representative Western blots are shown for each subfigure. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/21559462), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Western Blot: OS9 Antibody [BC100-520] - Initial characterization of the OS-9 protein.(A) OS-9 expression in various human cell lines. Equal protein amounts of total cell lysates were used for SDS-PAGE & subsequent Western blotting. For each cell line, two independent samples are shown. Endogenous OS-9 was detected with a polyclonal antibody raised against a peptide corresponding to amino acids 600-667 of isoform 1 of OS-9. (B) Protein stability assay of endogenous OS-9. U2OS cells were treated with the translational inhibitor cycloheximide (100 μ M). At indicated time points, whole cell lysates were analysed by immunoblotting. (C) Effect of hypoxia on OS-9 expression. For hypoxia, UT-7 cells were exposed to 1% O2 for 24 h prior to Western blot analysis. To determine any influence of HIF-1α on OS-9 expression under normoxia, cells were incubated with the prolyl hydroxylase inhibitor DMOG (0.5 mM) for 24 h. (D) Protein interaction between OS-9 & PHD2 in vitro. For co-immunoprecipitation, U2OS cells were transiently co-transfected with the plasmids pOS-9-V5 & pPHD2-His, lysed in NP40 buffer, & subjected to immunoisolation with anti-V5 antibody recognizing OS-9 by its V5-tag. OS-9 & its associated proteins were separated by SDS-PAGE & analyzed by Western blot (lane 2). As controls, samples of untransfected (lane 1) cells or cells transfected with a single plasmid (lanes 3–4) were loaded. Representative Western blots are shown for each subfigure. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/21559462), licensed under a CC-BY license. Not internally tested by Novus Biologicals.







Western Blot: OS9 Antibody [BC100-520] - Initial characterization of the D OS-9 protein.(A) OS-9 expression in various human cell lines. Equal protein amounts of total cell lysates were used for SDS-PAGE & subsequent Western blotting. For each cell line, two independent samples are shown. Endogenous OS-9 was detected with a polyclonal antibody raised against a peptide corresponding to amino acids 600–667 of isoform 1 of OS-9. (B) Protein stability assay of endogenous OS-9. U2OS cells were treated with the translational inhibitor cycloheximide (100 µM). At indicated time points, whole cell lysates were analysed by immunoblotting. (C) Effect of hypoxia on OS-9 expression. For hypoxia, UT-7 cells were exposed to 1% O2 for 24 h prior to Western blot analysis. To determine any influence of HIF-1a on OS-9 expression under normoxia, cells were incubated with the prolyl hydroxylase inhibitor DMOG (0.5 mM) for 24 h. (D) Protein interaction between OS-9 & PHD2 in vitro. For co-immunoprecipitation, U2OS cells were transiently co-transfected with the plasmids pOS-9-V5 & pPHD2-His, lysed in NP40 buffer, & subjected to immunoisolation with anti-V5 antibody recognizing OS-9 by its V5-tag. OS-9 & its associated proteins were separated by SDS-PAGE & analyzed by Western blot (lane 2). As controls, samples of untransfected (lane 1) cells or cells transfected with a single plasmid (lanes 3-4) were loaded. Representative Western blots are shown for each subfigure. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/21559462), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Western Blot: OS9 Antibody [BC100-520] - Cellular localization of OS-9 & PHD2.(A) A lectin gel-shift assay was conducted to test for glycosylated proteins. Total cell lysates of U2OS cells were incubated in the presence or absence of the endoglycosidases EndoH & PNGaseF for 6 h at 37°C. Digest products were separated on a reducing 10% SDS-PAGE gel which contained concanavalin A co-polymerized in the top layer of the separating gel to retard mobility of glycosylated proteins [56]. Glycosylated OS-9 is indicated as 'g', deglycosylated OS-9 as 'd'. (B) Detection of OS-9 & PHD2 in the nuclear fraction. HEK293 cells with & without transfection of the plasmid pcDNA3-OS-9 were separated into nuclear fraction (N) & postnuclear supernatant (PS), the latter containing cytoplasm & organelles. Western blot analysis included BiP, GAPDH & lamin A as typical marker proteins for the ER, the cytoplasm & the nucleus, respectively. (C) Detection of OS-9 & PHD2 in the cytoplasm. HEK293 cells were co-transfected with pcDNA3-OS-9 & pPHD2-V5. For hypoxia, cells were exposed to 3% O2 for 4 h. Cells were treated with 50 µg/ml digitonin & centrifuged to obtain a cytoplasmic (C) & an organelle fraction (O) & subjected to immunoblotting. (D) Isolation of cellular endomembranes. HEK293 cells were lysed mechanically by several passages through a 301/2G needle. The postnuclear supernatant was processed further by ultra-centrifugation to separate the organelles (O) from the cytosol (C). High salt treatment (1 M KCI) of the organelle fraction produced a wash fraction (W) that contained dissociated peripheral membrane proteins. For immunoblot analysis of subcellular fractionations, cell aliquots were normalized for cell number prior to loading (B–D). Representative Western blots are shown for each subfigure. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/21559462), licensed under a CC-BY license. Not internally tested by Novus Biologicals.





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Western Blot: OS9 Antibody [BC100-520] - OS-9 shows no effect on regulation of HIF-1α.Total cell lysates were used for SDS-PAGE & subsequent Western blotting. To generate nearly anoxic conditions, cells were exposed to an oxygen consuming chemical system or to 1% or 3% O2 for 4 h to generate hypoxia. (A) U2OS, HeLa & Hep3B cells were transiently transfected with the plasmid pOS-9-V5 48 h prior to the experiment. Lamin A & actin were used as loading controls. (B) U2OS cells were subjected to ER stress by incubation either with tunicamycin (1 µg/ml) or thapsigargin (0.5 µg/ml) for 20 h. To detect HIF-1a under normoxia, cells were treated with DMOG (1 mM) for 4 h. A sample of DMSO-only treated cells was loaded to exclude unspecific side effects of the solvent. (C) U2OS cells were transduced with lentiviral construct pLKO.1-shRNA-OS-9 (shOS-9) mediating a stable knockdown of OS-9 expression. Control cells (c) were transduced with plasmid pLKO.1-puro. Representative Western blots are shown for each subfigure. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/21559462), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Elisa Fasana, Ilaria Fregno, Carmela Galli, Tatiana Soldà, Maurizio Molinari ER-to-lysosome-associated degradation acts as failsafe mechanism upon ERAD dysfunction EMBO Reports 2024-05-21 [PMID: 38773321]

Fasana E, Fregno I, Galli C, Molinari M FAM134B regulates ER-to-lysosome-associated degradation of misfolded proteins upon pharmacologic or genetic inactivation of ER-associated degradation bioRxiv 2023-11-28 (WB, KD, Mouse)

Sun L, Xu C, Chen G et al. A Novel Role of OS-9 in the Maintenance of Intestinal Barrier Function from Hypoxiainduced Injury via p38-dependent Pathway. Int. J. Biol. Sci. 2015-05-22 [PMID: 25999789] (WB, Human)

Brockmeier U, Platzek C, Schneider K et al. The function of hypoxia-inducible factor (HIF) is independent of the endoplasmic reticulum protein OS-9. PLoS One. 2011-04-29 [PMID: 21559462] (WB, Human)

Dougan SK, Hu CC, Paquet ME et al. Derlin-2-deficient mice reveal an essential role for protein dislocation in chondrocytes Mol Cell Biol 2011-03-01 [PMID: 21220515] (WB, Mouse)

Bernasconi R, Galli C, Noack J et al. Role of the SEL1L:LC3-I Complex as an ERAD Tuning Receptor in the Mammalian ER. Mol Cell. 2012-05-23 [PMID: 22633958] (WB, Human)





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NBL1-13982	OS9 Overexpression Lysate
BC100-520PEP	OS9 Antibody Blocking Peptide
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NBP2-24891	Rabbit IgG Isotype Control

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