

Product Datasheet

beta Tubulin Antibody - BSA Free NB600-936

Unit Size: 0.1 ml

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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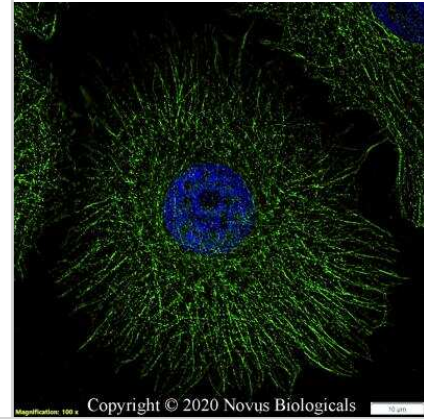
NB600-936

beta Tubulin Antibody - BSA Free

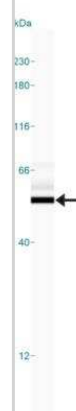
| Product Information | |
|-----------------------------|---|
| Unit Size | 0.1 ml |
| Concentration | 1.0 mg/ml |
| Storage | Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles. |
| Clonality | Polyclonal |
| Preservative | 0.02% Sodium Azide |
| Isotype | IgG |
| Purity | Immunogen affinity purified |
| Buffer | PBS |
| Target Molecular Weight | 50 kDa |
| Product Description | |
| Host | Rabbit |
| Gene Symbol | TUBB |
| Species | Human, Mouse, Rat, Porcine, Bovine, C. elegans, Chicken, Chinese Hamster, Insect, Invertebrate, Primate, Xenopus, Zebrafish |
| Reactivity Notes | Use in Rat reported in scientific literature (PMID:34519641). Invertebrate reactivity reported in scientific literature (PMID: 28114363). C. elegans reactivity reported in scientific literature (PMID: 27690361). Insect reactivity reported from a verified customer review. |
| Marker | Microtubule Marker |
| Immunogen | A synthetic peptide made to the N-terminal region of human beta Tubulin (within residues 1-100). [Swiss-Prot: P07437] |
| Product Application Details | |
| Applications | Western Blot, Simple Western, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin |
| Recommended Dilutions | Western Blot 1:1000, Simple Western 1:1000. Use reported in scientific literature (PMID 35008713), Immunohistochemistry 1:1000 - 1:2000, Immunocytochemistry/ Immunofluorescence 1:500 - 1:1000, Immunohistochemistry-Paraffin 1:1000 - 1:2000 |
| Application Notes | In Simple Western only 10 - 15 uL of the recommended dilution is used per data point. Separated by Size-Wes, Sally Sue/Peggy Sue. SCW validated using murine pancreatic cancer cells. |

Images

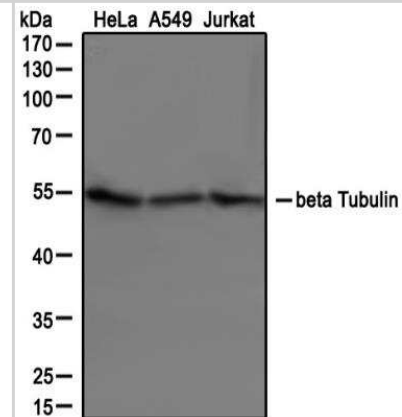
Immunocytochemistry/Immunofluorescence: beta Tubulin Antibody [NB600-936] - MCF7 cells were fixed in 4% paraformaldehyde for 10 minutes and permeabilized in 0.05% Triton X-100 in PBS for 5 minutes. The cells were incubated with anti- (NB600-936) at 2 ug/ml overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at a 1:1000 dilution for 60 minutes. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 100X objective and digitally deconvolved.



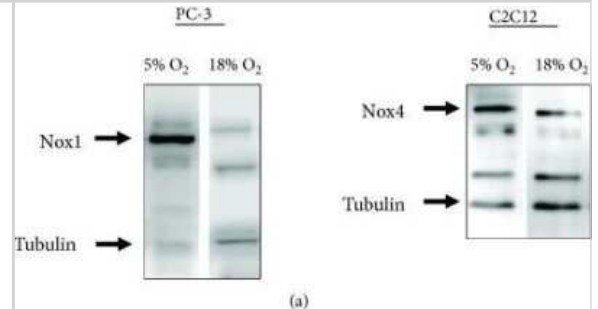
Simple Western: beta Tubulin Antibody [NB600-936] - Lane view shows a specific band for beta Tubulin in 0.05 mg/ml of MCF-7 lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.



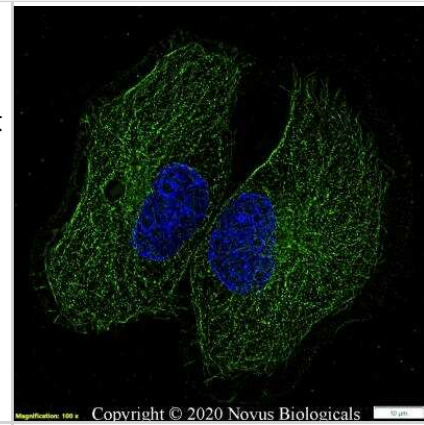
Western Blot: beta Tubulin Antibody [NB600-936] - Analysis of extracts from HeLa, A549 and Jurkat cells using NB600-936 Beta-tubulin antibody at 1:1000



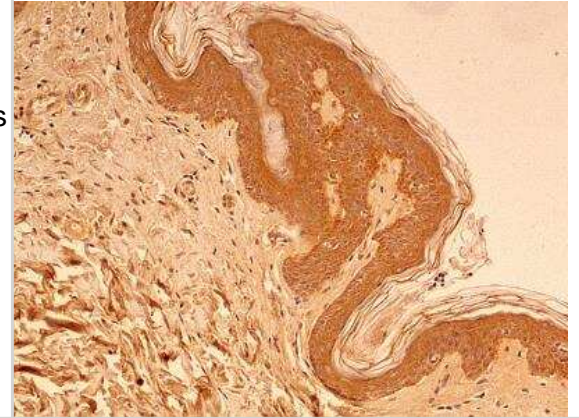
Western Blot: beta Tubulin Antibody - BSA Free [NB600-936] - beta Tubulin Antibody [NB600-936] - Reduced levels of NADPH oxidases 1 and 4 at 18% versus 5% O₂. (a) Representative Western blots showing Nox1 and Beta-tubulin in PC-3 cells or Nox4 and Beta-tubulin in C2C12 cells, at 5% and 18% O₂. (b, c) Average Nox1 signal How Supraphysiological Oxygen Levels in Standard Cell Culture Affect Oxygen-Consuming Reactions. Image collected and cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/30363917/>) licensed under a CC-BY license.



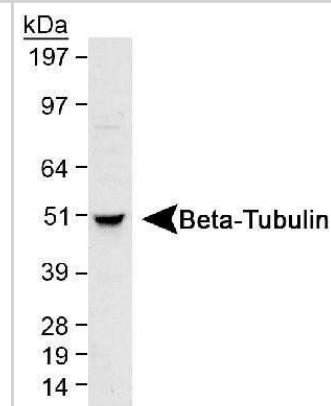
Immunocytochemistry/Immunofluorescence: beta Tubulin Antibody [NB600-936] - PC12 cells were fixed in 4% paraformaldehyde for 10 minutes and permeabilized in 0.05% Triton X-100 in PBS for 5 minutes. The cells were incubated with anti-beta Tubulin Antibody (NB600-936) at 2 ug/ml overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at a 1:1000 dilution for 60 minutes. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 100X objective and digitally deconvolved.



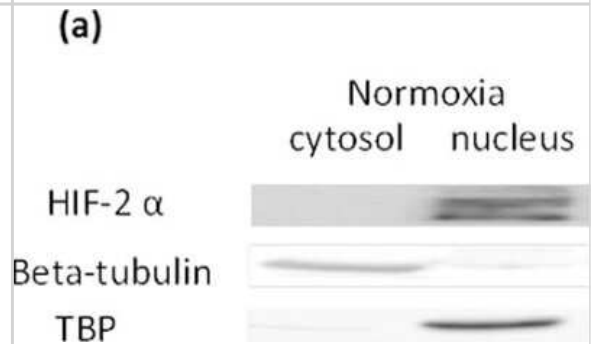
Immunohistochemistry-Paraffin: beta Tubulin Antibody [NB600-936] - Analysis of FFPE tissue section of normal human skin using 1:2000 dilution of beta Tubulin antibody. Intense cytoplasmic staining of beta Tubulin (TUBB) protein was observed in various cells of the epidermal as well as the dermal cells [10X Magnification].



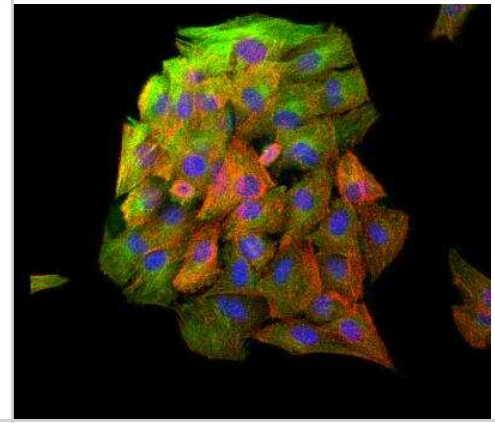
Western Blot: beta Tubulin Antibody [NB600-936] - Analysis in HeLa whole cell lysate at a 1:1,000 dilution.



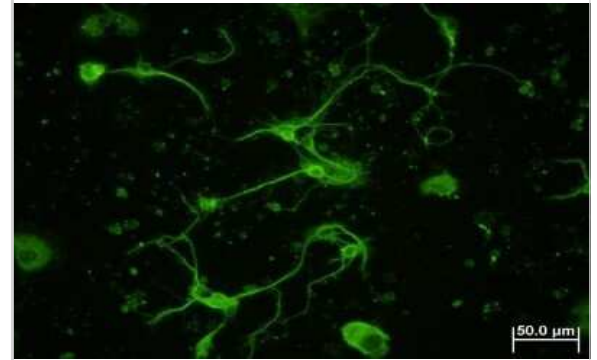
Western Blot: beta Tubulin Antibody [NB600-936] - Expression of pluripotency related markers and a sphere culture of BEAS-2B under normoxia. HIF-2alpha expression was detected in the nucleus of BEAS-2B under normoxia by western blotting. Beta tubulin and TATA binding protein (TBP) were used as protein markers for the cytosol and nucleus fraction, respectively. Image collected and cropped by CiteAb from the following publication (<https://www.nature.com/articles/srep29311>), licensed under a CC-BY license.



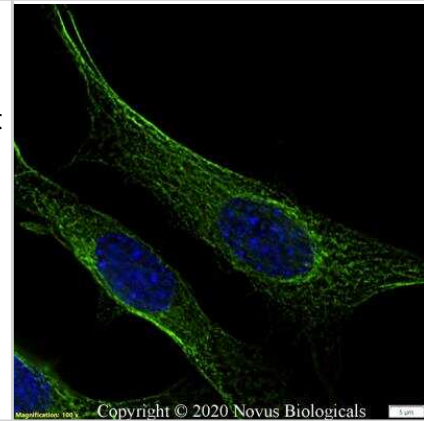
Immunocytochemistry/Immunofluorescence: beta Tubulin Antibody [NB600-936] - Confocal immunofluorescent analysis of C2C12 cells using beta Tubulin antibody (NB600-936, 1:5). An Alexa Fluor 488-conjugated Goat to rabbit IgG was used as secondary antibody (green). Actin filaments were labeled with Alexa Fluor 568 phalloidin (red). DAPI was used to stain the cell nuclei (blue).



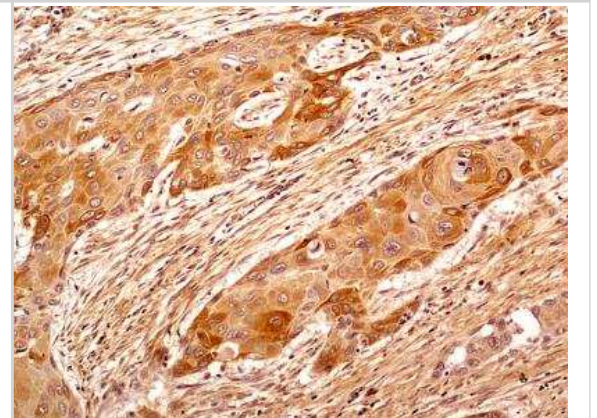
Immunocytochemistry/Immunofluorescence: beta Tubulin Antibody [NB600-936] - Analysis of beta Tubulin in mouse hippocampal primary culture. Image courtesy of product review by Lin Yi-Wen.



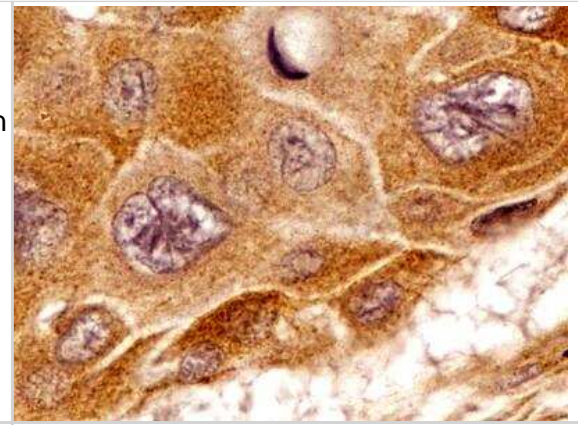
Immunocytochemistry/Immunofluorescence: beta Tubulin Antibody [NB600-936] - NIH3T3 cells were fixed in 4% paraformaldehyde for 10 minutes and permeabilized in 0.05% Triton X-100 in PBS for 5 minutes. The cells were incubated with anti-beta Tubulin Antibody (NB600-936) at 2 ug/ml overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at a 1:1000 dilution for 60 minutes. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 100X objective and digitally deconvolved.



Immunohistochemistry-Paraffin: beta Tubulin Antibody [NB600-936] - Analysis of FFPE tissue section of human esophageal squamous cell carcinoma (SCC) using 1:2000 dilution of beta Tubulin antibody. Strong cytoplasmic immuno-positivity of beta Tubulin (TUBB) was observed in SCC cells as well as the associated tumor stromal cells [10X Magnification].



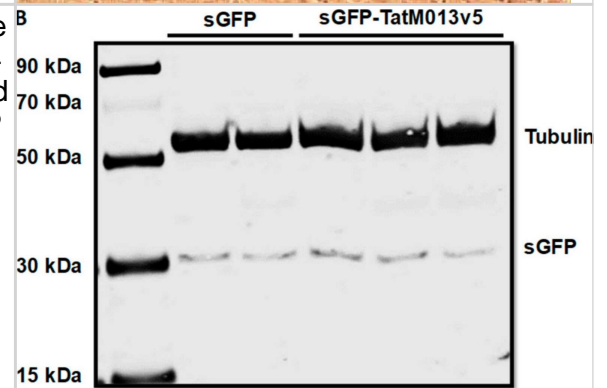
Immunohistochemistry-Paraffin: beta Tubulin Antibody [NB600-936] - Analysis of FFPE tissue section of human esophageal squamous cell carcinoma (SCC) using 1:2000 dilution of beta Tubulin antibody. This representative image shows a cytoplasm specific staining of beta Tubulin (TUBB) in SCC cells [60X Magnification].



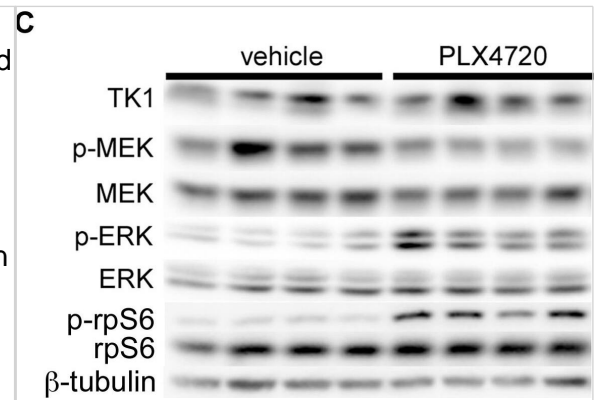
Immunohistochemistry-Paraffin: beta Tubulin Antibody [NB600-936] - Analysis of FFPE tissue section of normal human brain using 1:2000 dilution of beta Tubulin antibody. The various brain cells depicted strong cytoplasmic immunoreactivity of beta Tubulin (TUBB) protein [10X Magnification].



Expression of AAV transgene before and after experimental autoimmune uveoretinitis (EAU) induction. (B) Western blot from retinas harvested 14 days after IRBP immunization. Membrane was probed with anti-GFP and anti-Tubulin antibodies. Image shows expression of sGFP on both sGFP and sGFP-TatM013v5 retina lysates. (n = 2-3 retina samples from different mice per group). Image collected and cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/31795515>), licensed under a CC-BY licence.



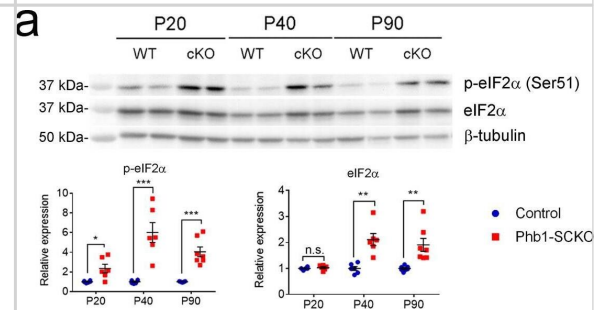
EBNA1 expression is associated with upregulation of MTH1 and oxidative damage repair pathways. The expression of MTH1, OGG1, and MUTYH was investigated by western blots and qPCR in BJAB-tTAE1 cells upon addition and withdrawal of doxycycline. a Representative western blots illustrating the correlation between the reversible down- and upregulation of MTH1 and EBNA1 in BJAB-tTAE1 cells upon addition and withdrawal of doxycycline. GAPDH was used as loading control. b Densitometry quantification of MTH1 and EBNA1 expression in BJAB-tTAE1 cells. The mean intensity of the MTH1 and EBNA1 specific bands relative to GAPDH in three independent experiments is shown in the figure. c Regression analysis of the relationship between expression levels of MTH1 and EBNA1. The data from three independent experiments were used for the plot. d Representative western blots illustrating the expression of MTH1, MUTYH, and OGG1 in BJAB-tTAE1 cells cultured for two weeks in the presence or absence of doxycycline. e Fold change is the ratio between the intensity of the specific band in cells cultured without or with doxycycline. The mean \pm SD of four independent experiments is shown. f qPCR analysis of the levels of MTH1, OGG1, and MUTYH transcripts in BJAB-tTAE1 cells cultured for 2 weeks in the presence or absence of doxycycline. The mean \pm SE of the fold change in six independent experiments each performed in triplicate is shown



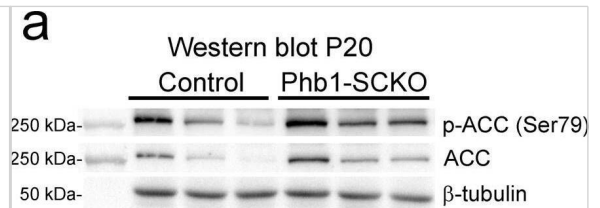
Characterization of immunosorted NCAM1+ subpopulation. A. RT-PCR analysis of gene expression in NCAM1 cell fractions. GAPDH was used as endogenous control and NCAM1- cell were used as the calibrator sample for RQ calculation (therefore = 1). Data were analysed using SDS 3.2 software and presented as average RQ \pm SDEV of three replicates. *** $p < 0.001$, * $p < 0.05$ versus NCAM1-.

B-D. Immunofluorescence staining of NCAM1+ and NCAM1- subpopulations for E-cadherin (E-cad) (B, red) vimentin (C, green) and SIX2 (D, green). Nuclei stained with Dapi (blue). (B-C) Images were obtained using Olympus DP72 camera attached to Olympus BX51 fluorescence microscope and processed via cellSens standard software, bar represents 200 μ m. (D) Images were obtained using Zeiss LSM 510 confocal microscope, bar represents 50 μ m. E. Fluorescent quantification of SIX2 immunostaining as represented in (D). F. Double labelling of sorted NCAM1+ cells for NCAM1 and SIX2: NCAM1 with DAPI (upper panel; red and blue channels), NCAM1 with SIX2 (lower panel; red and green channels), indicating both NCAM1+ SIX2+ (arrows) and NCAM1+ SIX2- (arrowheads) cells. G. Graph represents percentage of NCAM1+ SIX2+ cells and NCAM1+ SIX2- cells. H. Clonogenic efficiency of NCAM1+ cells sorted from hFK and cultured in SFM. Data are presented as average CE(%) \pm SDEV. ** $p < 0.01$ versus NCAM1-.

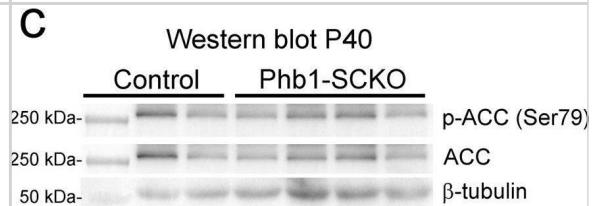
I. Representative morphology of NCAM1+ and NCAM1- clones. Cells were observed using a Nikon Digital Sight camera attached to a Nikon Eclipse TS100 microscope. J. Clonogenic capacity of hFK cells treated with IMG901 (ADC 55 nM), huN901 (Ab55 nM) or not treated (control). Data are presented as average CE(%) \pm SDEV. *** $p < 0.001$, * $p < 0.05$ versus control group. Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/23996934>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



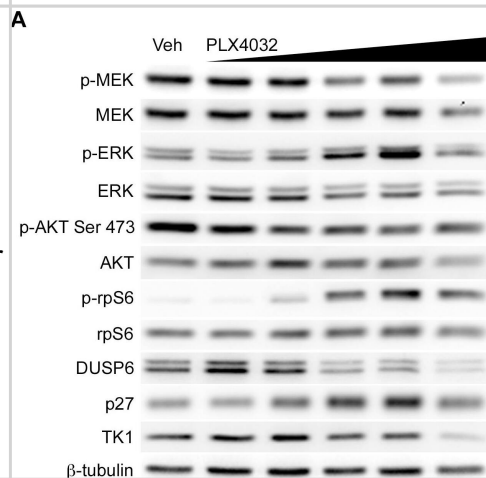
PGE1 induces HIF-1 α protein accumulation in vascular-derived cells. Human aortic smooth muscle cells (HASMCs) (A and B), human umbilical vein endothelial cells (HUVECs) (C and D), and HEK293 cells (G) were exposed to 1 or 10 μ M PGE1 under 20% O₂ or 1% O₂ conditions for 4 h. After treatment, cells were harvested and whole-cell lysates were subjected to immunoblot assay for HIF-1 α , HIF-1 β , and β -actin protein expression. Experiments were repeated thrice (A, C and G). Representative immunoblots are shown (A and C). Band intensities were analyzed densitometrically (B and D). Fold induction relative to lane 1 was plotted as mean \pm S.D. \square P < 0.05 compared with the control. HASMCs (E) and HUVECs (F) were exposed to 1 μ M PGE1 for the indicated times under 20% O₂ and were harvested for immunoblot assay for HIF-1 α protein. Experiments were repeated twice. Representative immunoblots are shown. (H) HASMCs and HUVECs were exposed to 10 μ M PGE1 for 4 h under 20% O₂ and were harvested for immunoblot assay for HIF-2 α protein. Experiments were repeated twice. Representative immunoblots are shown. I. HASMCs were exposed to 1 μ M PGE1, lipo-PGE1 and PGE1-alfadex under 20% O₂ conditions for 4 h. After treatment, cells were harvested and whole-cell lysates were subjected to immunoblot assay for HIF-1 α . Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/24349900>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



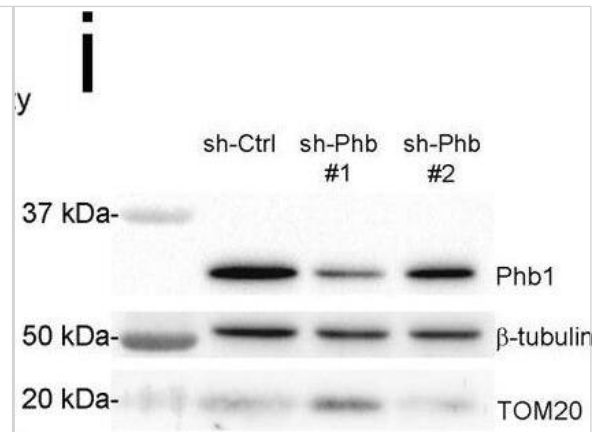
TIP30 was regulated by HIF-2 α at protein level in HCC cell lines. Lentivirus infection was used to establish a stable knocking-down or overexpression of HIF-2 α in MHCC97H cell lines. The relationship between HIF-2 α and TIP30 was detected by Western blot. a Knocking-down of HIF-2 α upregulated TIP30 expression. b Knocking-down of TIP30 did not influence the expression of HIF-2 α . c, d Metformin in combination with sorafenib synergistically inhibited HIF-2 α protein expression and subsequently upregulated TIP30 protein expression. e MHCC97H cells were exposed to CoCl₂ (400 μ M) for 6 h, and anti-HIF-2 α or anti-IgG was used for immunoprecipitation. Immunoprecipitated and purified DNA together with 1 % of input DNA were used for PCR amplification of a 214-bp product encompassing HRE region of TIP30 promoter. Image collected and cropped by CiteAb from the following open publication (<https://www.jhonline.org/content/9/1/20>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



ATM and NF- κ B activation are downregulated in Ercc1- Δ mice heterozygous for Atm. (A) Livers were collected at 12 weeks of age from WT, Ercc1- Δ and Ercc1- Δ Atm^{+/-} mice (n=3 per genotype) and lysates analyzed by western blot for activation of ATM and its downstream effectors. (B) Same liver lysates were used to measure phosphorylation of p65 and I κ B α . (C) Western blot analysis of livers from 16-week-old WT, Ercc1- Δ and Ercc1- Δ Atm^{+/-} mice (n=3 per genotype) probed for activation of ATM. GAPDH was used as a loading control. (D) Same liver lysates used to measure activation of NF- κ B. (E) Fourteen-week-old livers from Ercc1- Δ and Ercc1- Δ p65^{+/-} mice (n=3 per genotype) were analyzed by western blot for activation of ATM (F) and NF- κ B. Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/32201398>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Expression of the mitochondrial biogenesis proteins: SIRT1, PGC1 α , TFAM and SIRT3 in H1299, H1299r, P31 and P31r cell lysates as determined by immunoblot analysis. Whole cell lysates were prepared from confluent cultures of H1299, H1299r, P31 and P31r cells. Proteins (90 μ g) were resolved in 10% SDS-PAGE gels and transferred to a PVDF membrane. Blots were probed for SIRT1, PGC1 α , TFAM and SIRT3 or the loading control γ -tubulin. Figure shows representative blots of (A) of SIRT1, (B) PGC1 α , (C) SIRT3, (D) TFAM protein expression; each from three independent experiments. Image collected and cropped by CiteAb from the following open publication (<https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.21885>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Pre-treatment with 0.5 mM sodium arsenite (SA) enhances permissivity in a cell-type-specific manner across reovirus strains. (A) CV-1, HeLa, L929, or HPDE cells were left untreated (no SA) or were treated with 0.5 mM SA for 30 min prior to infection (Pre-SA). Following this, cells were infected with T3D such that ~20% to 50% of cells were infected and at 18 h p.i. cells were fixed and immunostained for μ NS and DAPI to visualize viral factories (VFs). The percent of cells containing VFs was quantified ($(\# \text{ of cells containing VFs} / \text{total } \# \text{ of cells}) \times 100$) from three independent experiments. The expression level of μ NS (B) and μ 1 (C) was determined in CV-1, L929, or HeLa cells either left untreated (no SA) or treated with 0.5 mM SA for 30 min (Pre-SA) before infection with T3D at MOI = 1. At 18 h p.i., cells were harvested and the expression level of the indicated proteins was determined by immunoblot. M = mock. Densitometry analysis of the band intensity for μ NS and μ 1 was adjusted to the matched α -tubulin loading control for two independent experiments. Columns represent mean \pm SEM. (D) CV-1; (E) L929; or (F) HeLa cells were left untreated (no SA) or were treated with 0.5 mM SA prior to infection (Pre-SA). Cells were then infected with the reovirus strains, T3D, T1L, or T3A, as described in (A). At 18 h p.i., cells were fixed and immunostained for μ NS and DAPI to detect VFs. The percent of cells containing VFs was quantified ($(\# \text{ of cells containing VFs} / \text{total } \# \text{ of cells}) \times 100$) from at least two independent experiments. * $p < 0.05$; ** $p < 0.01$; two-tailed unpaired t test. The error bars indicate S.D. Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/31216693>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Neggens JE, Vanstreels E, Baloglu E et al. Heterozygous mutation of cysteine528 in XPO1 is sufficient for resistance to selective inhibitors of nuclear export Oncotarget 2016-10-18 [PMID: 27634897]

Kang L, Li C, Yang Q et al. (64)Cu-labeled daratumumab F(ab')(2) fragment enables early visualization of CD38-positive lymphoma European Journal of Nuclear Medicine and Molecular Imaging 2022-04-01 [PMID: 34677626] (WB)

Otte K, Zhao K, Braun M et al. Eltanexor Effectively Reduces Viability of Glioblastoma and Glioblastoma Stem-Like Cells at Nano-Molar Concentrations and Sensitizes to Radiotherapy and Temozolomide Biomedicines 2022-08-31 [PMID: 36140245] (ICC/IF, B/N)

Del Rosario JS, Gabrielle M, Yudin Y, Rohacs T. TMEM120A/TACAN inhibits mechanically activated PIEZO2 channels Journal of General Physiology 2022-08-01 [PMID: 35819364] (WB)

Neggens JE, Kwanten B, Dierckx T et al. Target identification of small molecules using large-scale CRISPR-Cas mutagenesis scanning of essential genes Nature Communications 2018-02-05 [PMID: 29402884]

Kashio M, Masubuchi S, Tominaga M. Protein kinase C-mediated phosphorylation of transient receptor potential melastatin type 2 Thr738 counteracts the effect of cytosolic Ca(2+) and elevates the temperature threshold The Journal of Physiology 2022-10-01 [PMID: 36042566]

Affandi T, Haas A, Ohm A et al. PKC ϵ regulates chromatin remodeling and DNA repair through SIRT6 bioRxiv 2023-05-25 [PMID: 37292592] (WB)

Shah H, Olivetta M, Bhickta C et al. Life cycle-coupled evolution of mitosis in close relatives of animals bioRxiv 2023-05-10 (ICC/IF)

Li F, Liu Y, Li L et al. Brain-derived extracellular vesicles mediate traumatic brain injury associated multi-organ damage Biochemical and Biophysical Research Communications 2023-05-01 [PMID: 37163934] (ICC/IF, Mouse)

Ridley RB, Bowman BM, Lee J et al. Modulation of Retinal Inflammation Delays Degeneration in a Mouse Model of Geographic Atrophy bioRxiv : the preprint server for biology 2023-02-09 [PMID: 36798403] (WB, Mouse)

Mouchbahani-Constance S, Lagard C, Schweizer J et al. Modulating the activity of human nociceptors with a SCN10A-promoter specific viral vector tool Neurobiology of Pain 2023-01-01 [PMID: 36816616]

D'Otolo V Functional complementarity of mammalian coronins determines T cell population homeostasis Thesis 2022-01-01 (WB, Human)

More publications at <http://www.novusbio.com/NB600-936>



Procedures

Western Blot protocol for beta Tubulin Antibody (NB600-936)

Western Blot Protocol

1. Perform SDS-PAGE on samples to be analyzed, loading 10-25 ug of total protein per lane.
2. Transfer proteins to PVDF membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.
3. Stain the membrane with Ponceau S (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.
4. Rinse the blot TBS -0.05% Tween 20 (TBST).
5. Block the membrane in 5% Non-fat milk in TBST (blocking buffer) for at least 1 hour.
6. Wash the membrane in TBST three times for 10 minutes each.
7. Dilute primary antibody in blocking buffer and incubate overnight at 4C with gentle rocking.
8. Wash the membrane in TBST three times for 10 minutes each.
9. Incubate the membrane in diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturer's instructions) for 1 hour at room temperature.
10. Wash the blot in TBST three times for 10 minutes each (this step can be repeated as required to reduce background).
11. Apply the detection reagent of choice in accordance with the manufacturers instructions.

Immunocytochemistry/ Immunofluorescence Protocol for beta Tubulin Antibody (NB600-936)

Immunocytochemistry Protocol

Culture cells to appropriate density in 35 mm culture dishes or 6-well plates.

1. Remove culture medium and wash the cells briefly in PBS. Add 10% formalin to the dish and fix at room temperature for 10 minutes.
2. Remove the formalin and wash the cells in PBS.
3. Permeablize the cells with 0.1% Triton X100 or other suitable detergent for 10 min.
4. Remove the permeablization buffer and wash three times for 10 minutes each in PBS. Be sure to not let the specimen dry out.
5. To block nonspecific antibody binding, incubate in 10% normal goat serum from 1 hour to overnight at room temperature.
6. Add primary antibody at appropriate dilution and incubate overnight at 4C.
7. Remove primary antibody and replace with PBS. Wash three times for 10 minutes each.
8. Add secondary antibody at appropriate dilution. Incubate for 1 hour at room temperature.
9. Remove secondary antibody and replace with PBS. Wash three times for 10 minutes each.
10. Counter stain DNA with DAPI if required.



Immunohistochemistry-Paraffin Protocol for beta Tubulin Antibody (NB600-936)

Immunohistochemistry-Paraffin Embedded Sections

Antigen Unmasking:

Bring slides to a boil in 10 mM sodium citrate buffer (pH 6.0) then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench-top for 30 minutes (keep slides in the sodium citrate buffer all the time).

Staining:

1. Wash sections in deionized water three times for 5 minutes each.
2. Wash sections in PBS for 5 minutes.
3. Block each section with 100-400 ul blocking solution (1% BSA in PBS) for 1 hour at room temperature.
4. Remove blocking solution and add 100-400 ul diluted primary antibody. Incubate overnight at 4 C.
5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
6. Add 100-400 ul HRP polymer conjugated secondary antibody. Incubate 30 minutes at room temperature.
7. Wash sections three times in wash buffer for 5 minutes each.
8. Add 100-400 ul DAB substrate to each section and monitor staining closely.
9. As soon as the sections develop, immerse slides in deionized water.
10. Counterstain sections in hematoxylin.
11. Wash sections in deionized water two times for 5 minutes each.
12. Dehydrate sections.
13. Mount coverslips.





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