

Product Datasheet

PD-L1 Antibody - BSA Free NBP1-76769

Unit Size: 0.1 mg

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

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NBP1-76769

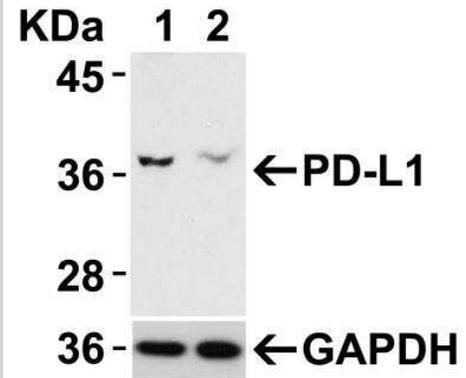
PD-L1 Antibody - BSA Free

| Product Information | |
|------------------------------------|--|
| Unit Size | 0.1 mg |
| Concentration | 1 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 | Peptide affinity purified |
| Buffer | PBS |
| Target Molecular Weight | 37 kDa |
| Product Description | |
| Host | Rabbit |
| Gene ID | 29126 |
| Gene Symbol | CD274 |
| Species | Human, Mouse, Rat |
| Specificity/Sensitivity | PD-L1 antibody has no cross-reactivity to PD-L2. |
| Immunogen | Antibody was raised against a 17 amino acid synthetic peptide from near the center of human PD-L1. The immunogen is located within amino acids 60-110 of PD-L1. |
| Product Application Details | |
| Applications | Western Blot, ELISA, Flow Cytometry, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation, Dual RNAscope ISH-IHC, Immunofluorescence, Immunohistochemistry Whole-Mount, Knockdown Validated |
| Recommended Dilutions | Western Blot 1:1000, Flow Cytometry 0.5 ug/ml, ELISA 1:100 - 1:2000, Immunohistochemistry 1:10-1:500, Immunocytochemistry/ Immunofluorescence 1-5 ug/ml, Immunoprecipitation, Immunohistochemistry-Paraffin 1:10-1:500, Immunohistochemistry-Frozen, Immunohistochemistry Whole-Mount, Immunofluorescence 20 ug/ml, Knockdown Validated, Dual RNAscope ISH-IHC |
| Application Notes | Use in Immunohistochemistry Whole-Mount reported in scientific literature (PMID:34944780). Use in ICC/IF reported in scientific literature (PMID:33220359) Use in IHC-Frozen was reported in scientific literature (PMID: 28402953). Use in immunoprecipitation reported in scientific literature (PMID: 28978117).. |

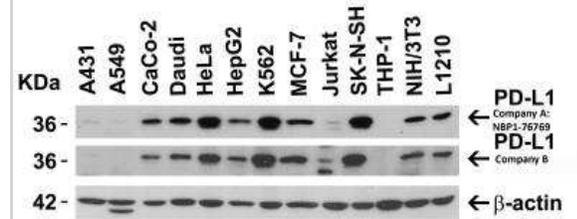


Images

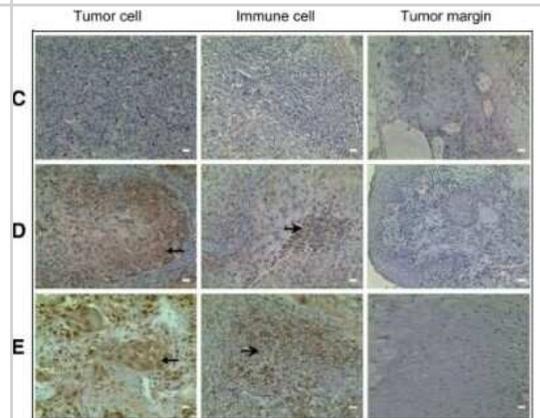
Knockdown Validated: PD-L1 Antibody [NBP1-76769] - Validation with PD-L1 siRNA Knockdown. HeLa cells were transfected with control siRNAs (lane 1) or PD-L1 siRNAs (lane 2). Loading: 10 ug of HeLa whole cell lysates per lane. Antibodies: NBP1-76769 (2 ug/mL) and GAPDH (0.02 ug/mL), 1 h incubation at RT in 5% NFDN/TBST. Secondary: Goat anti-rabbit IgG HRP conjugate at 1:10000 dilution.



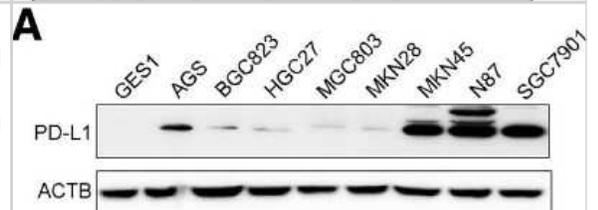
Western Blot: PD-L1 Antibody [NBP1-76769] - Independent Antibody Validation (IAV) via Protein Expression Profile Loading: 15 ug of lysates per lane. Antibodies: NBP1-76769 (2 ug/mL), PD-L1 (2 ug/mL), and beta-actin (1 ug/mL), 1 h incubation at RT in 5% NFDN/TBST. Secondary: Goat anti-rabbit and or anti-mouse IgG HRP conjugate at 1:10000 and 1:5000 dilution, respectively.



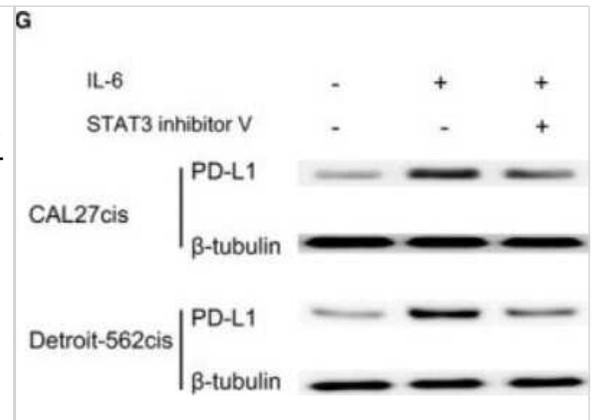
Immunohistochemistry-Paraffin: PD-L1 Antibody [NBP1-76769] - CD274 (PD-L1) expression in the HNSCC patients from the TCGA database, HNSCC tissue samples and the HNSCC cells. Immunostaining of PD-L1 obtained from HNSCC tumor cells, immune cells and tumor margin tissues in HNSCC tissue samples (magnification A-200, scale bars 50A11/4m): low tumor staining; moderate tumor staining; high tumor staining have been observed. Image collected and cropped by Citeab from the following publication (Lactoferricin B reverses cisplatin resistance in head and neck squamous cell carcinoma cells through targeting PD-L1. *Cancer Med* (2018)) licensed under a CC-BY license.



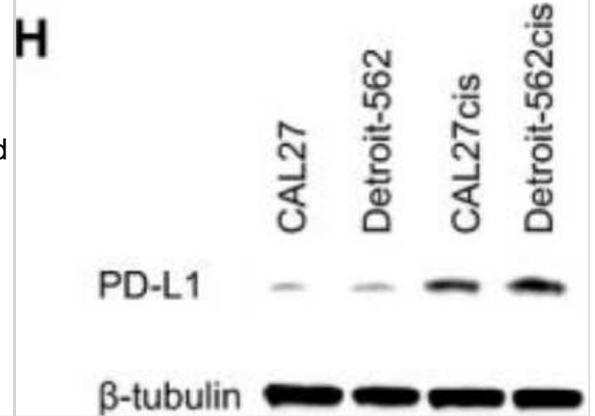
Western Blot: PD-L1 Antibody [NBP1-76769] - PD-L1 expression was evaluated in gastric cancer cells. Levels of PD-L1 protein were assessed in normal human gastric epithelial cells and 8 gastric cancer cell lines by Western blots. Image collected and cropped by Citeab from the following publication (Autophagy inhibition enhances PD-L1 expression in gastric cancer. *J Exp Clin Cancer Res* (2019)) licensed under a CC-BY license.



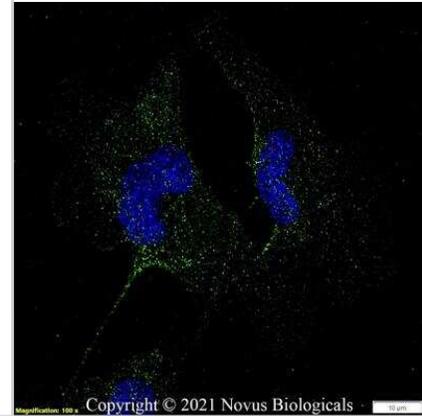
Western Blot: PD-L1 Antibody [NBP1-76769] - Upregulated expression of PD-L1 in cisplatin-resistant cells through IL-6/STAT3. Image collected and cropped by Citeab from the following publication (Lactoferricin B reverses cisplatin resistance in head and neck squamous cell carcinoma cells through targeting PD-L1. *Cancer Med* (2018)) licensed under a CC-BY license.



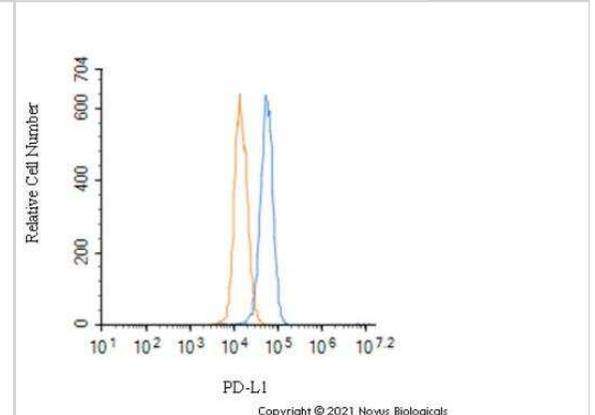
Western Blot: PD-L1 Antibody [NBP1-76769] - CD274 (PD-L1) expression in the HNSCC patients from the TCGA databas HNSCC cells. CD274 expression with survival and its relation with therapy in the HNSCC patients from the TCGA database. Expression of CD274 gene and PD-L1 protein in the established cisplatin-resistant HNSCC cells and cisplatin sensitive cells by qRT-PCR and WB (F, H): CD274 (PD-L1) expressed in cisplatin resistant cell. Image collected and cropped by Citeab from the following publication (Lactoferricin B reverses cisplatin resistance in head and neck squamous cell carcinoma cells through targeting PD-L1. *Cancer Med* (2018)) licensed under a CC-BY license.



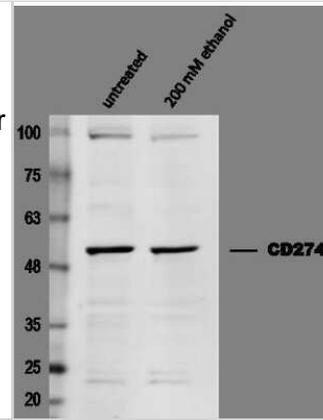
Immunocytochemistry/Immunofluorescence: PD-L1 Antibody [NBP1-76769] - U-251 MG cells were fixed in 4% paraformaldehyde for 10 minutes and permeabilized in 0.05% Triton X-100 in PBS for 5 minutes. The cells were incubated with anti- NBP1-76769 at 1 ug/ml overnight at 4C and detected with an anti-rabbit 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.



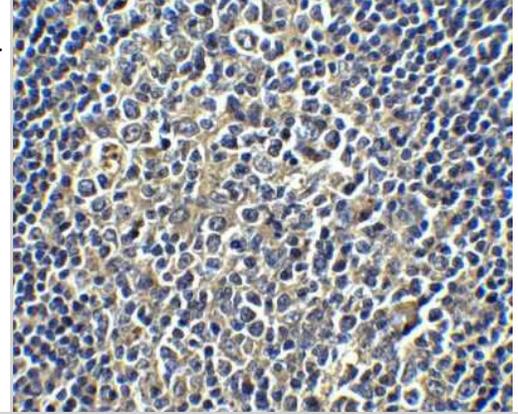
Flow Cytometry: PD-L1 Antibody [NBP1-76769] - An intracellular stain was performed on U-251 MG cells with PD-L1 Antibody NBP1-76769 (blue) and a matched isotype control NBP2-24891 (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 2.5 ug/mL for 30 minutes at room temperature, followed by Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody, Dylight 550 (SA5-10033, Thermo Fisher).



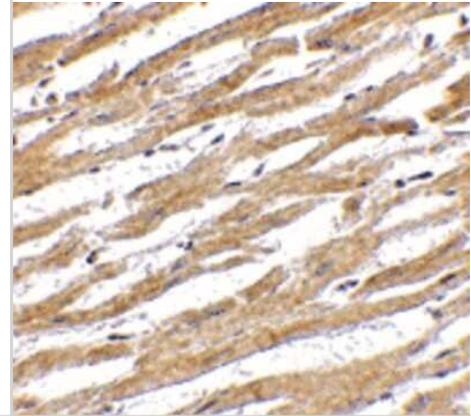
Western Blot: PD-L1 Antibody [NBP1-76769] - Expression of CD274 in Human melanoma SK-MEL-28 cell line upon treatment with 200 mM ethanol. Dilution: 1:1,000 in PBS with 5% BSA. Secondary Ab: anti-Rabbit IgG 1:5,000. Western blot image submitted by a verified customer review.



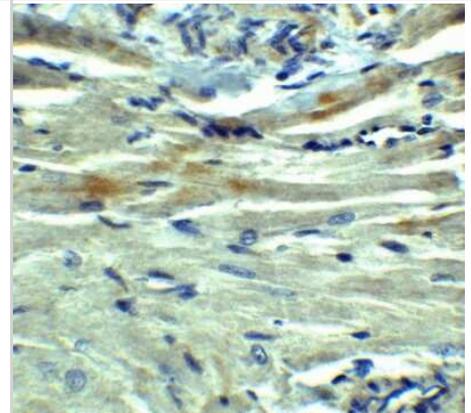
Immunohistochemistry-Paraffin: PD-L1 Antibody [NBP1-76769] - B7-H1/PD-L1/CD274 Antibody [NBP1-76769] - Human tonsil tissue with PD-L1 antibody at 5 ug/mL.



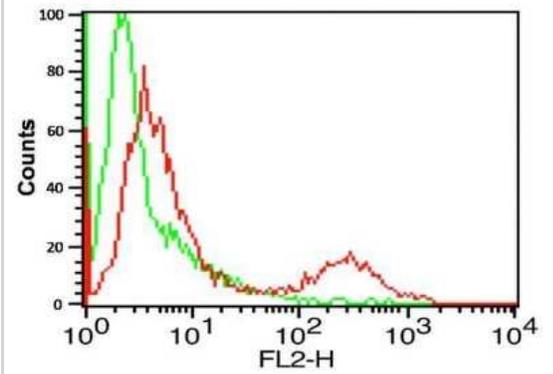
Immunohistochemistry-Paraffin: PD-L1 Antibody [NBP1-76769] - Staining of human heart tissue with antibody at 2.5 ug/mL.



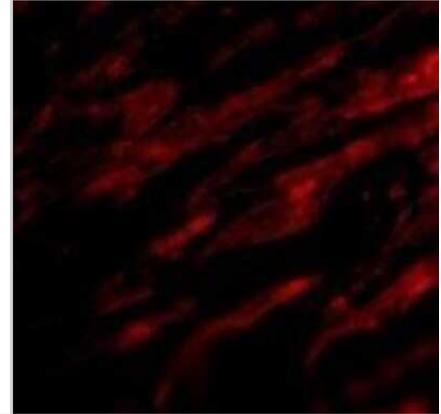
Immunohistochemistry-Paraffin: PD-L1 Antibody [NBP1-76769] - PD-L1/B7-H1 Antibody [NBP1-76769] - PD-L1 in rat heart tissue with at 5 ug/mL.



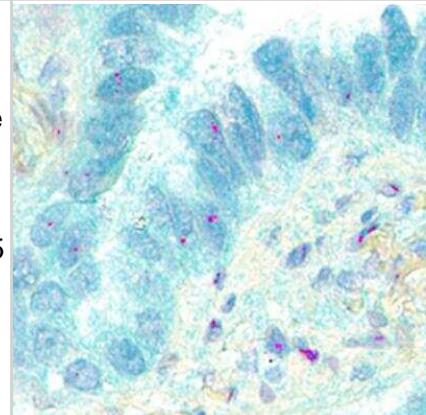
Flow Cytometry: PD-L1 Antibody [NBP1-76769] - B7-H1/PD-L1/CD274 Antibody [NBP1-76769] - Analysis of A-20 cells using B7-H1/PD-L1/CD274 antibody at 0.5 ug/mL. Green: Isotype control. Red : B7-H1/PD-L1/CD274 antibody.



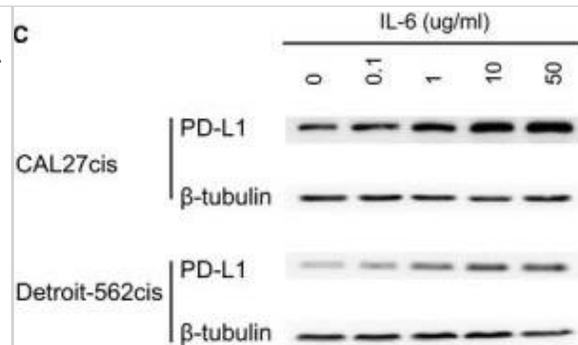
Immunofluorescence: PD-L1 Antibody [NBP1-76769] - Human Heart cells with PD-L1 antibody at 20 ug/mL.



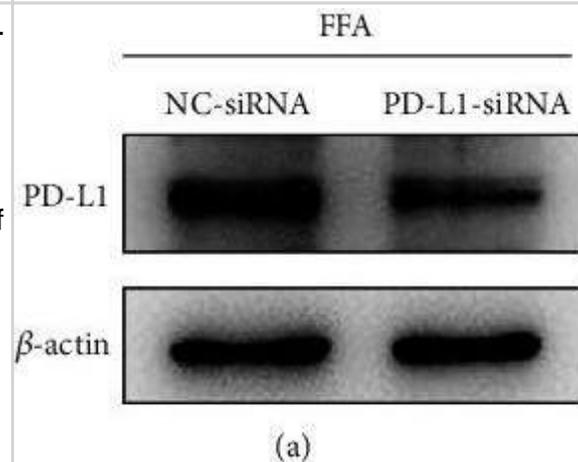
Dual RNAscope ISH-IHC: PD-L1 Antibody [NBP1-76769] - PDCD1 mRNA (red) and PD-L1 protein (green) were detected in formalin-fixed paraffin-embedded tissue sections of human lung cancer. ACD's Integrated Co-Detection Workflow was performed using ACD RNAscope Probe Hs-PDCD1 and PD-L1 antibody at 1:100 dilution. Tissue was stained on Leica Bond RX using RNAscope (TM) 2.5 LS Reagent Kit-RED, BOND Polymer Refine Detection (DAB) and Hematoxylin, BOND Polymer Refine Red Detection and Hematoxylin and RNAscope (TM) 2.5 LS Green Accessory Pack. Tissue was counterstained with 50% hematoxylin (blue).



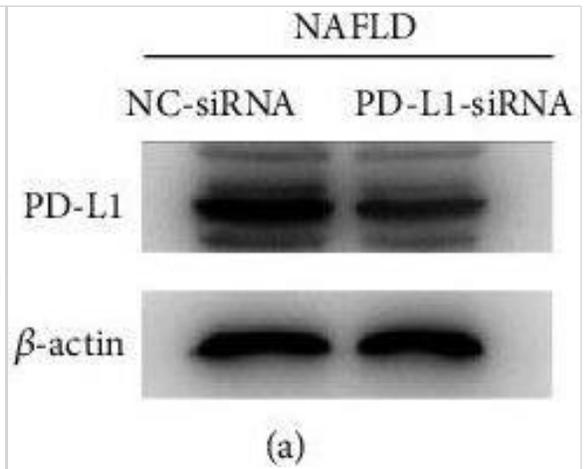
WNT11 is induced by hypoxia or hypoxic mimetics in different cell types. (A) Increased Wnt11 mRNA in EMSC adipocytes (Day 12) after hypoxia-mimetic treatments. EMSC adipocytes were treated with CoCl₂ (0.1 mM), DFO (0.1 mM) or DMOG (0.1 mM) for 24 hrs. Values were normalized to Tbp mRNA and are expressed relative to control (n = 3). (B,C) Increased Wnt11 mRNA by hypoxia in EMSC preadipocytes and adipocytes (Day 0–12 after differentiation) (B), and C2C12 myoblast and myocyte (Day 0 and 8 after differentiation) (C). Wnt11 mRNA was assessed by quantitative PCR in cells exposed to air (21% O₂) or hypoxia (1% O₂) for 24 hrs. (n = 4). Values were normalized to Tbp mRNA and are expressed relative to 21% O₂ samples (left panel). (D) Immunoblot analyses of HeLa cells under normal air or hypoxia for 24 hrs. (E,F) Induction of Wnt11 by increasing concentrations of DMOG in MDA-MB-231 cells (E) and 4T1 cells (F). (G) EMSCs treated with 0.1 mM DMOG for the indicated times. Wnt11 and Vegf mRNA expression was measured by qPCR and normalized to Tbp mRNA (n = 4). (H) WNT11 protein levels after DMOG treatment normalized to α -Tubulin (upper panel; n = 4). Representative immunoblots of EMSCs treated with 0.1 mM DMOG for the indicated times (Lower panel). (I) Protein expression in MDA-MB-231 cells treated with 0.1 mM DMOG. (J) Induction of Wnt11 promoter activity by hypoxia or hypoxia mimetics. pGL3-Wnt11 promoter plasmid was transfected into C2C12 cells. Cells were incubated with DMOG (left panel, n = 4) or under 21% O₂ or 1% O₂ (right panel, n = 8) for 24 hrs. For panels (A–C,G,H,J), values are mean \pm s.e.m. *p < 0.05, **p < 0.01. For panels of immunoblotting, laminin, α -tubulin, and ERK were used as loading controls, WNT11 normalized to α -Tubulin was shown. Image collected and cropped by CiteAb from the following open publication (<https://www.nature.com/articles/srep21520>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



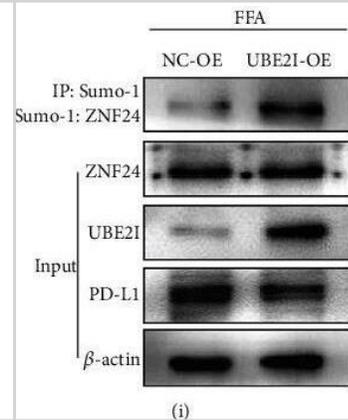
TIM-3 is highly expressed in hypoxic brain regions of a H/I mouse model. (a) TIM-3 transcript levels were examined in brain tissues from the contralateral cortex (C, boxed region) and ischaemic ipsilateral cortex (I, boxed region) of mouse model 24 h after H/I. The RT-PCR products were quantified with Image J and normalized with respect to the expression of actin. The HIF-1 α transcript level represents a positive control for hypoxia. The right panel shows representative TTC staining of three brain sections from the H/I mice. (b) Representative western blot analyses of the TIM-3 and HIF-1 α proteins (n=3). Relative levels of TIM-3 are shown as the mean \pm s.d. from three independent experiments. (c) Contralateral and ipsilateral cortical regions of coronal sections from the H/I mice were subjected to immunohistochemistry using an anti-TIM-3 antibody, and the number of TIM-3-expressing cells per mm² was counted. (d) Immunohistochemistry was performed on brain sections from the H/I mice using anti-TIM-3 and hypoxyprobe-1 (red, to detect hypoxic regions). Scale bars, 50 μ m (\times 20); 50 μ m (\times 40). (e,f) Brain cells were isolated from the ipsilateral and contralateral hemispheres of three mice per group, processed for simultaneous detection of TIM-3 plus Iba-1 (e) or GFAP (f), and analysed by FACS. The results are presented as relative TIM-3 levels in the indicated gated populations, as determined from three independent experiments. Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/25790768>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



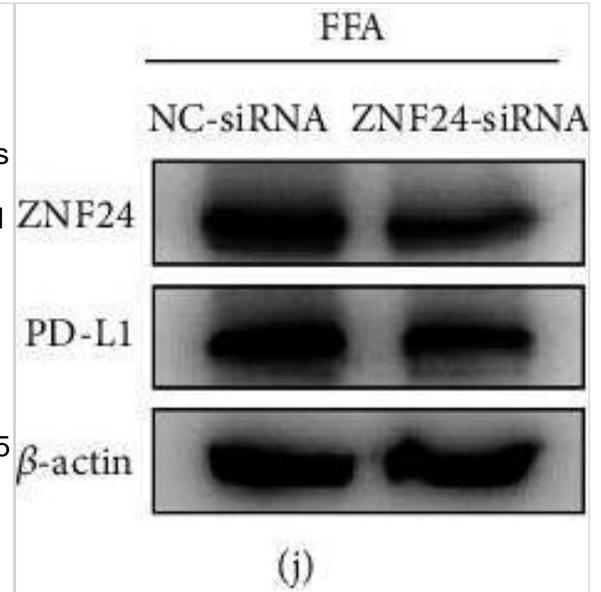
AcAPE1 is exclusively associated with chromatin and remains bound to the condensed chromosomes. (A and B) Asynchronous normal lung fibroblast IMR90 cells and lung adenocarcinoma A549 cells were immunostained with anti-APE1 and anti-AcAPE1 Abs, counterstained with DAPI, and visualized by confocal microscopy and 3D SIM. (C) Colocalization of AcAPE1 with histone H3 or active enhancer-specific histone marker acetylated H3K27 (H3K27Ac). (D) BJ-hTERT cells were serum starved for 72 h and then fixed at different time points. Cells were immunostained with anti-APE1 and anti-AcAPE1 Abs and counterstained with anti-TO-PRO-3 iodide Ab. (E) Mitotic A549 cells were immunostained with anti-APE1 and anti-AcAPE1 and visualized by 3D SIM. (F) BJ-hTERT cells were either serum starved for 72 h (G0/G1 phase), treated with nocodazole (mitotic cells) or aphidicolin (G1/S phase synchronized cells), or untreated, and whole-cell extracts were isolated using 150 mM or 300 mM salt-containing lysis buffer. Western blot analysis for anti-APE1 and anti-AcAPE1 levels was performed. Anti-HSC70 was used as loading control. (G) A proximal ligation assay was performed with mouse anti-APE1 and rabbit anti-APE1 (mAPE1 & Rabbit-APE1), mouse anti-mouse APE1 and rabbit anti-AcAPE1 (mAPE1 & rAcAPE1), and rabbit anti-AcAPE1 and mouse anti-histone H3 (mHistone H3 & rAcAPE1) to confirm the chromatin association of AcAPE1. Mouse IgG (mIgG) and rabbit anti-AcAPE1 were used as a control. At least 50 cells were counted for PLA foci. (H) Colocalization of p300 and AcAPE1 on chromatin (DAPI). (I) HCT116 cells were transfected with E1A and mutant E1A, and at 48 h after transfection, IF was performed. Cells were immunostained with anti-p300 and anti-APE1 or anti-AcAPE1 and counterstained with DAPI. Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/27994014>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Delayed, second window RIC maintains mTOR inhibition without activating autophagosome machinery. (A) Western blots for autophagy related signaling proteins. (B) Quantification of the protein fold change in 2W RIC compared to 2W controls. Values are means \pm S.E.M. n=6–8 per group. An (*) denotes a statistically significant difference ($P < 0.05$) compared to control. (P-: phospho-). Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/25347774>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

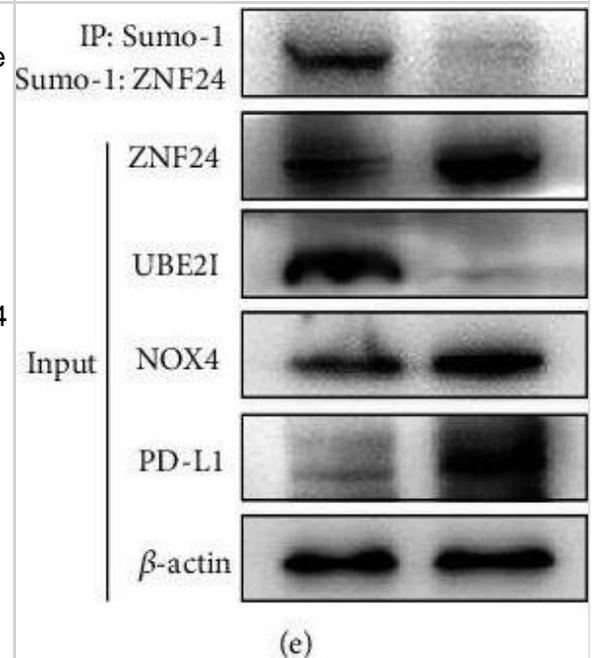


Regulation of FOXC1 on TLR expression. H9c2 cells were transfected with adenovirus or siRNA to overexpress or knock down FoxC1, with GFP adenovirus (Ad-GFP) and negative control (NC) siRNA serving as control, respectively. A, FoxC1 overexpression increased Tlr3/4 mRNA and protein levels, under both control and ischaemic conditions. P values from the one-way ANOVAs: $<.001$ (TLR3 mRNA), $.002$ (TLR4 mRNA), $<.001$ (FOXC1 mRNA), $<.001$ (TLR3 protein), $<.001$ (TLR4 protein) and $<.001$ (FOXC1 protein). B, FoxC1 knockdown decreased Tlr3/4 mRNA and protein levels. P values from the one-way ANOVAs: $.001$ (TLR3 mRNA), $<.001$ (TLR4 mRNA), $<.001$ (FOXC1 mRNA), $.002$ (TLR3 protein), $<.001$ (TLR4 protein) and $<.001$ (FOXC1 protein). C, FoxC1 overexpression up-regulated the mRNA expression of multiple Tlr subtypes. P values from the one-way ANOVAs: $.022$ (TLR1 mRNA), $.047$ (TLR2 mRNA), $.002$ (TLR3 mRNA), $.013$ (TLR4 mRNA), $.039$ (TLR5 mRNA), $.003$ (TLR6 mRNA), $.203$ (TLR7 mRNA), $.078$ (TLR8 mRNA) and $.009$ (TLR9 mRNA). Data are means \pm SEM of 4 independent experiments. aP $<.05$, AP $<.01$ vs. control; bP $<.05$, BP $<.01$ vs. ischaemia Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/31517441>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Macrophages regulate dormancy in tumor cells. a Representative image of triple immunofluorescently stained in E0771-GFP primary tumor tissue for tumor cells, macrophages, and NR2F1. Green = GFP; Red = NR2F1; White = IBA-1; Blue = DAPI. White arrow shows a macrophage. The yellow arrow shows the contact between an NR2F1-positive tumor cell and a macrophage. M ϕ =Macrophage. Scale bar=20 μ m. b Quantification showing the frequency of distances between NR2F1+ tumor cells to the nearest macrophage in the primary tumor. Data is normalized to the frequency of distances between all DAPI+ nuclei to the nearest TMEM. Bar = mean. Error bars = \pm SEM. n = 34 fields of view (551 \times 316 μ m²) in 4 animals. For comparison between the 0 and 200 μ m bins a two-tailed Mann-Whitney test was used ($p < 0.0001$). ****p < 0.0001 . c

Representative immunofluorescence images of NR2F1 expression in E0771-GFP tumor cells cultured alone, in direct contact with BAC1.2F5 macrophages, or in direct contact with HUVEC endothelial cells. White arrows show macrophages or endothelial cells in direct contact with a tumor cell. Green = GFP; Red = NR2F1; Blue = DAPI. TC = Tumor Cell. M ϕ = Macrophage. EC = Endothelial Cell. Scale bar = 15 μ m. d Percentage of NR2F1-positive tumor cells from each group in C. TC alone: n = 777 cells in 9 independent experiments; TC+M ϕ ; n = 226 cells in 6 independent experiments, TC+EC = n = 359 cells in 4 independent experiments. Bar = mean. Error bars = \pm SEM. For TC vs. TC+M ϕ ($p = 0.0039$), and for TC vs. TC+EC ($p = 1$), a two-tailed Kruskal-Wallis test with Dunn's multiple comparisons adjustment was used. For TC+M ϕ vs. TC+EC (0.012), a two-tailed one-way ANOVA with Sidak's multiple comparison adjustment was used. *p < 0.05 . **p < 0.01 ; ns = not significant. Source data are provided as a Source Data file. Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/35110548>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Jin Y, Zuo HX, Li MY et al. Anti-Tumor Effects of Carrimycin and Monomeric Isovalerylspiramycin I on Hepatocellular Carcinoma in Vitro and in Vivo *Frontiers in Pharmacology* 2021-11-26 [PMID: 34899336] (WB)

Zhang J, Qi J, Wei H et al. TGF β 1 in Cancer-Associated Fibroblasts Is Associated With Progression and Radiosensitivity in Small-Cell Lung Cancer *Frontiers in Cell and Developmental Biology* 2021-05-20 [PMID: 34095135] (IHC)

Holokai L, Chakrabarti J, Broda T et al. Increased Programmed Death-Ligand 1 is an Early Epithelial Cell Response to *Helicobacter pylori* Infection *PLoS Pathog.* 2019-01-01 [PMID: 30703170] (B/N)

Dong G, Huang X, Chen R et al. Increased PD-L1 Restricts Liver Injury in Nonalcoholic Fatty Liver Disease *Oxidative Medicine and Cellular Longevity* 2022-05-16 [PMID: 35615575] (WB, IP, IHC)

Zhong Y, Li MY, Han L et al. Galangin inhibits programmed cell death-ligand 1 expression by suppressing STAT3 and MYC and enhances T cell tumor-killing activity *Phytomedicine : international journal of phytotherapy and phytopharmacology* 2023-07-01 [PMID: 37267692] (WB, Human)

Details:

1:2000 WB dilution

Caron JM, Han X, Lary CW et al. Targeting the secreted RGDKGE collagen fragment reduces PD β L1 by a proteasome-dependent mechanism and inhibits tumor growth *Oncology reports* 2023-02-01 [PMID: 36633146] (ICC/IF, Mouse)

Riondato F, Colitti B, Rosati S et al. A method to test antibody cross-reactivity toward animal antigens for flow cytometry *Cytometry. Part A : the journal of the International Society for Analytical Cytology* 2022-09-26 [PMID: 36161760]

Dong M, Qian M, Ruan Z CUL3/SPOP complex prevents immune escape and enhances chemotherapy sensitivity of ovarian cancer cells through degradation of PD-L1 protein *Journal for immunotherapy of cancer* 2022-10-01 [PMID: 36198437] (IP, WB, Human)

Sakuma K, Kii T, Takahashi H et al. An In Vivo Study of Local Administration of Low-dose Anti-PD-1 Antibody Using an Oral Cancer Cell Line *Anticancer research* 2022-09-01 [PMID: 36039414]

Yang W, Han B, Chen Y, Geng F SAAL1, a novel oncogene, is associated with prognosis and immunotherapy in multiple types of cancer *Aging* 2022-08-13 [PMID: 35963646] (WB, Human)

Luo Z, Liao T, Zhang Y et al. Ex vivo anchored PD L1 functionally prevent in vivo renal allograft rejection *Bioengineering & Translational Medicine* 2022-04-06 [PMID: 36176616] (WB, ICC/IF, IHC-P, Rat)

Xia W, Zhang S, Duan H et al. The combination therapy of Everolimus and anti-PD-1 improves the antitumor effect by regulating CD8+ T cells in bladder cancer *Medical oncology (Northwood, London, England)* 2022-01-20 [PMID: 35059863]

More publications at <http://www.novusbio.com/NBP1-76769>





Novus Biologicals USA

10730 E. Briarwood Avenue
Centennial, CO 80112
USA
Phone: 303.730.1950
Toll Free: 1.888.506.6887
Fax: 303.730.1966
nb-customerservice@bio-techne.com

Bio-Techne Canada

21 Canmotor Ave
Toronto, ON M8Z 4E6
Canada
Phone: 905.827.6400
Toll Free: 855.668.8722
Fax: 905.827.6402
canada.inquires@bio-techne.com

Bio-Techne Ltd

19 Barton Lane
Abingdon Science Park
Abingdon, OX14 3NB, United Kingdom
Phone: (44) (0) 1235 529449
Free Phone: 0800 37 34 15
Fax: (44) (0) 1235 533420
info.EMEA@bio-techne.com

General Contact Information

www.novusbio.com
Technical Support: nb-technical@bio-techne.com
Orders: nb-customerservice@bio-techne.com
General: novus@novusbio.com

Products Related to NBP1-76769

| | |
|---------------------|---|
| NBP1-76769PEP-0.1mg | PD-L1 Antibody Blocking Peptide |
| HAF008 | Goat anti-Rabbit IgG Secondary Antibody [HRP] |
| NB7160 | Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP] |
| NBP2-24891 | Rabbit IgG Isotype Control |

Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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