Product Datasheet

N-Cadherin Antibody (13A9)
NBP1-48309

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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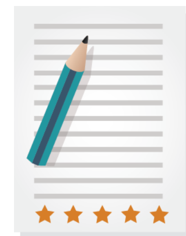


NBP1-48309

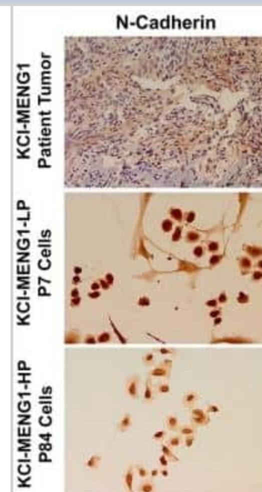
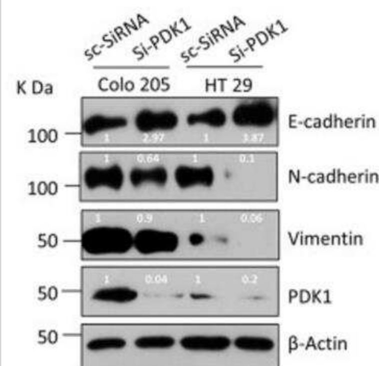
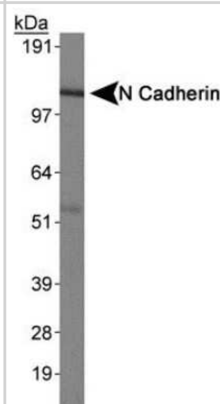
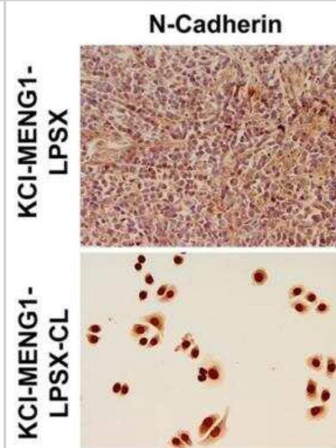
N-Cadherin Antibody (13A9)

Product Information	
Unit Size	0.1 ml
Concentration	1 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	13A9
Preservative	0.05% Sodium Azide
Isotype	IgG1
Purity	Protein G purified
Buffer	PBS
Target Molecular Weight	140 kDa

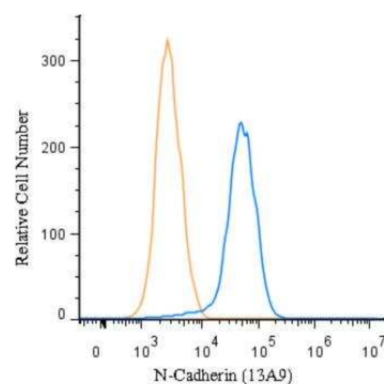
Product Description	
Host	Mouse
Gene ID	1000
Gene Symbol	CDH2
Species	Human, Mouse, Rat
Marker	Mesenchymal Cells Marker
Immunogen	This N-Cadherin Antibody (13A9) was developed against the cytoplasmic domain of human N Cadherin [Swiss-Prot# P19022].

Product Application Details	
Applications	Western Blot, Simple Western, Flow Cytometry, Flow (Intracellular), Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin, Immunoprecipitation, Immunocytochemistry
Recommended Dilutions	Western Blot 0.5 ug/ml, Simple Western 1:50, Flow Cytometry, Immunohistochemistry 1:50-1:200, Immunocytochemistry/ Immunofluorescence 1:100, Immunoprecipitation 1:10-1:500, Immunohistochemistry-Paraffin 1:50-1:100, Flow (Intracellular), Immunocytochemistry
Application Notes	In Western Blot a band is observed at approx. 140 kDa. In Simple Western only 10 - 15 uL of the recommended dilution is used per data point. 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

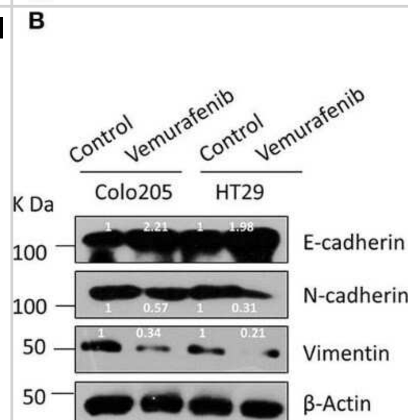
Immunocytochemistry: N-Cadherin Antibody (13A9) [NBP1-48309] - Immunostaining of original tumor, low passage, and high passage KCI-MENG1 cells. The original patient-derived tumor (top row) showed moderate and patchy immunoreactivity for epithelial membrane antigen (EMA); strong and diffuse immunostaining for progesterone receptor (PR); and a Ki-67 proliferative index of 2-3%. There was also strong immunostaining for N-cadherin and vimentin. KCI-MENG1-LP cells (middle row) and KCI-MENG1-HP cells (bottom row) maintained expression of EMA, N-cadherin, and vimentin but had significantly reduced PR expression compared to the original tumor. Whereas Ki-67 labeling was found in only a small number of cells in the original tumor and low passage cells, it was positive in virtually all P84 cells. Scale bar 50 um. Image collected and cropped by CiteAb from the following publication (https://www.translational-medicine.com/content/13/1/227), licensed under a CC-BY license.	
Western Blot: N-Cadherin Antibody (13A9) [NBP1-48309] - Western blots showing a reduction in epithelial mesenchymal transition (EMT) markers upon PDK1 knockdown in Colo205 and HT29 cells using E cadherin (NBP2-19051), N cadherin (NBP1-48309) and B-actin antibody (NB600-501). The corresponding secondary antibodies used were either goat anti-rabbit IgG-HRP (NB7160) or goat anti-mouse IgG-HRP (NB7539). Image collected and cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/33738242) licensed under a CC-BY license.	
Western Blot: N-Cadherin Antibody (13A9) [NBP1-48309] - Analysis of N-Cadherin expression in HeLa whole cell lysate.	
Immunocytochemistry: N-Cadherin Antibody (13A9) [NBP1-48309] - Human meningioma mouse xenograft model KCI-MENG1-LPSX generated with the spontaneously immortal cell line KCI-MENG1-LP. Tumors from immunocompromised SCID mice were dissected and the derivative cell line KCI-MENG1-LPSX CL was generated. The EMA, PR, and N-cadherin IHC of the mouse tumor highly resembled the original patient-derived tumor. The vimentin- and Ki-67-stained cells in the mouse tumor tissue were markedly more abundant and more intensely stained than in the original tumor. KCI-MENG1-LPSX CL cells displayed the same patterns of immunostaining as the high passage parent cell line KCI-MENG1-HP, including the loss of PR staining. Scale bar 50 um. Image collected and cropped by CiteAb from the following publication (https://www.translational-medicine.com/content/13/1/227), licensed under a CC-BY license.	

Flow (Intracellular): N-Cadherin Antibody (13A9) [NBP1-48309] - An intracellular stain was performed on HeLa with NBP1-48309 and a matched isotype control. Cells were fixed with 4% PFA and then permeablized with 0.1% saponin. Cells were incubated in an antibody dilution of 1 ug/mL for 30 minutes at room temperature, followed by Mouse F(ab)2 IgG (H+L) PE-conjugated Antibody.

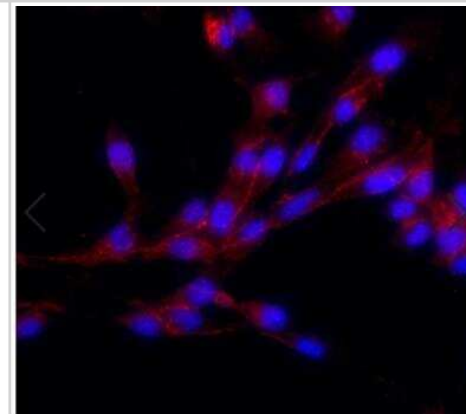


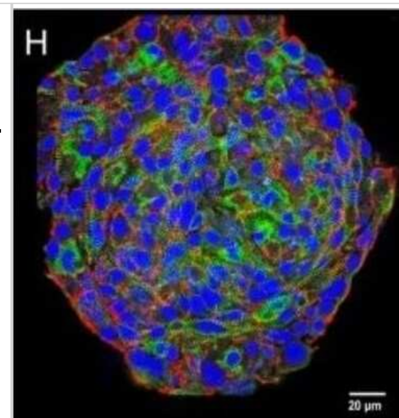
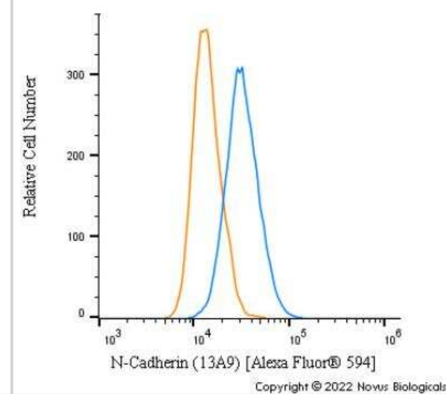
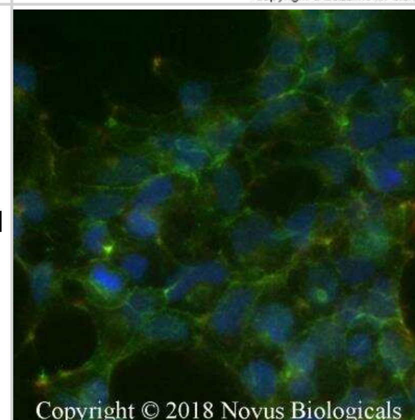
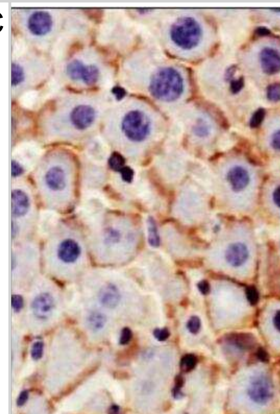
Simple Western: N-Cadherin Antibody (13A9) [NBP1-48309] - Simple Western lane view shows a specific band for N Cadherin in 1.0 mg/mL of HeLa lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.

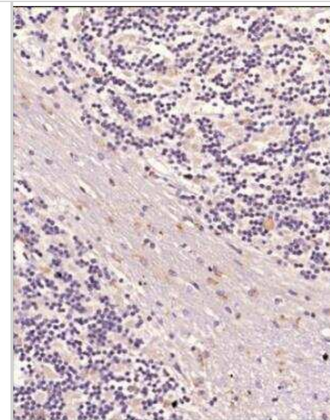
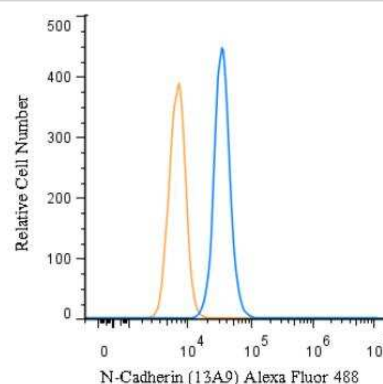
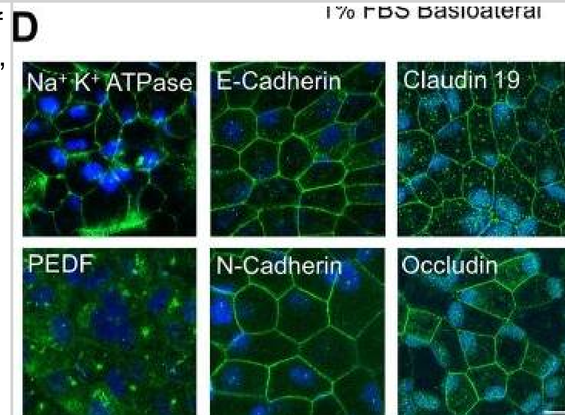
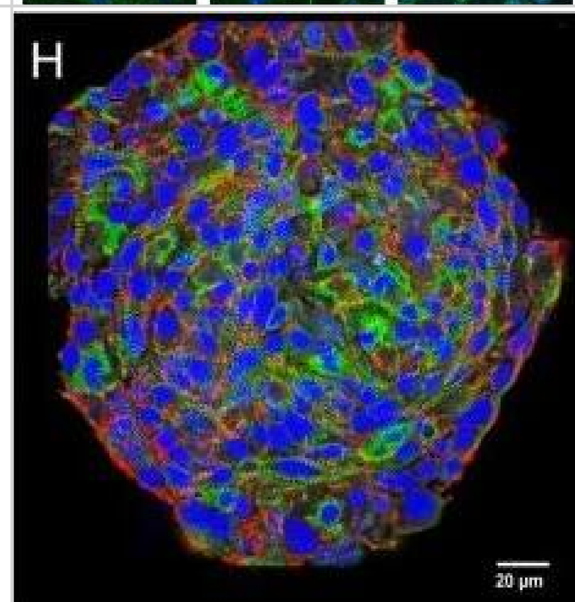
Western Blot: N-Cadherin Antibody (13A9) [NBP1-48309] - Mitochondrial fission regulates migration and invasion in BRAF^V600E CRC cells through glucose metabolic reprograming. Western blot showing a reduction in EMT markers upon vemurafenib treatment in BRAF^V600E cells. Image collected and cropped by CiteAb from the following publication (//pubmed.ncbi.nlm.nih.gov/33738242/) licensed under a CC-BY license.



Immunocytochemistry/Immunofluorescence: N-Cadherin Antibody (13A9) [NBP1-48309] - Adult mouse neural stem cells stained for N-Cadherin. Antibody at 1:100. ICC/IF image submitted by a verified customer review.



Immunohistochemistry: N-Cadherin Antibody (13A9) [NBP1-48309] - Immunohistochemical characterization of tissue cultures. Nuclei labeled with DAPI (blue). Whole-mount immunostaining of a cardiomyocyte-only spheroid 1 month in culture, N-cadherin (red) and EH-myomesin (green). Image collected and cropped by CiteAb from the following publication (//pubmed.ncbi.nlm.nih.gov/32118040/) licensed under a CC-BY license.	
Flow Cytometry: N-Cadherin Antibody (13A9) [NBP1-48309] - An intracellular stain was performed on U-251 MG cells with N-Cadherin Antibody (13A9) NBP1-48309AF594 (blue) and a matched isotype control (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. Both antibodies were conjugated to Alexa Fluor 594.	
Immunocytochemistry/Immunofluorescence: N-Cadherin Antibody (13A9) [NBP1-48309] - HEK 293 cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X PBS + 0.5% Triton X-100. The cells were incubated with anti- at 5 ug/mL overnight at 4C and detected with an anti-mouse DyLight 488 (Green) at a 1:500 dilution. Actin was detected with Phalloidin 568 (Red) at a 1:200 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective.	
Immunohistochemistry: N-Cadherin Antibody (13A9) [NBP1-48309] - IHC analysis of N Cadherin in mouse liver using DAB with hematoxylin counterstain.	

Immunohistochemistry-Paraffin: N-Cadherin Antibody (13A9) [NBP1-48309] - Analysis of a FFPE tissue section of human brain using 1:200 dilution of N-Cadherin antibody. The staining was developed using HRP labeled anti-mouse secondary antibody and DAB reagent, and nuclei of cells were counter-stained with hematoxylin.	
Flow Cytometry: N-Cadherin Antibody (13A9) [NBP1-48309] - An intracellular stain was performed on HeLa cells with NBP1-48309AF488 and a matched isotype control (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 10 ug/mL for 30 minutes at room temperature. Both antibodies were conjugated to Alexa Fluor 488.	
Polarized RPE cell model characterization. (D) Representative images of several characteristic RPE markers: Na+ K+ ATPase, PEDF, E-cadherin, N-cadherin, claudin 19 and occludin (green); DAPI (blue); scale bar, 10 um. Image collected and cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/29269042), licensed under a CC-BY licence.	
Expression of hypoxia-inducible factor (HIF) proteins after bilateral renal ischemia or sham ischemia. Immunoblots for HIF-1α (A–C) and HIF-2α (D–F) of tissue extracts from the cortex and outer and inner medulla of the left kidneys of rats 24 h and 5 days following recovery from either sham ischemia (○) or bilateral renal ischemia (●); n = 6 per group. G: typical image of the gel following electrophoresis. H: typical image of the nitrocellulose membrane following transfer. Values are expressed as medians (25th percentile, 75th percentile). Paired comparisons were performed using the Mann-Whitney U-test. Because paired comparisons were made at two time points, P values were conservatively adjusted using the Dunn-Sidak method with k = 2. PTr, PT, and PTr*T are the outcomes of two-way analysis of variance on ranking with the factors treatment (Tr) and time (T). AU, arbitrary unit; I1, 24 h after ischemia; I5, 5 days after ischemia; S1, 24 h after sham ischemia; S5, 5 days after sham ischemia. Image collected and cropped by CiteAb from the following open publication (https://pubmed.ncbi.nlm.nih.gov/30110566), licensed under a CC-BY license. Not internally tested by Novus Biologicals.	

Publications

Belghasem M, Yin W, Lotfollahzadeh S et al. Tryptophan Metabolites Target Transmembrane and Immunoglobulin Domain-Containing 1 Signaling to Augment Renal Tubular Injury The American journal of pathology 2023-10-01 [PMID: 37676196] (WB, Mouse)

Padder RA, Bhat ZI, Ahmad Z et al. DRP1 Promotes BRAF(V600E)-Driven Tumor Progression and Metabolic Reprogramming in Colorectal Cancer Frontiers in Oncology 2021-03-02 [PMID: 33738242] (WB)

Lin YK, Zhang F, Lei WJ et al. Amnion-derived serum amyloid A1 participates in sterile inflammation of fetal membranes at parturition Inflammation research : official journal of the European Histamine Research Society ... [et al.] 2023-03-06 [PMID: 36879064]

?wierczewska M, Sterzy?ska K, Ruci?ski M et al. The response and resistance to drugs in ovarian cancer cell lines in 2D monolayers and 3D spheroids Biomedicine & pharmacotherapy = Biomedecine & pharmacotherapie 2023-07-11 [PMID: 37442067] (IHC, Human)

Huang M, Wang C, Yao Y et al. Mebendazole-Induced Blood-Testis Barrier Injury in Mice Testes by Disrupting Microtubules in Addition to Triggering Programmed Cell Death International journal of molecular sciences 2022-04-11 [PMID: 35457043]

Karim S, Burzangi AS, Ahmad A Et al. PI3K-AKT Pathway Modulation by Thymoquinone Limits Tumor Growth and Glycolytic Metabolism in Colorectal Cancer Int J Mol Sci 2022-02-26 [PMID: 35216429] (WB, Human)

Details:
Citation using the HRP version of this antibody.

Prasad A, Mahmood A, Gupta R Et Al. In cardiac muscle cells, both adrenergic agonists and antagonists induce reactive oxygen species from NOX2 but mutually attenuate each other\'s effects European journal of pharmacology 2021-07-13 [PMID: 34265295] (WB)

Matta R, Yousafzai MS, Murrell M, Gonzalez AL Endothelial cell secreted metalloproteinase-2 enhances neural stem cell N-cadherin expression, clustering, and migration FASEB journal : official publication of the Federation of American Societies for Experimental Biology 2021-02-01 [PMID: 33417253]

SAenz-de-Santa-MarIa I, Celada L, San JosE MartInez A et al. Blockage of Squamous Cancer Cell Collective Invasion by FAK Inhibition Is Released by CAFs and MMP-2 Cancers 2020-12-10 [PMID: 33321813] (WB, Human)

Beauchamp P, Jackson C B et al. 3D Co-culture of hiPSC-Derived Cardiomyocytes With Cardiac Fibroblasts Improves Tissue-Like Features of Cardiac Spheroids. Front Mol Biosci 2020-03-03 [PMID: 32118040] (IF/IHC, Mouse)

Becker A B, Qian J et al. Heart and Nervous System Pathology in Compound Heterozygous Friedreich Ataxia. J Neuropathol Exp Neurol 2017-01-08 [PMID: 28789479] (IF/IHC, Human)

Farjood Farhad, Vargis Elizabeth Physical disruption of cell-cell contact induces VEGF expression in RPE cells Physical disruption of cell-cell contact induces VEGF expression in RPE cells. Mol Vis. 2017-07-17 [PMID: 28761317] (ICC, Human)

Details:
Citation using the DyLight 680 version of this antibody.

More publications at http://www.novusbio.com/NBP1-48309

Procedures

Protocol specific for N Cadherin Antibody (13A9) [NBP1-48309]

Western Blot Protocol
1. Perform SDS-PAGE (4-12% MOPS) on samples to be analyzed, loading 25 ug of total protein per lane.
2. Transfer proteins to Nitrocellulose according to the instructions provided by the manufacturer of the transfer apparatus.
3. Rinse membrane with dH2O and then stain the blot using Ponceau S for 1-2 minutes to access the transfer of proteins onto the nitrocellulose membrane. Rinse the blot in water to remove excess stain and mark the lane locations and locations of molecular weight markers using a pencil.
4. Rinse the blot in TBS for approximately 5 minutes.
5. Block the membrane using 5% NFDM + 1% BSA in TBS + Tween, 1 hour at RT.
6. Rinse the membrane in dH2O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.
7. Dilute the anti-N Cadherin primary antibody (NBP1-48309) in blocking buffer and incubate 1 hour at room temperature.
8. Rinse the membrane in dH2O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.
9. Apply the diluted mouse-IgG HRP-conjugated secondary antibody in blocking buffer (as per manufacturers instructions) and incubate 1 hour at room temperature.
10. Wash the blot in wash buffer [TBS + 0.1% Tween] 3 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 (Pierce ECL).
Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%, provided it does not interfere with antibody-antigen binding.

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.

Staining:
1. Wash sections in deionized water three times for 5 minutes each.
2. Wash sections in wash buffer for 5 minutes.
3. Block each section with 100-400 ul blocking solution 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 biotinylated diluted secondary antibody. Incubate 30 minutes at room temperature.
7. Remove secondary antibody solution and wash sections three times with wash buffer for 5 minutes each.
8. Add 100-400 ul Streptavidin-HRP reagent to each section and incubate for 30 minutes at room temperature.
9. Wash sections three times in wash buffer for 5 minutes each.
10. Add 100-400 ul DAB substrate to each section and monitor staining closely.
11. As soon as the sections develop, immerse slides in deionized water.
12. Counterstain sections in hematoxylin.
13. Wash sections in deionized water two times for 5 minutes each.
14. Dehydrate sections.
15. Mount coverslips.

Immunocytochemistry Protocol

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

1. Remove culture medium and add 10% formalin to the dish. Fix at room temperature for 30 minutes.
2. Remove the formalin and add ice cold methanol. Incubate for 5-10 minutes.
3. Remove methanol and add washing solution (i.e. PBS). Be sure to not let the specimen dry out. Wash three times for 10 minutes.

4. To block nonspecific antibody binding incubate in 10% normal goat serum from 1 hour to overnight at room temperature.
5. Add primary antibody at appropriate dilution and incubate at room temperature from 2 hours to overnight at room temperature.
6. Remove primary antibody and replace with washing solution. Wash three times for 10 minutes.
7. Add secondary antibody at appropriate dilution. Incubate for 1 hour at room temperature.
8. Remove antibody and replace with wash solution, then wash for 10 minutes. Add Hoechst 33258 to wash solution at 1:25,0000 and incubate for 10 minutes. Wash a third time for 10 minutes.
9. Cells can be viewed directly after washing. The plates can also be stored in PBS containing Azide covered in Parafilm (TM). Cells can also be cover-slipped using Fluoromount, with appropriate sealing.

*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow safe laboratory procedures.

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Products Related to NBP1-48309

NB800-PC1	HeLa Whole Cell Lysate
HAF007	Goat anti-Mouse IgG Secondary Antibody [HRP]
NB720-B	Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]
NBP1-97005-0.5mg	Mouse IgG1 Isotype Control (MG1)

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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