

# Product Datasheet

## p19ARF/CDKN2A Antibody - BSA Free NB200-106

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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**NB200-106**

p19ARF/CDKN2A 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

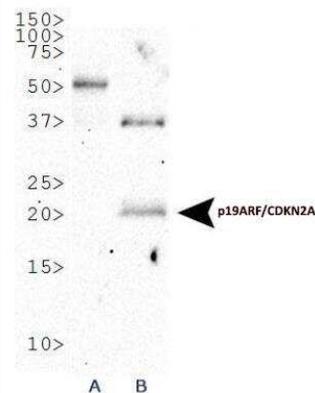
Product Description	
Description	Novus Biologicals Rabbit p19ARF/CDKN2A Antibody - BSA Free (NB200-106) is a polyclonal antibody validated for use in IHC, WB, ELISA and ICC/IF. Anti-p19ARF/CDKN2A Antibody: Cited in 48 publications. All Novus Biologicals antibodies are covered by our 100% guarantee.
Host	Rabbit
Gene ID	12578
Gene Symbol	Cdkn2a
Species	Mouse, Rat
Reactivity Notes	Rat reactivity reported in scientific literature (PMID: 31827530).
Immunogen	Synthetic peptide conjugated to KLH, derived from a sequence within residues 50-150 of mouse p19ARF/CDKN2A. [Swiss-Prot #Q64364]

Product Application Details	
Applications	Western Blot, Immunohistochemistry-Paraffin, ELISA, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen
Recommended Dilutions	Western Blot 2 ug/ml, ELISA 1:100-1:2000, Immunohistochemistry 10 ug/ml, Immunocytochemistry/ Immunofluorescence 1:50 - 1:100, Immunohistochemistry-Paraffin 10 ug/ml, Immunohistochemistry-Frozen 1 ug/ml
Application Notes	In WB a band can be seen at approximately 19 kDa. 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.

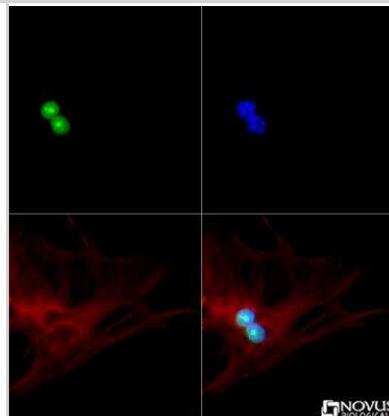


## Images

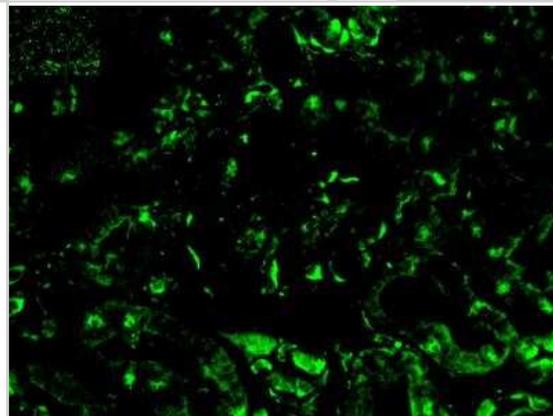
Western Blot: p19ARF/CDKN2A Antibody [NB200-106] - Analysis of p19ARF/CDKN2A in (A) NIH/3T3 and (B) MEF cells.



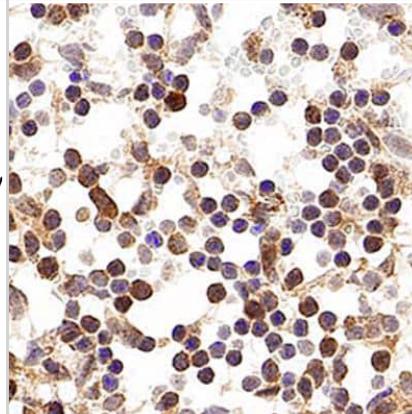
Immunocytochemistry/Immunofluorescence: p19ARF/CDKN2A Antibody [NB200-106] - Antibody was tested in MEF cells with DyLight 488 (green). Nuclei and alpha-tubulin were counterstained with DAPI (blue) and DyLight 550 (red).



Immunohistochemistry: p19ARF/CDKN2A Antibody [NB200-106] - Adult mouse kidney (glomerulus). Image from verified customer review.



Immunohistochemistry-Paraffin: p19ARF/CDKN2A Antibody [NB200-106] - Analysis of FFPE mouse spleen using p19ARF/CDKN2A antibody at 10 ug/mL on a Bond Rx autostainer (Leica Biosystems). The assay involved 20 minutes of heat induced antigen retrieval (HIER) using 10mM sodium citrate buffer (pH 6.0) and endogenous peroxidase quenching with peroxide block. The sections were incubated with primary antibody for 30 minutes and Bond Polymer Refine Detection (Leica Biosystems) with DAB was used for signal development followed by counterstaining with hematoxylin. Whole slide scanning and capturing of representative images was performed using Aperio AT2 (Leica Biosystems). Nuclear staining in the germinal centers of spleen was observed. Staining was performed by Histowiz.



## Publications

Ottone OK, Kim CJ, Collins JA, Risbud MV. The cGAS-STING Pathway Affects Vertebral Bone but Does Not Promote Intervertebral Disc Cell Senescence or Degeneration *Frontiers in Immunology* 2022-06-13 [PMID: 35769461] (Western Blot, Immunocytochemistry/ Immunofluorescence, Mouse)

Zhang Y, Pignolo RJ, Bram RJ. Accelerated Aging in Cyclophilin B-Deficient Mice Downstream of p21-Cip1/Waf1 *JBMR Plus* 2022-10-01 [PMID: 36248275] (Western Blot, Immunocytochemistry/ Immunofluorescence, Mouse)

Risbud M, Novais E, Tran V, et al. Long-term treatment with senolytic drugs Dasatinib and Quercetin ameliorates age-dependent intervertebral disc degeneration in mice *Nat Commun* 2021-09-04 [PMID: 34480023]

Chen H, Zhou J, Chen H et al. Bmi-1-RING1B prevents GATA4-dependent senescence-associated pathological cardiac hypertrophy by promoting autophagic degradation of GATA4 *Clinical and translational medicine* 2022-04-01 [PMID: 35390228] (WB, Mouse)

Brelstaff JH, Mason M, Katsinelos T Et al. Microglia become hypofunctional and release metalloproteases and tau seeds when phagocytosing live neurons with P301S tau aggregates *Science advances* 2021-10-22 [PMID: 34669475] (ICC/IF, Mouse)

Makino SI, Shirata N, Oliva Trejo JA, et al. Impairment of Proteasome Function in Podocytes Leads to CKD *Journal of the American Society of Nephrology : JASN* 2021-01-28 [PMID: 33510039]

Rui Y, Chen M, Li Y et al. CTGF Attenuates Tendon-Derived Stem/Progenitor Cell Aging *Stem Cells International* 2019-11-11 [PMID: 31827530] (WB, Rat)

Novais EM, Diekman BO, Shapiro IM, Risbud MV. p16Ink4a deletion in cells of the intervertebral disc affects their matrix homeostasis and senescence associated secretory phenotype without altering onset of senescence *Matrix Biol.* 2019-02-24 [PMID: 30811968] (IHC-P, Mouse)

Xie X, Tsai Sy, Tsai Mj. COUP-TFII regulates satellite cell function and muscular dystrophy. *J. Clin. Invest.* 2016-09-12 [PMID: 27617862]

Iaquinta PJ, Aslanian A, Lees JA. Regulation of the Arf/p53 tumor surveillance network by E2F. *Cold Spring Harb Symp Quant Biol* 2005-01-01 [PMID: 16869767] (Mouse)

Castillo JP, Frame FM, Rogoff HA et al. Human cytomegalovirus IE1-72 activates ataxia telangiectasia mutated kinase and a p53/p21-mediated growth arrest response. *J Virol* 2005-09-01 [PMID: 16103197]

Gapuzan ME, Schmah O, Pollock AD et al. Immortalized fibroblasts from NF-kappaB RelA knockout mice show phenotypic heterogeneity and maintain increased sensitivity to tumor necrosis factor alpha after transformation by v-Ras. *Oncogene* 2005-09-29 [PMID: 16027734] (Mouse)

More publications at <http://www.novusbio.com/NB200-106>



## Procedures

### Western blot protocol for p19ARF antibody (NB200-106)

#### 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

### ICC/IF protocol for p19ARF antibody (NB200-106)

#### 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 0.5% Triton X100 or other suitable detergent for 10 min.
4. Remove the permeablization buffer and wash three times for 10 minutes each in PBS with 0.1% Tween 20. 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.

\*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow proper laboratory procedures for the disposal of formalin.



**Immunohistochemistry-Paraffin Protocol for p19ARF/CDKN2A Antibody (NB200-106)**

## 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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### **Products Related to NB200-106**

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NB200-106PEP	p19ARF/CDKN2A 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

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### **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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