# **Product Datasheet**

## OGG1 Antibody NB100-106SS

Unit Size: 0.025 ml

Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.

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Updated 6/15/2014 v.20.1

## NB100-106SS

OGG1 Antibody

Product Information	
0.025 ml	
1.0 mg/ml	
Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.	
Polyclonal	
0.09% Sodium Azide	
Immunogen affinity purified	
Tris-citrate/phosphate, pH 7-8	
39 kDa	
Product Description	
Rabbit	
4968	
OGG1	
Human, Mouse, Rat, Primate	
Human, mouse, monkey, and rat.	
A peptide derived from human Ogg1 (within amino acids 1-100). [UniProt# O15527]	
Product Application Details	
Western Blot, ELISA, Flow Cytometry, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunoprecipitation	
ELISA, Flow Cytometry, Immunocytochemistry/Immunofluorescence 1:100 - 1:200, Immunohistochemistry, Immunohistochemistry-Frozen 1:25-1:100, Immunoprecipitation 1:10-1:500, Western Blot 1:500-1:1000	
This Ogg1 antibody is useful for Immunohistochemistry on frozen and paraffin- embedded sections, Immunoprecipitation Co-IP (PMID: 20956573), ELISA (PMID: 19506022) and Western Blot. In WB, it recognizes a band at ~39 kDa, representing Ogg1. In ICC/IF nuclear staining was observed in Hek293 cells. Flow data from customer review.	

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Western Blot: Ogg1 Antibody [NB100-106] - Western blot analysis of Ogg1 recombinant protein using Ogg1 antibody at 1 ug/ml.





Immunocytochemistry/Immunofluorescence: Ogg1 Antibody [NB100-106] - Ogg1 antibody was tested in Hek293 cells with DyLight 488 (green). Nuclei and alpha-tubulin were counterstained with DAPI (blue) and Dylight 550 (red).

Immunohistochemistry-Frozen: Ogg1 Antibody [NB100-106] - Detection of OGG1 in formaldehyde fixed frozen sections of the substantia nigra from a Rhesus macaque (Macaca mulatta) using NB100-106 at 10



ug/ml. Photo courtesy of Glen Kisby, Oregon Health Sciences University

Flow Cytometry: Ogg1 Antibody [NB100-106] - Baseline Ogg1 100 expression in human PBMC from a healthy donor. Image from confirmed customer review. of Max

Western Blot: Ogg1 Antibody [NB100-106] - Analysis of Ogg1 expression in Jurkat whole cell lysate.









#### **Publications**

Fang Q, Inanc B, Schamus S et al. HSP90 regulates DnA repair via the interaction between XRCC1 and DnA polymerase B. Nat Commun. 2014 Nov 26 [PMID: 25423885] (WB)

Wang H, Wang X, Chen G et al. Distinct Roles of Ape1 Protein, An Enzyme Involved in DNA Repair, in High or Low Linear Energy Transfer Ionizing Radiation-induced Cell Killing. J Biol Chem. 2014 Sep 10 [PMID: 25210033]

Kumar P, Rao GN, Pal BB, Pal A. Hyperglycemia-induced oxidative stress induces apoptosis by inhibiting PI3kinase/Akt and ERK1/2 MAPK mediated signaling pathway causing downregulation of 8-oxoG-DNA glycosylase levels in glial cells Int. J. Biochem. Cell Biol. 2014 Jun 04 [PMID: 24907397] (WB, ICC/IF, Rat)

Details:

Antibody used on Rat's astroglial cells/ C6 cell line using WB (Fig. 6B, Fig. 7 A, B, C) and ICC-IF (Fig. 7 G) in experiments involving hyperglycemia-induced oxidative stress in vitro model.

Sagar S, Kumar P, Behera RR, Pal A. Effects of CEES and LPS synergistically stimulate oxidative stress inactivates OGG1 signaling in macrophage cells J. Hazard. Mater. 2014 Jun 06 [PMID: 24976129] (WB, ICC/IF, Mouse)

Details:

Ogg1 antibody used for WB and ICC-IF in RAW264.7 macrophages cells exposed to 2-chloroethyl ethyl sulphide (CEES) or lipopolysaccharide (LPS) for 24h, and/or treated with N-acetylcysteine/NAC fluorescein isothiocyanate (FITC)-conjugated secondary antibody (Images: WB - Fig. 8b, 9a and b, Fig 10a; ICC-IF - Fig 10C)

Kim SJ, Cheresh P, Williams D et al. Mitochondria-targeted Ogg1 and Aconitase-2 Prevent Oxidant-induced Mitochondrial DNA Damage in Alveolar Epithelial Cells. J. Biol. Chem. 2014 Feb 28 [PMID: 24429287] (WB, Human)

Sony S. Oxidative damage to dna in alzheimers disease. PhD Theses -Chemistry 2013 (WB, Human)

Huang E, Qu D, Zhang Y et al. The role of Cdk5-mediated apurinic/apyrimidinic endonuclease 1 phosphorylation in neuronal death. Nat Cell Biol. 2010 Jun [PMID: 20473298] (WB, Mouse)

Chen X, Wang J, Guo W et al. Two functional variations in 5'-UTR of hoGG1 gene associated with the risk of breast cancer in Chinese. Breast Cancer Res Treat 2011 Jun [PMID: 21153698]

Cordero MD, Alcocer-Gomez E, Culic O et al. NLRP3 Inflammasome is activated in Fibromyalgia: the effect of Coenzyme Q10. Antioxid Redox Signal 2013 Jul 25 [PMID: 23886272]

Kubo N, Morita M, Nakashima Y et al. Oxidative DNA damage in Human esophageal cancer: clinicopathological analysis of 8-hydroxydeoxyguanosine and its repair enzyme. Dis Esophagus 2013 Aug 1 [PMID: 23902537] (IHC-P, Human)

Dziaman T, Banaszkiewicz Z, Roszkowski K et al. 8-Oxo-7,8-dihydroguanine and uric acid as efficient predictors of survival in colon cancer patients. Int J Cancer 2013 Jul 5 [PMID: 23832862] (IHC-P, Human)

Lee YK, Youn HG, Wang HJ, Yoon G. Decreased mitochondrial OGG1 expression is linked to mitochondrial defects and delayed hepatoma cell growth. Mol Cells 2013 May 14 [PMID: 23677377] (WB, Human)

More publications at http://www.novusbio.com/NB100-106



#### **Procedures**

## Western blot Protocol for Ogg1 Antibody (NB100-106)

Western Blot Procedure

- 1. Run 50 ug of protein on a 4-20% Tris-glycine mini-gel at 125V for 90 minutes.
- 2. Equilibrate gel, nitrocellulose membrane, Whatman paper, and blotting pads in transfer buffer for 15 minutes.
- 3. Transfer protein to the membrane at 25V for 90 minutes.
- 4. Allow membrane to air-dry.
- 5. Block membrane with 1XPBS/3% BSA for 1 hour at room temperature (23-27 degrees C).
- 6. Wash membrane twice, for 5 minutes each, with 1XPBS/0.05% Tween-20 (PBST).
- 7. Incubate membrane with NB100-106 (anti-hOGG1), diluted in 1XPBS/1% BSA, for 1 hour at room temperature.
- 8. Wash membrane once for 15 minutes, then four times for 5 minutes each, with PBST.
- 9. Incubate membrane with goat anti-rabbit IgG-HRP, diluted in 1XPBS/1% BSA, for 1 hour at room temperature.
- 10. Wash membrane once for 15 minutes, then four times for 5 minutes each, with PBST.
- 11. Detect cross-reacting proteins using Renaissance Chemiluminescence Reagent Plus kit from NEN Life Sciences.

## Immunohistochemistry Protocol for Ogg1 Antibody (NB100-106)

Immunohistochemistry - FFPE sections

I. Deparaffinization:

A. Treat slides with Xylene: 3 changes for 5 minutes each. Drain slides for 10 seconds between changes.

B. Treat slides with 100% Reagent Alcohol: 3 changes for 5 minutes each. Drain slides for 10 seconds between changes.

II. Quench Endogenous Peroxidase:

A. Place slides in peroxidase quenching solution: 15-30 minutes.

To Prepare 200 ml of Quenching Solution:

Add 3 ml of 30% Hydrogen Peroxide to 200 ml of Methanol.

Use within 4 hours of preparation

B. Place slides in distilled water: 2 changes for 2 minutes each.

III. Retrieve Epitopes:

A. Preheat Citrate Buffer. Place 200 ml of Citrate Buffer Working Solution into container, cover and place into steamer. Heat to 90-96 degrees Celcius.

- B. Place rack of slides into hot Citrate Buffer for 20 minutes. Cover.
- C. Carefully remove container with slides from steamer and cool on bench, uncovered, for 20 minutes.
- D. Slowly add distilled water to further cool for 5 minutes.

E. Rinse slides with distilled water. 2 changes for 2 minutes each.

IV. Immunostaining Procedure:

A. Remove each slide from rack and circle tissue section with a hydrophobic barrier pen (e.g. Liquid Blocker-Super Pap Pen).

- B. Flood slide with Wash Solution. Do not allow tissue sections to dry for the rest of the procedure.
- C. Drain wash solution and apply 4 drops of Blocking Reagent to each slide and incubate for 15 minutes.

D. Drain Blocking Reagent (do not wash off the Blocking Reagent), apply 200 ul of primary antibody solution to each slide, and incubate for 1 hour.

- E. Wash slides with Wash Solution: 3 changes for 5 minutes each.
- F. Drain wash solution, apply 4 drops of Secondary antibody to each slide and incubate for 1 hour.
- G. Wash slides with Wash Solution: 3 changes for 5 minutes each.

H. Drain wash solution, apply 4 drops of DAB Substrate to each slide and develop for 5-10 minutes. Check development with microscope.

I. Wash slides with Wash Solution: 3 changes for 5 minutes each.

J. Drain wash solution, apply 4 drops of Hematoxylin to each slide and stain for 1-3 minutes. Increase time if darker counterstaining is desired.



K. Wash slides with Wash Solution: 2-3 changes for 2 minutes each.

L. Drain wash solution and apply 4 drops of Bluing Solution to each slide for 1-2 minutes.

M. Rinse slides in distilled water.

N. Soak slides in 70% reagent alcohol: 3 minutes with intermittent agitation.

O. Soak slides in 95% reagent alcohol: 2 changes for 3 minutes each with intermittent agitation.

P. Soak slides in 100% reagent alcohol: 3 changes for 3 minutes each with intermittent agitation. Drain slides for 10 seconds between each change.

Q. Soak slides in Xylene: 3 changes for 3 minutes each with intermittent agitation. Drain slides for 10 seconds between each change.

R. Apply 2-3 drops of non-aqueous mounting media to each slide and mount coverslip.

S. Lay slides on a flat surface to dry prior to viewing under microscope.

#### NOTES:

Use treated slides (e.g. HistoBond) to assure adherence of FFPE sections to slide.

Prior to deparaffinization, heat slides overnight in a 60 degrees Celcius oven.

All steps in which Xylene is used should be performed in a fume hood.

For Epitope Retrieval, a microwave or pressure cooker may be substituted for the steamer method. Adjust times as necessary depending on conditions.

For the initial IHC run with a new primary antibody, test tissues with and without Epitope Retrieval. In some instances, Epitope Retrieval may not be necessary.

200 ul is the recommended maximum volume to apply to a slide for full coverage. Using more than 200 ul may allow solutions to wick off the slide and create drying artifacts. For small tissue sections less than 200 ul may be used. 5 minutes of development with DAB Substrate should be sufficient. Do not develop for more than 10 minutes. If 5 minutes of development causes background staining, further dilution of the primary antibody may be necessary. Hematoxylin should produce a light nuclear counterstain so as not to obscure the DAB staining. Counterstain for 1-1 1/2 minutes for nuclear antigens. Counterstain for 2-3 minutes for cytoplasmic and membranous antigens. If darker counterstaining is desired increase time (up to 10 minutes).

#### ICC/IF Protocol for Ogg1 Antibody (NB100-106)

Immunocytochemistry Protocol

Culture cells to appropriate density on suitable glass coverslips 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 5-10 minutes.

2. Remove the formalin and add 0.5% Triton-X 100 in TBS to permeabilize the cells. Incubate for 5-10 minutes.

3. Remove the permeabilization buffer and add wash buffer (i.e. PBS or PBS with 0.1% Tween-20). Be sure to not let the specimen dry out. Gently wash three times for 10 minutes.

4. Alternatively, cells can be fixed with -20C methanol for 10 min at room temperature. Remove the methanol and rehydrate in PBS for 10 min before proceeding.

5. To block nonspecific antibody binding incubate in 10% normal goat serum for 1 hour at room temperature.

6. Add primary antibody at appropriate dilution and incubate at room temperature for 1 hour or at 4 degrees C overnight.

7. Remove primary antibody and replace with wash buffer. Gently wash three times for 10 minutes.

8. Add secondary antibody at the appropriate dilution. Incubate for 1 hour at room temperature.

9. Remove antibody and replace with wash buffer. Gently wash three times for 10 minutes.

10. Nuclei can be staining with 4',6' diamino phenylindole (DAPI) at 0.1 ug/ml, or coverslips can be directly mounted in media containing DAPI.

11. Cells can now be viewed with a fluorescence microscope.

\*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.







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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.

For more information on our guarantee, please visit www.novusbio.com/guarantee.

