# **Product Datasheet**

# DNA/RNA Damage Antibody (15A3) - BSA Free NB110-96878

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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# NB110-96878

DNA/RNA Damage Antibody (15A3) - 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	Monoclonal
Clone	15A3
Preservative	0.09% Sodium Azide
Isotype	lgG2b
Purity	Protein G purified
Buffer	PBS, 50% Glycerol
Product Description	
Host	Mouse
Species	Human, Mouse, Non-species specific
Reactivity Notes	Use in Human reported in scientific literature (PMID:33482333) Mouse reactivity reported in scientific literature (PMID: 31226694).
Specificity/Sensitivity	Recognizes markers of oxidative damage to DNA (8-hydroxy-2'-deoxyguanosine, 8-hydroxyguanine and 8-hydroxyguanosine).
Immunogen	8-hydroxy-guanosine-BSA and-casein conjugates
Product Application Details	
Applications	Dot Blot, ELISA, Flow Cytometry, Immunoassay, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin, Immunoprecipitation, Functional Assay
Recommended Dilutions	Flow Cytometry, ELISA, Immunohistochemistry 1:1000, Immunocytochemistry/ Immunofluorescence, Immunoprecipitation, Immunohistochemistry-Paraffin 1:1000, Immunoassay, Dot Blot, Functional Assay
Application Notes	Use in immoprecipitation reported in scientific literature (PMID 26510519). Use in Immunoassay reported in scientific literature (PMID:31625228).



#### Images

Immunocytochemistry/Immunofluorescence: DNA/RNA Damage Antibody (15A3) [NB110-96878] - visualized on a retinal injury



Immunohistochemistry: DNA/RNA Damage Antibody (15A3) [NB110-96878] - Immunohistochemistry analysis using Mouse Anti-DNA/RNA DamageMonoclonal Antibody, Clone 15A3 (NB110-96878). Tissue: inflamed colon. Species: Mouse. Fixation: Formalin. Primary Antibody: Mouse Anti-DNA/RNA DamageMonoclonal Antibody (NB110-96878) at 1:1000000 for 12 hours at 4C. Secondary Antibody: Biotin Goat Anti-Mouse at 1:2000 for 1 hour at RT. Counterstain: Mayer Hematoxylin (purple/blue) nuclear stain at 200 I for 2 minutes at RT. Magnification: 40x. With anti-microbial.

DNA/RNA Damage Antibody (15A3) [NB110-96878] -Immunohistochemistry analysis using Mouse Anti-DNA/RNA DamageMonoclonal Antibody, Clone 15A3 (NB110-96878). Tissue: backskin. Species: Mouse. Fixation: Bouin's Fixative and paraffinembedded. Primary Antibody: Mouse Anti-DNA/RNA DamageMonoclonal Antibody (NB110-96878) at 1:100 for 1 hour at RT. Secondary Antibody: FITC Goat Anti-Mouse (green) at 1:50 for 1 hour at RT.







#### **Publications**

Jie Tang, Xiaoxue Lu, Tao Zhang, Yuyang Feng, Qiaolin Xu, Jing Li, Yuanzhi Lan, Huaxing Luo, Linghai Zeng, Yuanyuan Xiang, Yan Zhang, Qian Li, Xuhu Mao, Bin Tang, Dongzhu Zeng Shiga toxin 2 A-subunit induces mitochondrial damage, mitophagy and apoptosis via the interaction of Tom20 in Caco-2 cells Heliyon 2023-09-09 [PMID: 37809632]

Gonzalez M, Prashar T, Connaughton H et al Restoring Sperm Quality Post-Cryopreservation Using Mitochondrial-Targeted Compounds Antioxidants (Basel) 2022-09-14 [PMID: 36139882] (Immunocytochemistry/ Immunofluorescence, Human)

Soria-Meneses PJ, Jurado-Campos A, G□mez-Rubio V et al. Determination of Ram (Ovis aries) Sperm DNA Damage Due to Oxidative Stress: 8-OHdG Immunodetection Assay vs. SCSA(□) Animals (Basel) 2022-11-25 [PMID: 36496807] (Flow Cytometry)

Fraser BA, Wilkins AL, De Iuliis GN et al. Development of a model for studying the developmental consequences of oxidative sperm DNA damage by targeting redox-cycling naphthoquinones to the Sertoli cell population Free radical biology & medicine 2023-06-23 [PMID: 37356777] (Immunohistochemistry, Mouse)

Langford MP, Srur L, Redens TB, Byrd WA Conjunctival epitheliopathy induced by topical exposure to bacterial peptidoglycan, muramyl dipeptide Experimental eye research 2023-01-10 [PMID: 36634837] (IHC-P, ICC/IF, Rabbit)

Shrestha S, Erikson G, Lyon J et al. Aging compromises human islet beta cell function and identity by decreasing transcription factor activity and inducing ER stress Science advances 2022-10-07 [PMID: 36197983] (IHC-P, Human)

Sadeghi N, Tavalaee M, Kiani-Esfahani A Et al. Apoptotic M540 bodies present in human semen interfere with flow cytometry-assisted assessment of sperm DNA fragmentation and oxidation Basic and clinical andrology 2021-10-21 [PMID: 34670490] (FLOW)

Berby B, Bichara C, Rives-Feraille A et al. Oxidative Stress Is Associated with Telomere Interaction Impairment and Chromatin Condensation Defects in Spermatozoa of Infertile Males Antioxidants (Basel, Switzerland) 2021-04-12 [PMID: 33921485]

Zhao F, Whiting S, Lambourne S, et al. Melatonin alleviates heat stress-induced oxidative stress and apoptosis in human spermatozoa Free radical biology & medicine 2021-01-19 [PMID: 33482333] (ICC/IF, Human)

Vorilhon S, Brugnon F et al. Accuracy of human sperm DNA oxidation quantification and threshold determination using an 8-OHdG immuno-detection assay. Hum Reprod 2018-01-04 [PMID: 29579272] (FLOW, Human)

Champroux A, Goubely C, Henry-Berger J et al. Three-Dimensional Confocal Analysis of Chromosome Positioning Coupled with Immunofluorescence in Mouse Sperm Nuclei Methods Mol. Biol. 2020-08-22 [PMID: 32822037]

Aitken R, Whiting S, Connaughton H et al. A novel pathway for the induction of DNA damage in human spermatozoa involving extracellular cell-free DNA Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis 2020-09-01 [PMID: 32920458] (Human)

More publications at <a href="http://www.novusbio.com/NB110-96878">http://www.novusbio.com/NB110-96878</a>



#### **Procedures**

Serum protocol for DNA/RNA Damage Antibody (NB110-96878) Protocol specific for DNA/RNA Damage Antibody (NB110-96878): Tissue Preparation

8-OHdG monoclonal antibody reacts on both 50 um frozen tissue sections and paraffin-embedded sections. Tissue should be dissected fresh and fixed in periodate-lysine-paraformaldehyde (PLP) at 4C over night.

PLP

Heat 1 L dH2O to 60C.

Add 60 g paraformaldehyde. Add 33 g dibasic NaPO4.

Cool to room temperature in a cold water bath. Add 9 g monobasic NaPO4.

Add 6.45 g Na-m-periodate. Add 41.1 g lysine (HCl salt). Filter and dilute to 3 L with dH2O.

Adjust pH to 7.6 with 1.0 N NaOH approx. (20-30 ml).

Tissue prepared for frozen sectioning must be cryoprotected in a 20% glycerol-2% DMSO solution in phosphate buffer for 24-48 hours. Tissue will sink to the bottom of container when fully penetrated. This will eliminate freezing artifact from cutting.

Glycerol-DMSO (for 3 L) 2.4 L 0.1 M phosphate buffer 600 ml glycerol 60 ml DMSO

0.1 M Phosphate Buffer, pH 7.4 (for 1 L)

1 L dH2O

11 g dibasic NaPO4

3 g monobasic NaPO4

After frozen sectioning, tissue should be stored in phosphate buffer with 0.08% sodium azide.

Staining Sections By DAB Procedure

Paraffin-embedded sections must be deparaffinized by sequential immersion in the following for 3 minutes each: xylene (twice), absolute ethanol (twice). Agitate gently in each solution. Proceed with the following procedure.

 Pretreat sections with a methanol-peroxide solution to eliminate endogenous peroxidases. Methanol-Peroxide
 ml absolute methanol
 ml 33% H2O2
 Incubate sections in methanol-peroxide solution for 30 minutes, room temperature.

2. Wash sections 3 times for 10 minutes each in 0.1 M phosphate buffered saline (PBS) PBS, pH 7.4 (for 1 L)
1 L dH2O
11 g dibasic NaPO4



3 g monobasic NaPO4 8.5 g NaCl

3. Incubate sections for 1 hour in 10% normal goat serum in PBS.

4. Incubate sections in the primary antibody for 18-24 hours at room temperature. Depending on the nature of the sample, a shorter incubation time may be used. It is recommended that a concentration range of 1-10 ug/ml be evaluated in order to determine the optimal concentration for each type of tissue sample. Dilute antibody in PBS containing 0.3% Triton X-100, 0.08% sodium azide and 2% normal goat serum.

NOTE: A humidified chamber is necessary when staining paraffin sections. Slides should be placed flat and primary antibody applied over the section, covering it completely.

5. Rinse sections 3 times for 10 minutes each in PBS.

6. Incubate for 3 hours with peroxidase-conjugated goat anti-mouse IgG (Boehringer-Mannheim, Indianapolis, IN) diluted 1:300 in PBS with 2% normal goat serum.

7. Rinse sections 3 times for 10 minutes each in PBS.

8. Incubate sections for 5-10 minutes in a solution of 0.5 mg/ml 3,3' diamino-benzidine tetrahydrochloride (DAB, Sigma, St. Louis, MO) and 0.005% hydrogen peroxide in 0.05 M tris HCl buffer, pH 7.6 plus imidazole (10 ml/110 ml Tris buffer).

50 mM Tris Buffer, pH 7.6 1 L dH2O 6 g Trizma base 3 ml concentrated HCI (37%)

Sodium Imidazole 100 ml 0.1 M phosphate buffer 0.7 g sodium imidazole

9. Rinse sections 3 times for 10 minutes each in PBS.

10. Mount free-floating sections on subbed slides and air dry.

Subbing Solution 500 ml dH2O

2.5 g gelatin

0.25 g chromium potassium sulfate

Heat to 60C. Filter and proceed to coat slides. Once slides are air dried, sections can be mounted.

11. Dehydrate mounted/paraffin sections by sequential immersion in the following for 3 minutes each: 70% ethanol, 95% ethanol, absolute ethanol, xylene. Agitate gently in each solution.

12. Apply coverslip with Permount in a chemical fume hood





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# Products Related to NB110-96878

HAF007	Goat anti-Mouse IgG Secondary Antibody [HRP]
NB720-B	Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]
NBP2-27231	Mouse IgG2b Isotype Control (MPC-11)

### 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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