# **Product Datasheet**

# GFP Antibody - BSA Free NB100-1770SS

Unit Size: 0.02 mg

Store at -20C. Avoid freeze-thaw cycles.

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# NB100-1770SS

GFP Antibody - BSA Free

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Product Information	
Unit Size	0.02 mg
Concentration	Please see the vial label for concentration. If unlisted please contact technical services.
Storage	Store at -20C. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.01% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Product Description	
Description	GFP antibody was prepared from monospecific antiserum by immunoaffinity chromatography using Green Fluorescent Protein (Aequorea victoria) coupled to agarose beads followed by solid phase adsorption(s) to remove any unwanted reactivities. Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Goat Serum and purified and partially purified Green Fluorescent Protein (Aequorea victoria)  Store this antibody at -20C prior to opening. Aliquot contents and freeze at -20C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4C as an undiluted liquid. Dilute only prior to immediate use.
Host	Goat
Species	Non-species specific
Reactivity Notes	No reaction was observed against Human, or Rat serum proteins. Known Cross Reactivity: rGFP. YFP differs from GFP due to a mutation at Thr203Tyr; antibodies raised against full-length GFP should also detect YFP and other variants. Reactivity in transgenic mice with GFP. Reactivity in human cell lines transfected will a GFP construct.
Specificity/Sensitivity	No reaction was observed against Human, Mouse or Rat serum proteins.
Immunogen	The immunogen is a Green Fluorescent Protein (GFP) fusion protein corresponding to the full length amino acid sequence (246aa) derived from the jellyfish Aequorea victoria. (Uniprot: P42212)
Product Application Details	
Applications	Western Blot, ELISA, Electron Microscopy, Flow Cytometry, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation, Proximity Ligation Assay, Immunohistochemistry Free-Floating, Knockdown Validated, Knockout Validated
Recommended Dilutions	Western Blot 1:1000-1:10000, Flow Cytometry 1:10 - 1:1000, ELISA 1:10000-1:30000, Immunohistochemistry 1:200-1:1000, Immunocytochemistry/ Immunofluorescence 1:500, Immunoprecipitation 1:10 - 1:500, Immunohistochemistry-Paraffin 1:10 - 1:500, Immunohistochemistry-Frozen 1:10 - 1:500, Proximity Ligation Assay, Electron Microscopy 1:10 - 1:500, Immunohistochemistry Free-Floating 1:10 - 1:500, Knockout Validated, Knockdown Validated

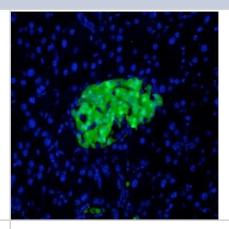


### **Application Notes**

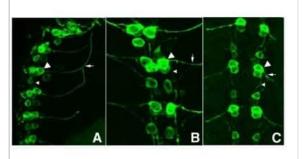
This product is designed to detect GFP and its variants. Goat This product has been tested by ELISA, SDS-PAGE, Western blot, and Immunofluorescence. This product is ideal for western blotting, ELISA, immunofluorescence, IHC, and IP. This antibody can be used to detect GFP by ELISA (sandwich or capture) for the direct binding of antigen and recognizes wild type, recombinant and enhanced forms of GFP. Biotin conjugated polyclonal anti-GFP used in a sandwich ELISA is well suited to titrate GFP in solution using this antibody in combination with monoclonal anti-GFP using either form of the antibody as the capture or detection antibody. However, use the monoclonal form only for the detection of wild type or recombinant GFP as this form does not sufficiently detect 'enhanced' GFP. The detection antibody is typically conjugated to biotin and subsequently reacted with streptavidin-HRP. Fluorochrome conjugated polyclonal anti-GFP can be used to detect GFP by immunofluorescence microscopy in prokaryotic (E.coli) and eukaryotic (CHO cells) expression systems and detects GFP containing inserts. Significant amplification of signal is achieved using fluorochrome conjugated polyclonal anti-GFP relative to the fluorescence of GFP alone. For immunoblotting use either alkaline phosphatase or peroxidase conjugated polyclonal anti-GFP to detect GFP or GFP-containing proteins on western blots. Researchers should determine optimal titers for applications.

## **Images**

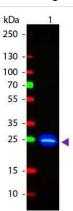
Analysis using the Biotin conjugate of NB100-1770. Staining of transgenic mouse pancreas, expressing GFP in beta cells. Image courtesy of product review by Dr. Yves Heremans of Vrije Universiteit Brussel.



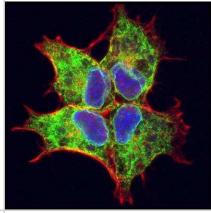
Analysis of Biotin conjugate of NB100-1770. Tissue: Drosophila melanogaster late stage embryonic central nervous system. Fixation: 0.5% PFA. Antigen retrieval: not required. Primary antibody: Anti-GFP antibody at a 1:1000 for 1 h at RT.



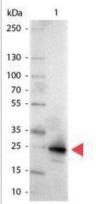
Analysis using the FITC conjugate of NB100-1770. Lane 1: GFP (50 ug). Primary antibody: None. Secondary antibody: Fluorescein goat secondary antibody at 1:1000 for 60 min at RT. Block for 30 minutes.



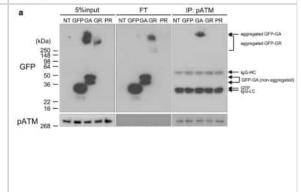
HEK293 cells stable transfected with integrin alpha8-GFP, were immunostained for GFP (green) and counterstained with phalloidin (red) and DAPI (BLUE). ICC/IF image submitted by a verified customer review.



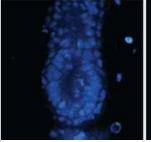
Analysis using the Alkaline Phosphatase conjugate of NB100-1770. Lane 1: GFP (50 ng). Alkaline Phosphatase GFP secondary antibody at 1:1000 for 60 min at RT. Predicted/Observed size: 28 kDa for GFP.

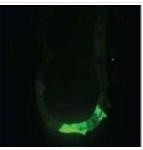


9 Poly-GA co-immuno - precipitation with pATM. Aggregated poly-GA selectively co-immunoprecipitates with pATM. Image collected and cropped by CiteAb from the following publication (www.link.springer.com/article/10.1007%2Fs00401-019-02082-0) licensed under a CC-BY license.

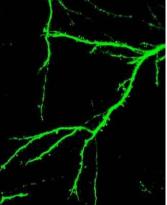


E5.5 Hex-GFP transgenic mouse embryo tissue. Primary antibody: Goat anti-GFP at 1:500 dilution. Secondary antibody: Fluorochrome conjugated Anti-goat IgG antibody at 1:10,000 for 45 min at RT. Staining: GFP as green fluorescent signal with DAPI blue counterstain.

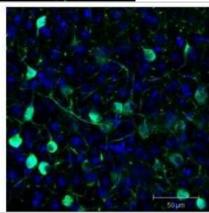




IF analysis of GFP in mouse brain. Image courtesy of product review by Tatyana Pivneva.



Tissue: Sf-1:Cre mice crossed to the Z/EG reporter line. Mouse brain (coronal view, 20X magnification). Fixation: 4%PFA/PBS o/n, and subsequently transferred to a 30% sucrose solution. Antigen retrieval: frozen in OCT freezing medium (Sakura) and cryostat sectioned at 40 microns. Primary antibody: Goat anti-GFP was used at 1:500 dilution in free floating immunohistochemistry to detect GFP. Secondary antibody: Fluorochrome conjugated Anti-goat IgG antibody at 1:500 for 45 min at RT. Localization: Sf-1+ neurons and their processes of the ventromedial nucleus of the hypothalamus. Staining: eGFP as green fluorescent signal and sections were counterstained with DAPI.



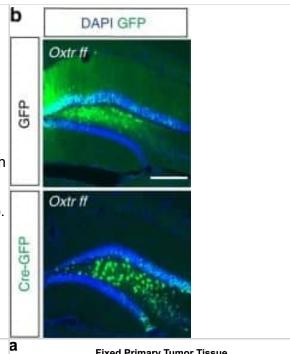
Analysis of DyLight 488 conjugate of NB100-1770. Human breast carcinoma tissue. Fixation: 0.5% PFA. Antigen retrieval: not required. Primary antibody: Anti-Histone and Anti-Tubulin antibody at 10 ug/mL for 1 h at RT. Secondary antibody: DyLight 488.

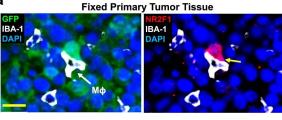


GFP-positive transplanted NPCs have morphological features of hippocampal pyramidal neurons at day 90 after grafting. IHC image submitted by a verified customer review. 1 2 Lane 1: HeLa cells. Lane 2: mock transfected HeLa cell lysate. Load: 35 ug per lane. Primary antibody: GFP antibody at 1 ug/ml for 1 h at room temperature. Secondary antibody: IRDye(R) 800 conjugated Donkey-a-Goat IgG [H&L] MX7 () secondary antibody at 1:2,500 for 45 min at RT. Block: 5% BLOTTO overnight at 4C. Predicted/Observed size: 27 kDa, 33 kDa for GFP. Other band(s): none. 33 kDa Western Blot of GFP Antibody. Lane 1: Opal Prestained Molecular Weight Marker Multi-lysate Western Blot of GFP Antibody. Marker: Opal Pre-stained ladder kDa 7 8 9 10 11 12 13 245 135 100 - 100 75 - 63 48 35 25 20 20 17 - 17

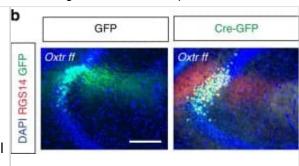
Immunocytochemistry/ Immunofluorescence: GFP Antibody [NB100-1770] - Viral recombination of Oxtr in anterior DG hilar neurons impairs discrimination of social, but not non-social, stimuli. a Schematic illustrating viral injection & behavioral testing timeline. b Representative images of Cre & GFP virus infection in aDG. Scale bar, 200 um. c Representative images & quantifications of cFos immunoreactivity in granule cell layer of aDG (GFP: n = 7, Cre: n = 4). Scale bar, 75 µm. d Behavioral schematic (top) & quantification (bottom) of single object exploration (GFP: n = 11, Cre: n = 6). e Behavioral schematic (top) & quantification (bottom) of novel objection recognition (GFP: n = 11, Cre: n = 6). Quantifications are displayed as Habituation (trials 1–3), Test (trial 4), & discrimination ratio (trial 4). f Behavioral schematic (top) & quantification (bottom) of social exploration test (GFP: n = 11, Cre: n = 6). g Behavioral schematic (top) & quantification (bottom) of social discrimination task (GFP: n = 11, Cre: n = 6). Quantifications are displayed as Habituation (trials 1-3), Test (trial 4), & discrimination ratio (trial 4). All data are displayed as mean ± SEM Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/29222469), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Immunohistochemistry: GFP Antibody [NB100-1770] - Macrophages regulate dormancy in tumor cells, a Representative image of triple immunofluorescently stained in E0771-GFP primary tumor tissue for tumor cells, macrophages, & NR2F1. Green = GFP; Red = NR2F1; White = IBA-1; Blue = DAPI. White arrow shows a macrophage. The vellow arrow contact between an NR2F1-positive tumor cell & a macrophage. Mo=Macrophage. Scale bar=20 µm. b Quantification showing frequency of distances between NR2F1+ tumor cells to nearest macrophage in primary tumor. Data is normalized to frequency of distances between all DAPI+ nuclei to nearest TMEM. Bar = mean. Error bars = ±SEM, n = 34 fields of view (551 × 316 µm2) in 4 animals. For comparison between 0 & 200 µm bins a two-tailed Mann-Whitney test used (p < 0.0001). \*\*\*\*p < 0.0001. c Representative immunofluorescence images of NR2F1 expression in E0771-GFP tumor cells cultured alone, in direct contact w/ BAC1.2F5 macrophages, or in direct contact w/ HUVEC endothelial cells. White arrows show macrophages or endothelial cells in direct contact w/ a tumor cell. Green = GFP; Red = NR2F1; Blue = DAPI. TC = Tumor Cell. Mφ = Macrophage. EC = Endothelial Cell. Scale bar = 15 µm. d Percentage of NR2F1-positive tumor cells from each group in C. TC alone: n = 777 cells in 9 independent experiments;  $TC+M\phi$ ; 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 $\phi$  (p = 0.0039), & for TC vs. TC+EC (p = 1), a two-tailed Kruskal-Wallis test w/ Dunn's multiple comparisons adjustment used. For TC+M\phi vs. TC+EC (0.012), a twotailed one-way ANOVA w/ Sidak's multiple comparison adjustment used. \*p < 0.05. \*\*p < 0.01; ns = not significant. Source data are provided as a Source Data file. Image collected & cropped by CiteAb from following publication (https://pubmed.ncbi.nlm.nih.gov/35110548), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

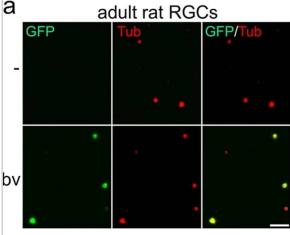




Immunocytochemistry/ Immunofluorescence: GFP Antibody [NB100-1770] - Viral recombination of Oxtr in aCA2/CA3distal impairs discrimination of social, but not non-social, stimuli. a Schematic illustrating viral injection & behavioral testing timeline. b Representative images of Cre & GFP virus infection in aCA2/CA3. Scale bar, 200 µm. c Behavioral schematic (top) & quantification (bottom) of single object exploration (GFP: n = 7, Cre: n = 10). d Behavioral schematic (top) & quantification (bottom) of novel objection recognition (GFP: n = 7, Cre: n = 7= 10). Quantifications are displayed as Habituation (trials 1–3), Test (trial 4), & discrimination ratio (trial 4), e Behavioral schematic (top) & quantification (bottom) of social exploration test (GFP: n = 7, Cre: n = 10). f Behavioral schematic (top) & quantification (bottom) of social discrimination task (GFP: n = 7, Cre: n = 10). Quantifications are displayed as Habituation (trials 1-3), Test (trial 4), & discrimination ratio (trial 4). All data are displayed as mean ± SEM Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/29222469), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

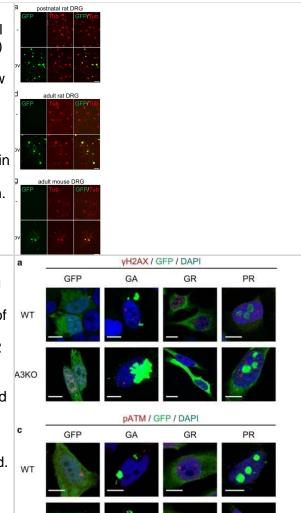


Immunocytochemistry/ Immunofluorescence: GFP Antibody [NB100-1770] - Transduction of mature retinal ganglion cells. Dissociated retinal ganglion cells (RGCs) isolated from adult rats (a-c), mice (d-f) & zebrafish (q-i) were transduced with baculovirus (bv) encoding fGFP (rat & mice) or DsRed (zebrafish). Representative pictures of vehicle- (-) & by -treated cultures (a,d,g) visualize transduced RGCs (GFP & DsRed, respectively) that were either co-stained with the neuronal marker βIIItubulin (Tub, red) at 2 days after transduction (d.a.t) for rat & mice RGCs (a,d) or identified by EGFP expression for zebrafish RGCs at 6 d.a.t. (g). The percentage of transduced RGCs was determined at 1, 2 & 3 days after transduction (d.a.t.) for rat & mice (b,e) & at 4 & 6 d.a.t. for zebrafish (h). Treatment effects compared to vehicle-treated controls: \*\*\*p < 0.001, \*\*p < 0.01, Scale bars: 50 µm, RGC survival (c.f.i) was not affected by baculovirus application (by) compared to vehicle-treated controls (-). (j) Delayed transduction of adult zebrafish RGCs with DsRed-by after 4 days in culture. Scale bar: 50 µm. (k,l) Co-transduction of adult rat RGCs with fGFP-bv & DsRed-bv. The two viruses were added to retinal cultures simultaneously at half the concentration of single transductions. Representative pictures show co-transduced, fGFP-& DsRed-expressing RGCs (green & red, respectively) that were costained against the neuronal marker BIII-tubulin (white) at 2 d.a.t. Scale bar: 50 µm. (k). The percentage of transduced RGCs was determined at 2 & 3 d.a.t. (I). Non-significant difference in co-tranduction efficiency compared to single transductions. Image collected & cropped by CiteAb from the following publication (https://www.nature.com/articles/srep38928), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Immunocytochemistry/ Immunofluorescence: GFP Antibody [NB100-1770] - Transduction of dorsal root ganglion neurons. Dissociated dorsal root ganglion neurons (DRG) isolated from postnatal (a–c) or adult (d–f) rats & adult mice (g–i) were transduced with fGFP-baculovirus (bv). Representative pictures of vehicle (-) & bv -treated cultures (a,d,g) show transduced, GFP-expressing neurons (green) that were co-stained with the neuronal marker βIII-tubulin (Tub, red) at 2 days after transduction (d.a.t.). The percentage of transduced, GFP-expressing DRG neurons was determined at 1, 2 & 3 d.a.t. (b,e,h), revealing similar transduction efficiencies of 80–90% in postnatal & adult DRG neurons. Cell survival in postnatal (c) & adult (f, i) DRG cultures was not affected by bvapplication compared to vehicle-treated controls (-). Scale bars: 100 μm. Image collected & cropped by CiteAb from the following publication (https://www.nature.com/articles/srep38928), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Immunocytochemistry/ Immunofluorescence: GFP Antibody [NB100-1770] - hnRNPA3 knockout enhances DNA damage in DPR expressing HeLa cells. a Immunocytochemical detection of yH2AX foci in GFPtagged DPR-expressing HeLa WT & A3KO HeLa cells. b Fold change of vH2AX foci in HeLa WT & A3KO cells expressing the indicated DPRs relative to HeLa WT cells with GFP expression. N = 47–127 cells from 2 biological replicates. c Immunocytochemical detection of pATM foci in GFP-tagged DPR-expressing HeLa WT & A3KO HeLa cells. d Fold change of pATM foci in HeLa WT & A3KO cells expressing the indicated DPRs relative to HeLa WT cells with GFP expression. N = 71–263 cells from 2 biological replicates. GFP EGFP transfected, GA EGFP-tagged poly-GA 175 repeats transfected, GR EGFP-tagged poly-GR 177 repeats transfected, PR: EGFP-tagged poly-PR 176 repeats transfected. All graphs are shown as mean ± SEM. \*p < 0.05, \*\*p < 0.01; one-way ANOVA & Tukey's post-hoc test. Scale bar 10 µm Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/31642962), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



**АЗКО** 

#### **Publications**

Nichol H, Amilhon B, Manseau F, Badrinarayanan S et Al. Electrophysiological and Morphological Characterization of Chrna2 Cells in the Subiculum and CA1 of the Hippocampus: An Optogenetic Investigation Front Cell Neurosci 2018-03-01 [PMID: 29487503]

Leibinger M, Hilla AM, Andreadaki A, Fischer D. et Al. GSK3-CRMP2 signaling mediates axonal regeneration induced by Pten knockout Commun Biol 2019-08-28 [PMID: 31453382]

Terheyden-Keighley D, Leibinger M, Zeitler C, Fischer D. et Al. Transneuronal Delivery of Cytokines to Stimulate Mammalian Spinal Cord Regeneration Methods Mol Biol 2023-03-07 [PMID: 36881297]

Leibinger M, Andreadaki A, Golla R, Levin E et Al. Boosting CNS axon regeneration by harnessing antagonistic effects of GSK3 activity Proc Natl Acad Sci U S A 2017-06-21 [PMID: 28630333]

Gobrecht P, Gebel J, Hilla A, Gisselmann G et Al. Targeting Vasohibins to Promote Axon Regeneration J Neurosci 2024-03-01 [PMID: 38429108]

Leibinger M, Zeitler C, Gobrecht P, Andreadaki A et Al. Transneuronal delivery of hyper-interleukin-6 enables functional recovery after severe spinal cord injury in mice Nat Commun 2021-01-16 [PMID: 33452250]

Cornebois A, Sorbara M, Cristol M, Vigne E et Al. Discovery of SOCS7 as a versatile E3 ligase for protein-based degraders iScience 2024-05-15 [PMID: 38746666]

McGinn TE, Galicia CA, Leoni DC, Partington N et Al. Rewiring the Regenerated Zebrafish Retina: Reemergence of Bipolar Neurons and Cone-Bipolar Circuitry Following an Inner Retinal Lesion Front Cell Dev Biol 2019-06-28 [PMID: 31245369]

Hilla AM, Baehr A, Leibinger M, Andreadaki A et Al. CXCR4/CXCL12-mediated entrapment of axons at the injury site compromises optic nerve regeneration Proc Natl Acad Sci U S A 2021-05-20 [PMID: 34011605]

Billipp TE, Fung C, Webeck LM, Sargent DB et Al. Tuft cell-derived acetylcholine promotes epithelial chloride secretion and intestinal helminth clearance Immunity 2024-05-14 [PMID: 38744291]

Whitworth GB, Watson FL., , et Al. Translating Ribosome Affinity Purification (TRAP) and Bioinformatic RNA-Seq Analysis in Post-metamorphic Xenopus laevis Methods Mol Biol 2023-03-07 [PMID: 36881307]

Zhang H, Pandey S, Travers M, Sun H et Al. Targeting CDK9 Reactivates Epigenetically Silenced Genes in Cancer Cell 2018-11-21 [PMID: 30454645]

More publications at <a href="http://www.novusbio.com/NB100-1770">http://www.novusbio.com/NB100-1770</a>





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General: novus@novusbio.com

#### 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 100% guarantee, please visit www.novusbio.com/guarantee

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