

Product Datasheet

GFP Antibody - BSA Free NB600-308

Unit Size: 0.1 mg

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

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NB600-308

GFP Antibody - BSA Free

Product Information	
Unit Size	0.1 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	<p>This 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-Rabbit Serum and purified and partially purified Green Fluorescent Protein (Aequorea victoria)</p> <p>GFP antibody is stable for several weeks at 4C as an undiluted liquid.</p>
Host	Rabbit
Species	Non-species specific
Reactivity Notes	<p>No reaction was observed against Human, Mouse serum proteins. Suitable for detecting fusion proteins containing the GFP sequence expressed in Human, Mouse, Rat, C. elegans, Drosophila and in vitro transcription/translation systems and transgenic animals. Known cross reactivity with wt and all variants such as rGFP, eGFP, S65T-GFP, RS-GFP, YFP and EGFP.

GFP Transgenic Rat reactivity reported in scientific literature (PMID:25724725).

Mouse reactivity reported in multiple pieces of scientific literature.

Transgenic C. elegans reactivity reported in scientific literature (PMID: 27110099).

Use in Mouse reported in scientific publication (PMID: 32765228).

Plant reactivity reported in scientific literature (PMID:32896843)</p>
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.

Product Application Details	
Applications	Western Blot, Immunohistochemistry-Paraffin, Dot Blot, ELISA, Electron Microscopy, Flow Cytometry, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunoprecipitation, Immunohistochemistry Free-Floating, Immunohistochemistry Whole-Mount, Knockdown Validated
Recommended Dilutions	Western Blot 1:500-1:5000, Flow Cytometry 1:10-1:1000, ELISA 1:20000-1:120000, Immunohistochemistry 1:200-1:3000, Immunocytochemistry/ Immunofluorescence 1:500-1:5000, Immunoprecipitation 1:10-1:500, Immunohistochemistry-Paraffin 1:10-1:500, Immunohistochemistry-Frozen 1:50-1:250, Dot Blot, Electron Microscopy 1:10-1:500, Immunohistochemistry Free-Floating, Immunohistochemistry Whole-Mount, Knockdown Validated



Application Notes

This product is designed to detect GFP and its variants. GFP antibody has been tested by western blot and ELISA. This product 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 antibodies. 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 conjugated 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 can detect 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. Optimal titers for applications should be determined by the researcher.

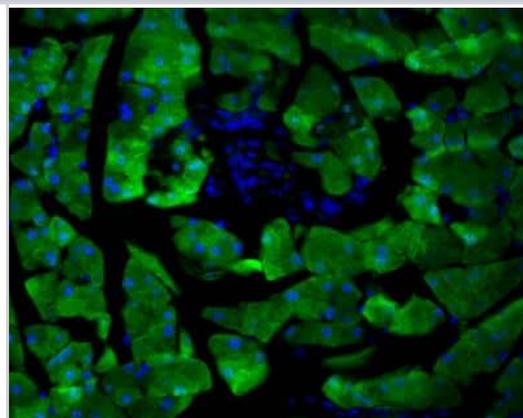
Use in Immunoprecipitation reported in scientific literature (PMID:34887587).

Use in DB reported in scientific literature (PMID:34242364).

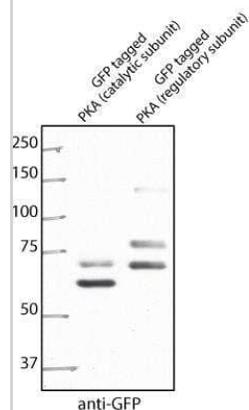
Use in Knockdown Validated reported in scientific literature (PMID: 32905777).

Images

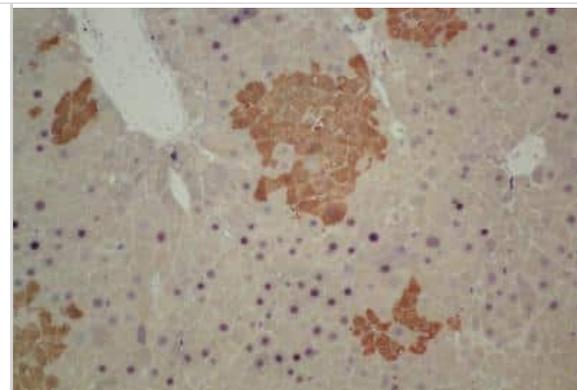
Analysis of GFP in transgenic mouse pancreas in OCT. The blue region in the center is an islet which does not express GFP and is negative. Image from verified customer review.



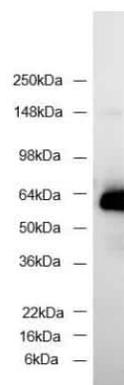
Analysis of GFP tagged PKA subunits over-expressed in HEK293T cells using GFP antibody. Image from verified customer review.



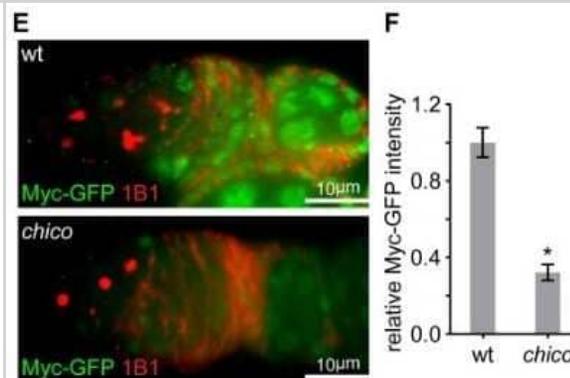
Staining of GFP+ hepatocytes (brown) transplanted into the liver of a recipient animal using anti-GFP antibody. Cyclin D1 double staining in blue. Image from verified customer review.



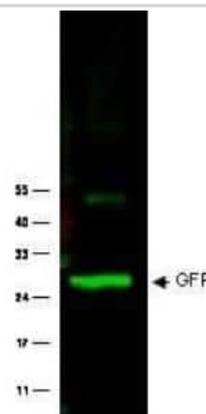
Total cell lysates (20ug) with protein of interest tagged with GFP was loaded. Blotted with GFP antibody [HRP] NB600-308H, without any secondary antibody. Image from verified customer review.



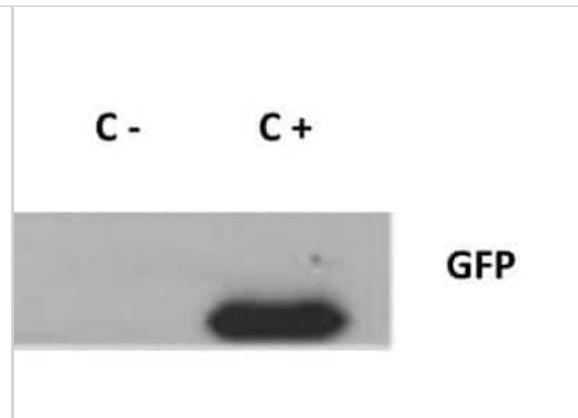
Germaria from wt or *chico1*/KG mutant ovaries endogenously expressing Myc-GFP stained with anti-GFP, anti-1B1, and DAPI. Scale bars, 10 μ m. (F) Quantification of relative Myc-GFP intensity in germarium region 2B from wt or *chico1*/KG mutant ovaries. $n = 10$ germaria for each genotype. Error bars represent SEM. $*p < 0.005$. Image collected and cropped by CiteAb from the following publication ([//pubmed.ncbi.nlm.nih.gov/31612862/](https://pubmed.ncbi.nlm.nih.gov/31612862/)) licensed under a CC-BY license.



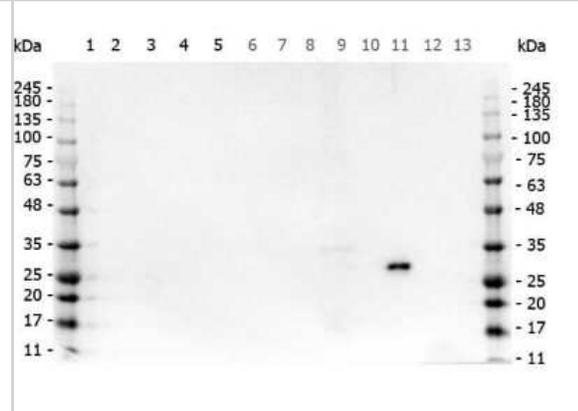
Western Blot of GFP antibody. Lane 1: Wild type GFP (0.1 ug) was used to spike HeLa whole cell lysate. Lane 2: none. Load: 30 ug per lane. Primary antibody: GFP antibody at 1:1000 for overnight at 4C. Secondary antibody: IRDye800(TM) Goat-a-Rabbit IgG [H&L] MX10 at 1:10,000 for 45 min at RT. Block: 5% BLOTTO in PBS overnight at 4C. Predicted/Observed size: 27 kDa for epitope tag GFP. Other band(s): none.



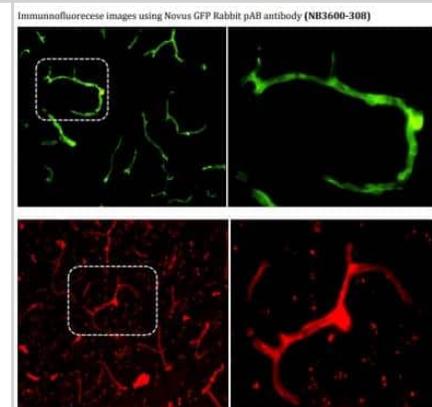
Western Blot of GFP antibody. Lane 1: 293FT cells transfected with CDK4 dominant negative (C-). Lane 2: 293FT cells positive control (C+). Load: 25 ug per lane. Primary antibody: GFP antibody at 1:400 for overnight at 4C. Secondary antibody: IRDye800(TM) rabbit secondary antibody at 1:10,000 for 45 min at RT. Block: 5% BLOTTO overnight at 4C. Predicted/Observed size: 27 kDa for GFP.



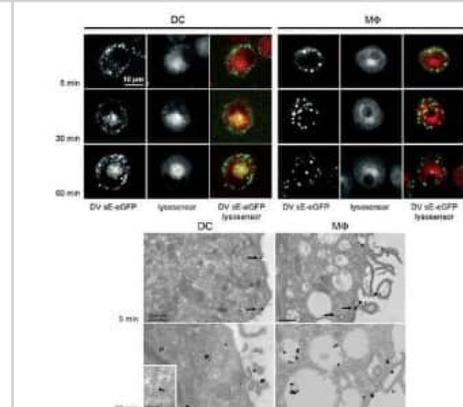
Western Blot of GFP antibody. Marker: Opal Pre-stained ladder. Lane 1: HEK293 lysate. Lane 2: HeLa Lysate. Lane 3: CHO/K1 Lysate. Lane 4: MDA-MB-231. Lane 5: A431 Lysate. Lane 6: Jurkat Lysate. Lane 7: NIH/3T3 Lysate. Lane 8: E-coli HCP Control. Lane 9: FLAG Positive Control Lysate. Lane 10: Red Fluorescent Protein. Lane 11: Green Fluorescent Protein. Lane 12: Glutathione-S-Transferase Protein. Lane 13: Maltose Binding Protein. Load: 10 ug of lysate or 50ng of purified protein per lane. Primary antibody: GFP antibody at 1ug/mL overnight at 4C. Secondary antibody: Peroxidase rabbit secondary antibody at 1:30,000 for 60 min at RT. Blocking Buffer: 1% Casein-TTBS for 30 min at RT. Predicted/Observed size: 30 kDa for GFP.



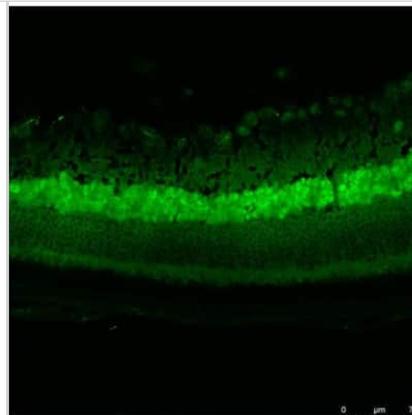
Adult mouse brain tissue. This image was submitted via customer review.



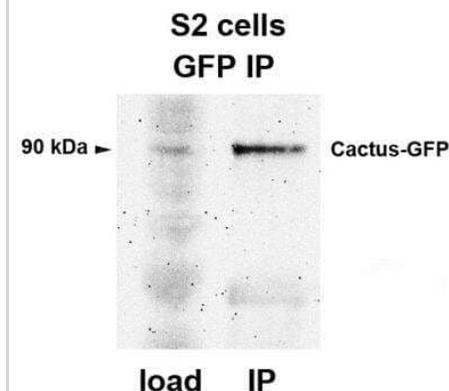
Immuno-microscopy of GFP antibody. Monocyte derived dendritic cells and dermal macrophages were challenged and directly visualized with eGFP labeled Dengue virus to localize sequestration of virus particles in the different cells (upper). The location of the GFP was confirmed by TEM (lower magnified view) using rabbit anti GFP Primary antibody (1:200) and a gold labeled secondary antibody.



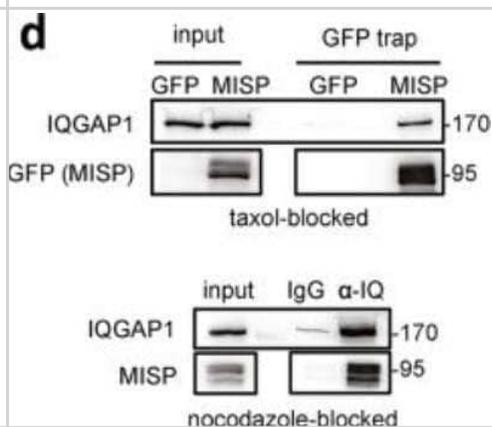
Analysis of GFP in mouse retinal cells. Image courtesy of anonymous customer review.



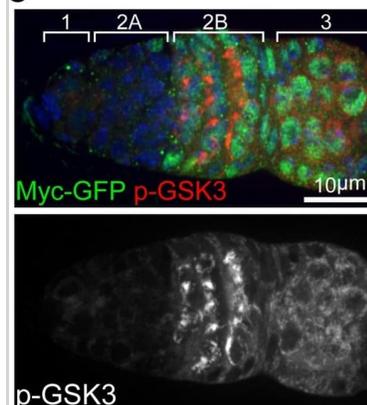
IP analysis of GFP in S2 Cells (Drosophila). Image from verified customer review.



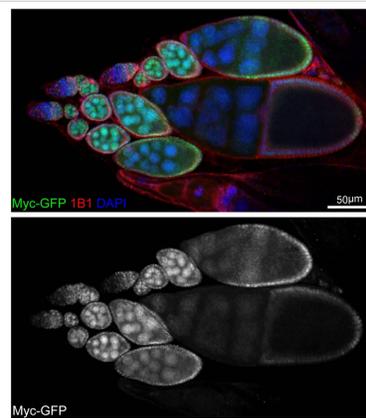
Immunoprecipitation experiments showing that GFP-MISP binds endogenous IQGAP1 (upper panel) and MISP binds to IQGAP1 endogenously using specific antibodies (lower panel) in mitotically arrested (taxol and nocodazole, respectively) HeLa cell lysates. Image collected and cropped by CiteAb from the following publication (nature.com/articles/s41598-018-24682-8), licensed under a CC-BY license.



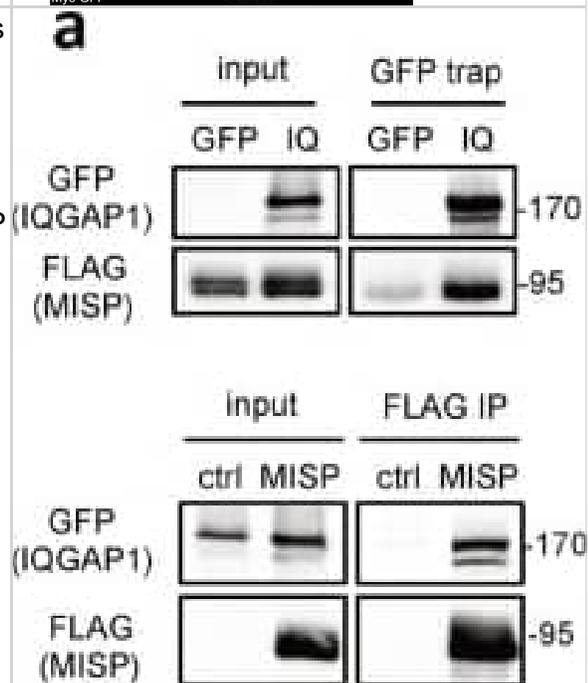
Immunocytochemistry/ Immunofluorescence: GFP Antibody [NB600-308] **C**
 - Myc is post-transcriptionally regulated by the IIS-GSK3 cascade. (A) Germaria from ovaries of ctrl, chicoRNAi, & bskRNAi driven by nos-Gal4 expressing LacZ under the control of myc endogenous promoter stained with anti-β-galactosidase & anti-1B1. Scale bars, 10 μm. (B) A representative live imaging of germaria from flies expressing GFP-tagged Sgg under its endogenous promoter. Scale bar, 10 μm. (C) A germarium from wt flies endogenously expressing Myc-GFP stained with anti-GFP (green) & anti-p-GSK3 (red). Germarium regions are indicated. Myc protein highly corresponds to inhibited form of GSK3. Scale bar, 10 μm. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/31612862>), licensed under a CC0-1.0 license. Not internally tested by Novus Biologicals.



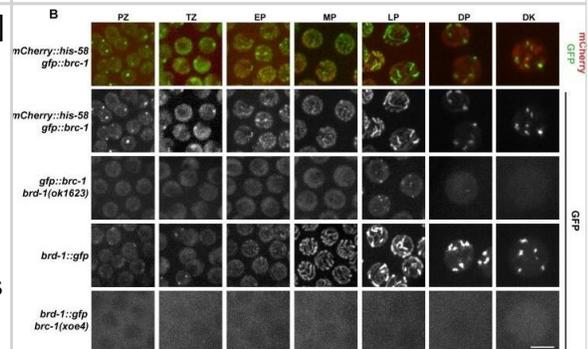
Immunocytochemistry/ Immunofluorescence: GFP Antibody [NB600-308]
 - Myc protein pattern in the ovary. (A) A representative low-magnification image of ovarioles from flies endogenously expressing Myc-GFP stained with anti-GFP, anti-1B1, & DAPI. Note that Myc level is low in early germarium stages, becomes high from germarium region 2B, & reduces from the stage-10 egg chamber. Scale bar, 50 μ m. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/31612862>), licensed under a CC0-1.0 license. Not internally tested by Novus Biologicals.



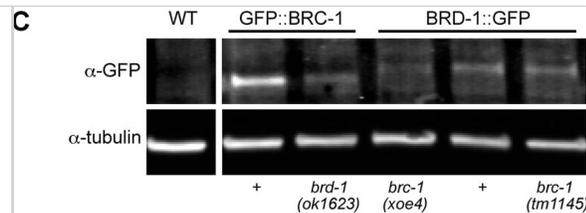
Western Blot: GFP Antibody [NB600-308] - MISP interacts & co-localizes with IQGAP1. (a) HEK293T cells transiently overexpressing GFP-IQGAP1 & FLAG-MISP were used for co-immunoprecipitation experiments from both sides (GFP trap or FLAG M2 beads). (b) Endogenous immunoprecipitation using IQGAP1 antibody or control immunoglobulin (IgG) was carried out in HeLa cell lysates & MISP was detected in the eluate. (c) In vitro interaction between purified MBP-MISP & GST-IQGAP1 was detected by GST & MBP pull-down experiments. (d) Immunoprecipitation experiments showing that GFP-MISP binds endogenous IQGAP1 (upper panel) & MISP binds to IQGAP1 endogenously using specific antibodies (lower panel) in mitotically arrested (taxol & nocodazole, respectively) HeLa cell lysates. (a–d) Images were gained from the same Western blot membrane after cutting/cropping & presented with different exposure times or contrast enhancement for better presentation purposes. The dividing lane marks the grouping of images of the same (or different) membrane. Uncropped blots, where applicable, are included in Supplementary Fig. 4. (e) HeLa cells were immunostained for MISP & IQGAP1 & co-localization was visualized in mitosis in single-plane confocal images, scale bar: 5 μ m. Lower pictures: After manual thresholding white spots mark the co-localizing areas in the merge images. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/29679050>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



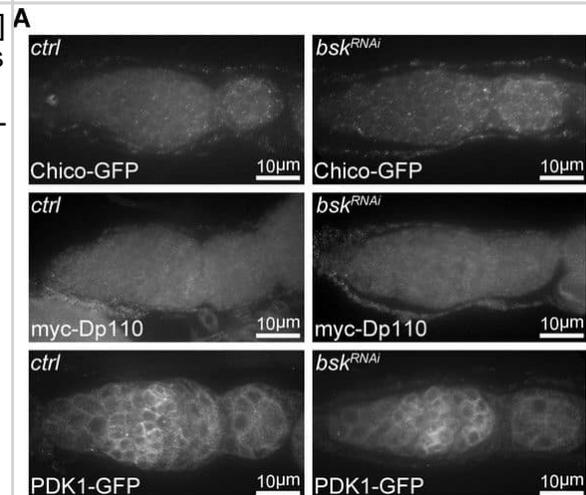
Immunocytochemistry/ Immunofluorescence: GFP Antibody [NB600-308]
 - BRC-1 & BRD-1 are inter-dependent for localization. (A) Co-localization between BRD-1::GFP (green) & TagRFP-T::BRC-1 (red) at late pachytene in live worms. Scale bar = 10 μ m. (B) Stills of germline nuclei from live worms expressing GFP::BRC-1 & mCherry::Histone H2B (mCherry::his-58; gfp::brc-1); merge & GFP channel; top two panels, respectively. GFP::BRC-1 expression in *brd-1(ok1623)* mutant at indicated meiotic stages. Bottom two panels show BRD-1::GFP localization in wild type & the *brc-1(xoe4)* mutant. Images are projections through half of the gonad. TZ = transition zone; EP = early pachytene; MP = mid pachytene; LP = late pachytene; DP = diplotene; DK = diakinesis. Scale bar = 5 μ m. (C) Immunoblot of whole worm extracts from indicated worms probed with anti-GFP & α -tubulin antibodies. Lane 1 = N2: wild type; Lane 2 = JEL515: gfp::brc-1; Lane 3 = JEL520: gfp::brc-1 *brd-1(ok1623)*; Lane 4 = JEL744: *brc-1(xoe4)* *brd-1::gfp*; Lane 5 = JEL657: *brd-1::gfp*; Lane 6 = JEL678: *brc-1(tm1145)* *brd-1::gfp*. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/30383767>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



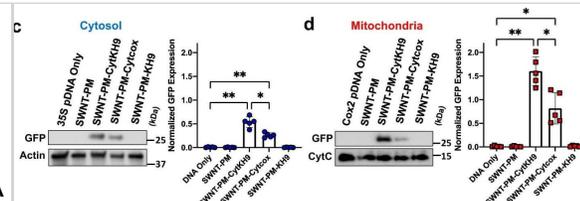
Western Blot: GFP Antibody [NB600-308] - BRC-1 & BRD-1 are inter-dependent for localization. A) Co-localization between BRD-1::GFP (green) & TagRFP-T::BRC-1 (red) at late pachytene in live worms. Scale bar = 10 μ m. B) Stills of germline nuclei from live worms expressing GFP::BRC-1 & mCherry::Histone H2B (mCherry::his-58; gfp::brc-1); merge & GFP channel; top two panels, respectively. GFP::BRC-1 expression in *brd-1(ok1623)* mutant at indicated meiotic stages. Bottom two panels show BRD-1::GFP localization in wild type & the *brc-1(xoe4)* mutant. Images are projections through half of the gonad. TZ = transition zone; EP = early pachytene; MP = mid pachytene; LP = late pachytene; DP = diplotene; DK = diakinesis. Scale bar = 5 μ m. C) Immunoblot of whole worm extracts from indicated worms probed with anti-GFP & α -tubulin antibodies. Lane 1 = N2: wild type; Lane 2 = JEL515: gfp::brc-1; Lane 3 = JEL520: gfp::brc-1 *brd-1(ok1623)*; Lane 4 = JEL744: *brc-1(xoe4)* *brd-1::gfp*; Lane 5 = JEL657: *brd-1::gfp*; Lane 6 = JEL678: *brc-1(tm1145)* *brd-1::gfp*. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/30383767>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



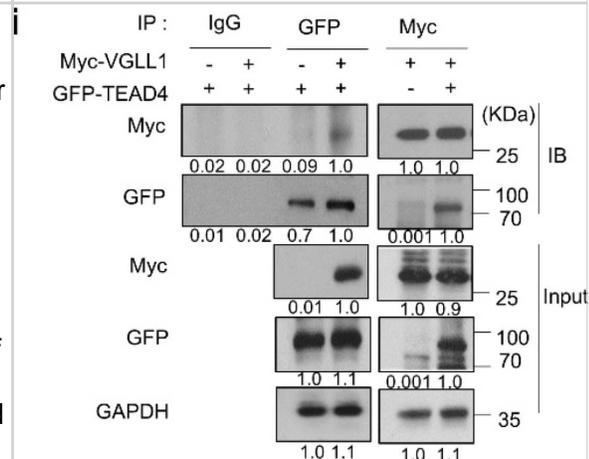
Immunocytochemistry/ Immunofluorescence: GFP Antibody [NB600-308] - Other IIS components are not altered by *bsk* RNAi. (A) Confocal images for germaria expressing Chico-GFP, myc-Dp110 or PDK1-GFP in the background of *ctrl* or *bsk*RNAi from ovaries stained with anti-GFP or anti-myc. Scale bars, 10 μ m. (B) Ovaries expressing LacZ driven by the *puc* promoter stained with anti- β -galactosidase, anti-1B1, & DAPI. Note that JNK activity is moderately induced in late germarium & decreased in the following stages, while JNK activity is much stronger in follicle cells of a maturing egg. Scale bars, 20 μ m. (C) Germaria from ovaries of *ctrl* & *bsk*RNAi driven by *nos*-Gal4 stained with anti-AKT & anti-1B1. Scale bars, 10 μ m. (D) Quantification of AKT intensity from cysts in germarium region 2A & 2B of ovaries with *ctrl* & *bsk*RNAi driven by *nos*-Gal4. Intensities are normalized to the value of *ctrl* at region 2B. $n = 7$ & 8 germaria for *ctrl* & *bsk*RNAi, respectively. Error bars represent SEM. $*p < 0.005$. (E) Visualization of the *InR* mRNA by FISH in germaria from ovaries of *ctrl* & *bsk*RNAi driven by *nos*-Gal4. Note that *InR* mRNA level is decreased by *bsk* RNAi. Germaria are outlined with dotted lines. Scale bars, 10 μ m. (F) Quantification of *InR* mRNA density in region 2A & 2B germ cells from ovaries with indicated genotypes. $n = 9$ germaria for each genotype. Error bars represent SEM. $*p < 0.005$. (G) Quantification of relative Myc-GFP intensity in region 2B cysts of ovaries illustrated in Figure 5H. $n = 10$ germaria for each genotype. Error bars represent SEM. $*p < 0.005$. 10.7554/eLife.49309.017Figure 5—figure supplement 1—source data 1. Relative AKT intensity, *InR* mRNA density, & Myc-GFP intensity in the germarium. Relative AKT intensity, *InR* mRNA density, & Myc-GFP intensity in the germarium. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/31612862>), licensed under a CC0-1.0 license. Not internally tested by Novus Biologicals.



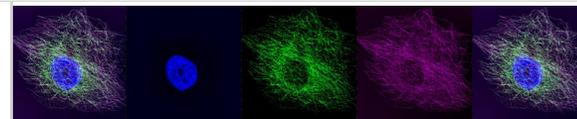
Western Blot: GFP Antibody [NB600-308] - GFP Expression within *A. thaliana* upon infiltration with SWNT-PM-CytKH9/pDNA complexes. a Reporter construct design of transient GFP expression for the nucleus & mitochondria. b Representative confocal laser scanning microscopy images of *A. thaliana* root cells from a minimum of five seedlings for each condition (n = 5) 18 h post infiltration with SWNT-PM-CytKH9/pDNA complexes containing pDONR-35S-GFP or pDONR-Cox2-GFP reporter constructs for nuclear or mitochondrial expression, respectively. Mitochondria were stained with MitoTracker Red (CMXRos), & colocalization analysis was performed on GFP expression & MitoTracker signals. Scale bars represent 20 μ m. c, d Representative western blots & their quantification from cytosolic (c) & mitochondrial (d) fractions isolated from approximately 60 seedlings 18 h post infiltration with SWNT NCs probed for GFP expression. Actin & cytochrome c were used as loading controls for cytosolic & mitochondrial proteins, respectively. Data points from five biological replicates (n = 5) are represented as the mean \pm standard deviation. Statistical significance was determined by Brown-Forsythe & Welch one-way ANOVA test. For the cytosolic samples, P-values are 0.0027, & 0.0020 between DNA only & SWNT-PM-CytKH9, SWNT-PM-Cytcox, respectively, & 0.0253 between SWNT-PM-CytKH9 & SWNT-PM-Cytcox. For the mitochondrial samples, P-values are 0.0019, & 0.0333 between DNA only & SWNT-PM-CytKH9, & SWNT-PM-Cytcox, respectively, & 0.0359 between SWNT-PM-CytKH9 & SWNT-PM-Cytcox. ns – not statistically significant, *P < 0.05, **P < 0.01. Source data are provided as a Source Data file. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/35577779>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Western Blot: GFP Antibody [NB600-308] - VGLL1 regulates MMP9 transcription in gastric cancer cells. (a,b) Viability (a) & invasion (b) assay of NUGC3 cells. Cells were treated with 20 nM siScramble (SC) or siMMP9 for 48 h, & then stained with sulforhodamine B. Data are presented as mean \pm SD. n = 3; * p < 0.05 (Student's t-test). (c) MMP9 mRNA expression regulation by VGLL1 in gastric cancer cells treated with siVGLL1 or siSC (control) was measured by qPCR. (d) Changes in MMP9 expression upon knockdown or overexpression of VGLL1 assayed by western blotting in NUGC3 cells. (e) IHC of VGLL1 & MMP9 expression in the liver & lungs of an in vivo metastasis mouse model using surgical resection of tumors. Scale bar, 200 μ m. (f) Construction of various luciferase reporter systems under control of the MMP9 promoter. (g) MMP9 promoter activities of the reporter systems containing modified TEA-binding sites were measured in NUGC3 cells. n = 3; * p < 0.05 (Student's t-test). (h) Effect of TEAD4 on VGLL1-regulated MMP9 transcriptional activity. NUGC3 cells treated with siSC or siTEAD4 for 24 h were transfected with MMP9-luc, Renilla-luc, pCDNA3.1, & pCDNA3.1-myc-VGLL1 vectors for 48 h. n = 3; * p < 0.05 (Student's t-test). (i) Interaction between VGLL1 & TEAD4. Lysates of NUGC3 cells that were transfected with pCDNA3.1-myc-VGLL1 & pEGFP-N1-TEAD4 were immunoprecipitated using anti-IgG, anti-GFP, & anti-Myc antibodies. Protein expression was analyzed by immunoblotting. (j) ChIP assays while using nuclear extracts of NUGC3 cells treated with siTEAD4. The ChIP-enriched DNA was subjected to PCR. (k) Target genes of VGLL1 & YAP. NUGC3 cells were transfected with pcDNA3, pcDNA3-myc-VGLL1, or pcDNA3-myc-YAP, & the mRNA expression levels were analyzed by RT-PCR. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/31816819>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Immunocytochemistry/Immunofluorescence: Rabbit GFP pAb [NB600-308] - Analysis of GFP in human Hs746T gastric cancer cell line. Image from a verified customer review.



Publications

Sidorov MS, Kim H, Rougie M et al. Visual Sequences Drive Experience-Dependent Plasticity in Mouse Anterior Cingulate Cortex Cell Reports 2020-09-15 [PMID: 32937128]

McNally KP, Beath EA, Danlasky BM et al. Cohesin is required for meiotic spindle assembly independent of its role in cohesion in *C. elegans* PLOS Genetics 2022-10-24 [PMID: 36279281]

Yamada S, Mizukoshi T, Tokunaga A, Sakakibara SI. Inka2, a novel Pak4 inhibitor, regulates actin dynamics in neuronal development PLOS Genetics 2022-10-27 [PMID: 36301793]

Kim H, Gao EB, Draper A et al. Rescue of behavioral and electrophysiological phenotypes in a Pitt-Hopkins syndrome mouse model by genetic restoration of Tcf4 expression eLife 2022-05-10 [PMID: 35535852]

Kong JH, Young CB, Pusapati GV et al. A Membrane-Tethered Ubiquitination Pathway Regulates Hedgehog Signaling and Heart Development Developmental Cell 2020-11-01 [PMID: 32966817]

Toth BA, Chang KS, Fechtali S, Burgess CR. Dopamine release in the nucleus accumbens promotes REM sleep and cataplexy iScience 2023-09-15 [PMID: 37664637]

Huang YH, Chen CW, Sundaramurthy V et al. Systematic Profiling of DNMT3A Variants Reveals Protein Instability Mediated by the DCAF8 E3 Ubiquitin Ligase Adaptor Cancer Discovery 2022-01-01 [PMID: 34429321]

Toth BA, Chang KS, Burgess CR. Striatal dopamine regulates sleep states and narcolepsy-cataplexy bioRxiv 2023-06-03 [PMID: 37397994]

Ansell TB, Corey RA, Viti LV et al. The energetics and ion coupling of cholesterol transport through Patched1 Science Advances 2023-08-25 [PMID: 37611095]

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