abcam

Product datasheet

Anti-GFP antibody ab13970

★★★★★ 93 Abreviews 3182 References 8 Images

Overview

Product name Anti-GFP antibody

Description Chicken polyclonal to GFP

Host species Chicken

Specificity Our GFP antibody does cross-react with the many fluorescent proteins that are derived from the

jellyfish Aequorea victoria. These are all proteins that differ from the original GFP by just a few

point mutations (EGFP, YFP, mVenus, CFP, BFP etc.).

Tested applications Suitable for: WB, ICC/IF

Species reactivity Reacts with: Species independent

Immunogen Recombinant full length protein corresponding to GFP.

Database link: P42212

Positive control ICC: GFP-transfected NIH/3T3 (Mouse embryo fibroblast cell line). WB: Transgenic mouse spinal

cords.

General notes

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

Storage buffer pH: 7

Preservative: 0.01% Thimerosal (merthiolate)

Constituents: PBS, 50% Glycerol, 0.16% Sodium phosphate

Purity lgY fraction

Purification notes Sterile filtered.

Clonality Polyclonal

1

Isotype IgY

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab13970 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	★★★★★ (13)	1/5000.
ICC/IF	★★★★★ (22)	1/2000. Used at a dilution of 1/2000 for 1 hr (see Abreview for further information).

Target

Relevance

Function: Energy-transfer acceptor. Its role is to transduce the blue chemiluminescence of the protein aequorin into green fluorescent light by energy transfer. Fluoresces in vivo upon receiving energy from the Ca²⁺ -activated photoprotein aequorin.

Subunit structure: Monomer.

Tissue specificity: Photocytes.

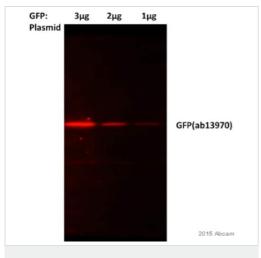
Post-translational modification: Contains a chromophore consisting of modified amino acid residues. The chromophore is formed by autocatalytic backbone condensation between Ser-65 and Gly-67, and oxidation of Tyr-66 to didehydrotyrosine. Maturation of the chromophore requires nothing other than molecular oxygen.

Biotechnological use: Green fluorescent protein has been engineered to produce a vast number of variously colored mutants, fusion proteins, and biosensors. Fluorescent proteins and its mutated allelic forms, blue, cyan and yellow have become a useful and ubiquitous tool for making chimeric proteins, where they function as a fluorescent protein tag. Typically they tolerate N- and C-terminal fusion to a broad variety of proteins. They have been expressed in most known cell types and are used as a noninvasive fluorescent marker in living cells and organisms. They enable a wide range of applications where they have functioned as a cell lineage tracer, reporter of gene expression, or as a measure of protein-protein interactions. Can also be used as a molecular thermometer, allowing accurate temperature measurements in fluids. The measurement process relies on the detection of the blinking of GFP using fluorescence correlation spectroscopy.

Sequence similarities: Belongs to the GFP family.

Biophysicochemical properties: Absorption: Abs(max)=395 nm

Exhibits a smaller absorbance peak at 470 nm. The fluorescence emission spectrum peaks at 509 nm with a shoulder at 540 nm.



Western blot - Anti-GFP antibody (ab13970)

This image is courtesy of an anonymous Abreview

All lanes : Anti-GFP antibody (ab13970) at 1/2000 dilution (Diluent 1x TBS /4 hours at 4° C)

Lane 1 : 3 μg of GFP plasmid overexpressed in mouse cardiomyocytes whole cell lysate with BSA / for 1 hour at room temperature

Lane 2 : 2 μg of GFP plasmid overexpressed in mouse cardiomyocytes whole cell lysate with BSA / for 1 hour at room temperature

Lane 3 : 1 μg of GFP plasmid overexpressed in mouse cardiomyocytes whole cell lysate with BSA / for 1 hour at room temperature

Lysates/proteins at 25 µg per lane.

Blocking peptides at 5 % per lane.

Secondary

All lanes : Goat Anti-Chicken IgY H&L (Alexa Fluor® 594) preadsorbed (ab150176) at 1/5000 dilution

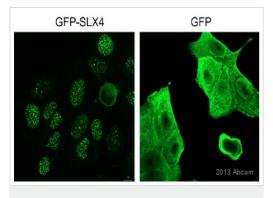
Performed under reducing conditions.

Additional bands at: 25 kDa. We are unsure as to the identity of these extra bands.

Exposure time: 30 seconds

Gel Running Conditions: Reduced Denaturing (15% PAGE)

Detection method: Fluorescent Secondary Antibodies

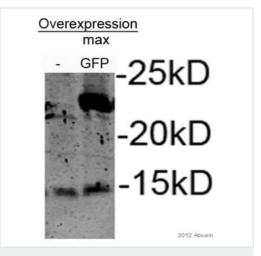


Immunocytochemistry/ Immunofluorescence - Anti-GFP antibody (ab13970)

This image is courtesy of an Abreview submitted by Christophe Lachaud

ab13970 staining GFP in U-2 OS (Human bone osteosarcoma epithelial cell line) cells by ICC/IF.

Cells were paraformaldehyde fixed, permeabilized with 0.5% triton and blocked with 2% antibody dilution buffer for 2 hours. Cells were incubated with the primary antibody (1/1000) for 1 hour at 25°C. An undiluted Alexa Fluor[®] 488 conjugated Goat anti-chicken polyclonal was used as the secondary antibody.



Western blot - Anti-GFP antibody (ab13970)

Image courtesy of an anonymous Abreview.

All lanes: Anti-GFP antibody (ab13970) at 1/1000 dilution

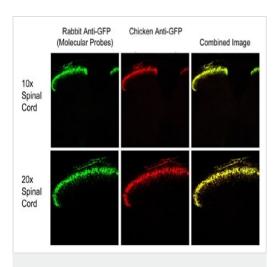
Lane 1 : Whole cell lysate prepared from HeLa (Human epithelial cell line from cervix adenocarcinoma) cells.

Lane 2: Whole cell lysate prepared from HeLa cells.

Lysates/proteins at 25 µg per lane.

Secondary

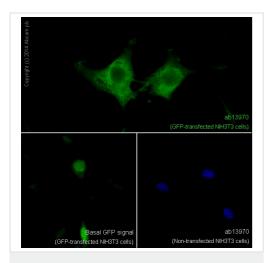
All lanes : IRDye 800CW conjugated goat anti-chicken polyclonal at 1/15000 dilution



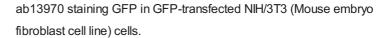
Immunocytochemistry/ Immunofluorescence - Anti-GFP antibody (ab13970)

Transgenic mice expressing GFP selectively in lamina II of the spinal cord.

In the right panels, note the correspondance between the green (rabbit anti-GFP) and red signals (chicken anti-GFP from Abcam) indicating that these two antibody preparations recognized the same gene product. The secondary antibody used with ab13970 was an FITC-labeled goat anti-chicken



Immunocytochemistry/ Immunofluorescence - Anti-GFP antibody (ab13970)



The cells were fixed with 4% formaldehyde (10 minutes) and then blocked in 1% BSA / 0.3M glycine in 0.1%PBS-Tween for 1 hour. The cells were then incubated with ab13970 at 1/2000 dilution overnight at +4°C followed by incubation with Goat Anti-Chicken lgY H&L (Alexa Fluor $^{\tiny (8)}$ 488) preadsorbed (ab150173), for 1 hour, at 1 $\mu g/ml$.

Under identical experimental conditions, when compared to the basal level of GFP expression in transfected NIH/3T3 cells, the cells upon which ab13970 was applied gave a stronger signal in the 488 channel, indicating that ab13970 is binding to GFP and therefore eliciting signal amplification.

ab13970 was also applied to non-GFP-transfected NIH/3T3 cells, which produced no positive staining, indicating specificity for GFP. Nuclear DNA was labeled with 1.43 μ M DAPI (blue).

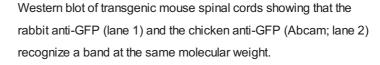


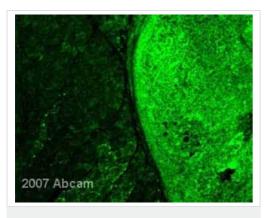
Western blot - Anti-GFP antibody (ab13970)

Lane 1: Rabbit anti-GFP

Lane 2: Anti-GFP antibody (ab13970)

All lanes: Transgenic mouse spinal cords



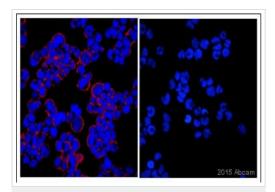


Immunocytochemistry/ Immunofluorescence - Anti-GFP antibody (ab13970)

This image is courtesy of an Abreview submitted by Dr Radbod Darabi

ab13970 staining GFP + tumor in mouse muscle cells by ICC/IF.

Cells were formaldehyde fixed and blocked with 3% BSA for 1 hour at 24°C prior to incubation with the primary antibody (1/500) for 1 hour at 24°C. An Alexa Fluor[®] 488 conjugated goat anti-chicken was used as the secondary.



Immunocytochemistry/ Immunofluorescence - Anti-GFP antibody (ab13970)

Image courtesy of an AbReview submitted by Dr Francois Daubeuf.

Immunocytochemical/ Immunoflurescence analysis of cytospined HEK-293 cells (Human epithelial cell line from embryonic kidney) transfected with GFP, labeling GFP with ab13970 at 1/200 incubated for 16 hours at 4°C with 1% BSA in PBS. Secondary used was a donkey anti-chicken polyclonal DyLight[®] 594 at 1/500.

GFP is shown in red (DyLight® 594).

Nuclei are counterstained in blue (DAPI).

The left panel shows HEK-293 cells transfected with GFP and the right panel shows non-transfected HEK-293 cells.

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