abcam

Product datasheet

FITC Anti-GFP antibody ab6662

15 Abreviews 171 References 5 Images

Overview

Product name	FITC Anti-GFP antibody
Description	FITC Goat polyclonal to GFP
Host species	Goat
Conjugation	FITC. Ex: 493nm, Em: 528nm
Tested applications	**Suitable for:** IHC-FoFr, IHC-Fr, WB, ICC/IF
Species reactivity	**Reacts with:** Species independent
Immunogen	Recombinant full length protein corresponding to GFP aa 1-246. Sequence: MSKGEELFTGVVPILVELDGDVNGHKFSVSGEGEGDATY GKLTLKFICTT GKLPVPWPTLVTTFSYGVQCFSRYPDHMKQHDFFKSAM PEGYVQERTIFF KDDGNYKTRAEVKFEGDTLVNRIELKGIDFKEDGNILGHKL EYNYNSHNV YIMADKQKNGIKVNFKIRHNIEDGSVQLADHYQQNTPIGDG PVLLPDNHY LSTQSALSKDPNEKRDHMVLLEFVTAAGITHGMDELYK Database link: **P42212** Run BLAST with Run BLAST with
Positive control	WB: Recombinant A. victoria GFP protein.
General notes	Designed to detect GFP and its variants in ELISA (sandwich or capture), immunoblotting and immunoprecipitation. Fluorescein conjugated anti-GFP was assayed by immunofluorescence microscopy on prokaryotic (*E.coli*) and eukaryotic (CHO cells) expression systems and was shown to detect GFP containing inserts. Significant amplification of signal was detected using fluorochrome conjugated anti-GFP relative to the fluorescence of GFP alone. In case of unexpected background, use pre-adsorbed secondary antibodies. Fluorescein isothiocyanate (FITC) (MW 390 daltons) Absorption Wavelength: 495 nm Emission Wavelength: 528 nm Fluorochrome/Protein Ratio: 3.5 moles FITC per mole of Goat IgG 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). Store at -20°C. Avoid freeze / thaw cycle.
Storage buffer	pH: 6.50 Preservative: 0.01% Sodium azide Constituents: 0.42% Potassium phosphate, 0.87% Sodium chloride, 1% BSA BSA Immunoglobulin and Protease free
Purity	Protein G purified
Purification notes	GFP Fluorescein Conjugated 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.
Primary antibody notes	Designed to detect GFP and its variants in ELISA (sandwich or capture), immunoblotting and immunoprecipitation. Fluorescein conjugated anti-GFP was assayed by immunofluorescence microscopy on prokaryotic (*E.coli*) and eukaryotic (CHO cells) expression systems and was shown to detect GFP containing inserts. Significant amplification of signal was detected using fluorochrome conjugated anti-GFP relative to the fluorescence of GFP alone.
Clonality	Polyclonal
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab6662 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
IHC-FoFr	★★★★★ (1)	1/250.
IHC-Fr	★★★★★ (5)	Use at an assay dependent concentration.
WB	★★★☆☆ (1)	1/10000.
ICC/IF	★★★★☆ (4)	1/500 - 1/2500.

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 Ca^{2+} -activated photoprotein aequorin.

Subunit structure: Monomer.

Tissue specificity: Photocytes.

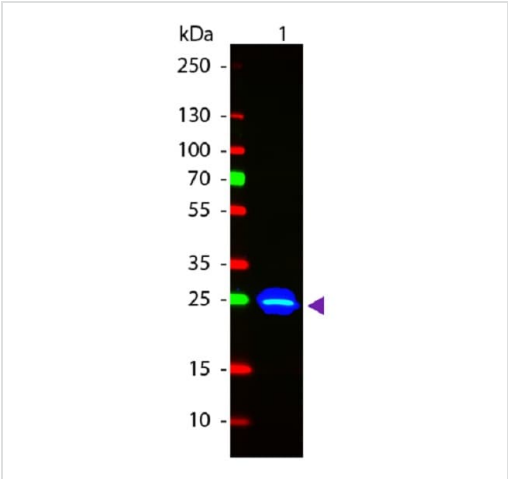
Post-translational modification: Contains a chromophore consisting of modified amino acid residues. The chromophore is formed by autocatalytic backbone condensation between Ser-65 and Gly-67, and oxidation of Tyr-66 to didehydrotyrosine. Maturation of the chromophore requires nothing other than molecular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

Images



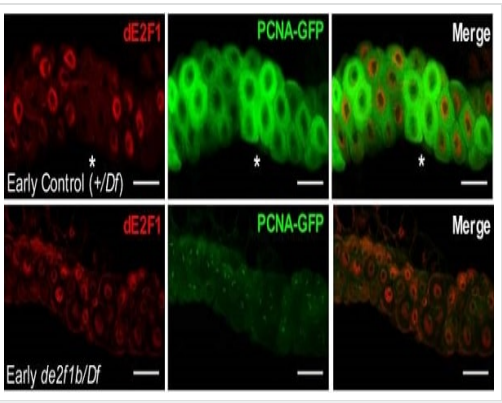
Western blot - FITC Anti-GFP antibody (ab6662)

FITC Anti-GFP antibody (ab6662) + Recombinant A. victoria GFP protein (**ab84191**)

Secondary

Fluorescein goat secondary antibody for 60 minutes at RT at 1/1000 dilution

Block: Blocking buffer for 30 minutes at RT.



Immunohistochemistry (Frozen sections) - FITC

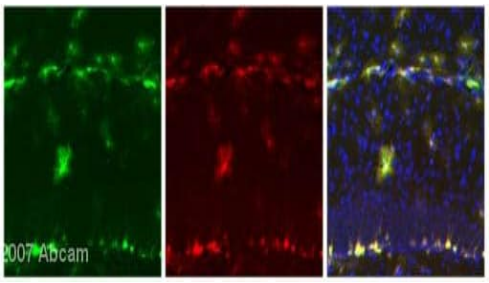
Anti-GFP antibody (ab6662)

Kim at el PLoS Genet. 2018 Feb 8;14(2):e1007204. doi: 10.1371/journal.pgen.1007204. eCollection 2018 Feb. Fig 4. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Oscillation of Cyclin E and E2F target gene expression is deregulated in *de2f1b* mutant salivary glands.

Salivary glands of control and *de2f1b* mutant early (80–85 hr AEL) third instar larvae expressing PCNA-GFP (green, ab6662) are stained with anti-dE2F1 (red). The region where high PCNA-GFP is observed with low dE2F1 is marked by an asterisk.

For Immunostaining, third instar imaginal discs and salivary glands were dissected in PBS and immediately fixed in 4% formaldehyde in PBS for 20 minutes at room temperature with the exception of tissues subjected to anti-dE2F1 staining that were fixed for 30 minutes on ice. Fixed tissues were then washed with 0.3% PBST (0.3% TritonX-100 in 1XPBS) and 0.1% PBST (0.1% TritonX-100 in 1XPBS). Samples were incubated with appropriate amount of primary antibody in 0.1% PBST and 1%BSA overnight. Samples were then washed with 0.1% PBST, incubated in secondary antibody in 0.1% PBST and 1% BSA for 2 hours, followed by several washes in 0.1% PBST prior to mounting.



Immunohistochemistry (Frozen sections) - FITC

Anti-GFP antibody (ab6662)

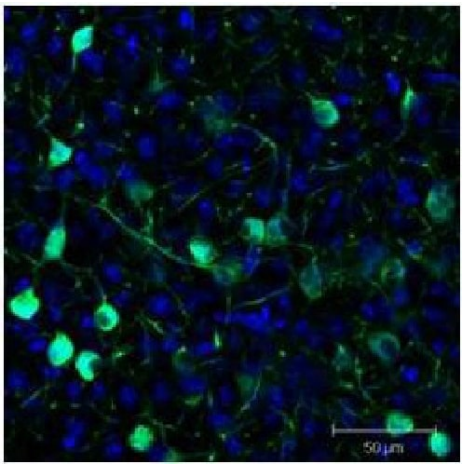
This image is courtesy of an Abreview submitted by Dr Joshua Breunig

ab6662 staining mouse brain tissue sections (inducible GFP reporter) by IHC-Fr.

The tissue was paraformaldehyde fixed and blocked with serum and then incubated with the antibody at a 1/1000 dilution for 1 hour.

Staining is shown in the left hand panel. The middle panel shows staining with a rabbit anti-GFP antibody and the right hand panel shows the merged images (plus DAPI).

ab6662 gives no noticable background and it is found that when viewing on an epifluorescent the exposure time is significantly reduced.



Immunohistochemistry (Frozen sections) - FITC Anti-GFP antibody (ab6662)

Immunofluorescence Microscopy using ab6662.

Tissue: Sf-1:Cre mice crossed to the Z/EG reporter line. Mouse brain (coronal view, 20X magnification).

Fixation: 4% PFA/PBS with o/n fixation, and subsequently transferred to a 30% sucrose solution.

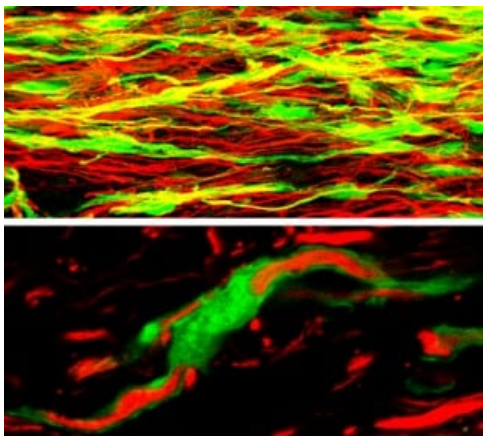
Antigen retrieval: Frozen in OCT freezing medium (Sakura) and cryostat sectioned at 40 microns.

Goat anti-GFP was used at 1/500 dilution in free floating imunnohistochemistry to detect GFP.

Secondary antibody: Fluorchrome conjugated Anti-goat IgG secondary antibody was used for detection at 1:500 at 1/10,000 for 45 minutes 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.



Immunohistochemistry (Frozen sections) - FITC Anti-GFP antibody (ab6662)

These pictures show confocal immunofluorescence using GFP-expressing glial cells (green) transplanted into the lesioned rat spinal cord.

Detected using ab6662 and a standard FITC filter set. Axons are labeled red by an antibody to neurofilament-200 and a rhodamine secondary antibody. The upper panel shows the centre of the transplant site at low power. Numerous GFP-positive cells can be seen mingling with axons. The lower panel shows, at high power in a single optical section, how ab6662 reveals the morphology of the transplanted cells to such an extent that their close interactions with axons are obvious - the cell depicted can be seen wrapping around a neurofilament-200 positive axon.

These images were kindly supplied as part of the review submitted by Andrew Toft.

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