

Product datasheet

Sepharose Anti-GFP antibody ab69314

★★★★★ 1 Abreviews 25 References 1 Image

Overview

Product name	Sepharose Anti-GFP antibody
Description	Sepharose Rabbit polyclonal to GFP
Host species	Rabbit
Conjugation	Sepharose
Specificity	This product is a suspension of affinity purified anti-GFP antibody (ab6556) covalently linked to sepharose beads. It is reactive against all variants of <i>Aequorea victoria</i> GFP such as S65T-GFP, RS-GFP, YFP, CFP, RFP, and EGFP. The unit sold contains 25µg of affinity purified rabbit anti-GFP IgG cross-linked to 125 µl sepharose beads in a total volume of 250 µl buffer. The product is supplied as a 50% slurry to facilitate pipetting. Pipetting the slurry is facilitated when pipet tips are blunted by cutting 3-5mm off from the tip.
Tested applications	Suitable for: IP
Species reactivity	Reacts with: Species independent
Immunogen	Recombinant full length protein corresponding to GFP. Database link: P42212
Positive control	Pure GFP protein, or cells known to overexpress GFP.
General notes	<p>This product should be kept refrigerated at all times whilst in short term storage. Using sterilised equipment will reduce the risk of bacterial contamination.</p> <p>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.</p> <p>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</p>

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	Preservative: 0.05% Sodium azide Constituent: PBS

Purity	Immunogen affinity purified
Purification notes	This antibody is an affinity purified rabbit anti-GFP antibody purified on an affinity chromatography column made with highly purified recombinant GFP.
Clonality	Polyclonal
Isotype	IgG

Applications

The Abpromise guarantee Our [Abpromise guarantee](#) covers the use of ab69314 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
IP	★★★★★ (1)	Use at an assay dependent concentration. We routinely recommend using 20 µL of slurry per IP, which corresponds to 2 µg of IgG cross-linked to beads. this results in a clearly visible 10 µL bead pellet upon centrifugation. Unconjugated version: ab6556 . FITC conjugated version: ab66180 . Biotin conjugated version: ab69313 .

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 dihydroxytyrosine. Maturation of the chromophore requires nothing other than molecular oxygen.

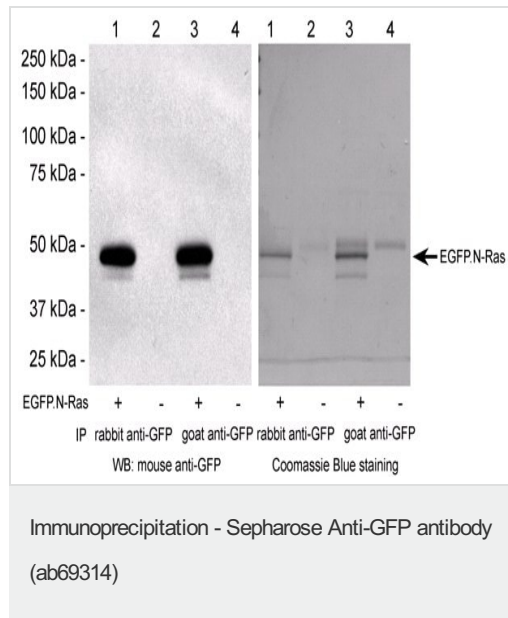
Biotechnological use: Green fluorescent protein has been engineered to produce a vast number of variously colored mutants, fusion proteins, and biosensors. Fluorescent proteins and its mutated allelic forms, blue, cyan and yellow have become a useful and ubiquitous tool for making chimeric proteins, 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



Lane 1 & 3: IP from COS 7 cells transfected with EGFP.N-Ras (48 kDa)

Lane 2 & 4: IP from untransfected COS 7 cells

Lane 1 & 2: IP using 15 ul of rabbit anti-GFP conjugated to sepharose beads (0.5 mg IgG per ml of beads)

Lane 3 & 4: IP using 15 ul of goat anti-GFP conjugated to sepharose beads (1 mg IgG per ml of beads)

Immunoprecipitation: COS 7 cell lysates containing 100 ug of total protein in 200 ul of 0.1% SDS-RIPA buffer with addition of complete protease inhibitor were used for each immunoprecipitation. Cell lysates were incubated with anti-GFP sepharose beads for 2 hours at 4oC with rocking. Beads were washed. Proteins were eluted with 1% SDS 50 mM HEPES (pH 7.4) at 80oC (15 min). Half of the IP sample was loaded on each lane of 10 % SDS PAGE gel and gel was processed for Western blotting/ECL.

Western blot: Primary antibody: monoclonal mouse anti-GFP (ab291) at 0.2 ug/ml in 5% non-fat milk/TBS-T; 1 hour incubation at room

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