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SP3005P Polyclonal Antibody to GFP - Purified

Alternate names: GFP-Tag, Green fluorescent protein

Quantity: 0.1 mg

Concentration: 1.0 mg/ml

Background: Green fluorescence protein (GFP) is a 27 KDa protein derived from the bioluminiscent jellyfish Aquorea victoria, emiting green light (509 nm) when excited (excitation by Blue or UV light, absorption peak at 395 nm).
GFP is a useful tool in cell biology research, as its intrinsic fluorescence can be visualized in living cells. Light-stimulated GFP fluorescence is species-independent and a fluorescence has been reported from many different types of GFP-expressing hosts, including microbes, invertebrates, vertebrates and plants. No exogenous substrates and cofactors are required for the fluorescence of GFP, since GFP autocatalytically forms a fluorescent pigment from natural amino acids present in the nascent protein.
GFP fluorescence is stable under fixation conditions and suitable for a variety of applications. GFP is widely used as a reporter (tag) for gene expression, enabling researchers to visualize and localize GFP-tagged proteins within living cells without any further staining. Other applications of GFP include measurement of distance between proteins through fluorescence energy transfer (FRET) protocols.
To increase a fluorescence intensity of GFP, chromophore mutations have been created. The Enhanced GFP has a fluorescence 35 times more intense than the wt-GFP. Mutagenesis of GFP has produced also many mutants (e.g. Yellow Fluorescent Protein, Cyan Fluorescent Protein) with warying spectral properties. Antibodies raised against full-length GFP variants should also detect other variants of the protein.

Uniprot ID: P42212

NCBI: 6100

Host: Rabbit

Immunogen: EGFP, a native full-length protein
Genename: GFP

Format: **State:** Liquid purified Ig fraction (› 95% pure by SDS-PAGE)
Purification: Affinity Chromatography on Protein A
Buffer System: PBS
Preservatives: 15 mM Sodium Azide

Applications: **Immunoprecipitation:** 10-20 µg/sample.
Immunocytochemistry: 1-3 µg/ml.
Western blot: 0.5-1.5 µg/ml.
Positive Control: transfected cells.
Negative Control: non-transfected cells.
Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.

Specificity: The antibody reacts specifically with GFP, EGFP, EYFP fusion proteins in all species.

Storage: Store undiluted at 2-8°C for one month or (in aliquots) at -20°C for longer.
Avoid repeated freezing and thawing.
Shelf life: one year from despatch.

Product Citations:

Purchased from Acris:

1. Ling YH, Wong CC, Li KW, Chan KM, Boukamp P, Liu WK. CCHCR1 interacts with EDC4, suggesting its localization in P-bodies. Exp Cell Res. 2014 Sep 10;327(1):12-23. doi: 10.1016/j.yexcr.2014.05.008. Epub 2014 May 21. PubMed PMID: 24858563.
2. Gentzel M, Schille C, Rauschenberger V, Schambony A. Distinct functionality of Dishevelled isoforms on Ca2+/Calmodulin dependent protein kinase 2 (CamKII) in Xenopus gastrulation. Mol Biol Cell. 2015 Jan 7. pii: mbc.E14-06-1089. PubMed PMID: 25568338.

Originator or purchased from resellers:

1. Valenta T, Lukas J, Doubravska L, Fafilek B, Korinek V. HIC1 attenuates Wnt signaling by recruitment of TCF-4 and beta-catenin to the nuclear bodies. EMBO J. 2006 Jun 7;25(11):2326-37. Epub 2006 May 25. PubMed PMID: 16724116.

Pictures:

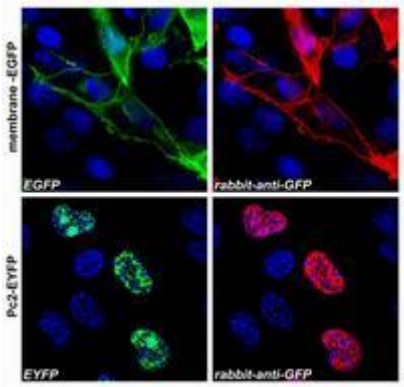

Figure 1. Confocal microscopy images of COS-7 cells transfected with expression constructs encoding membrane-tethered EGFP (membrane-EGFP; top) or nuclear Polycomb 2-EYFP fusion protein (Pc2-EYFP; bottom). The natural fluorescence of the produced proteins is shown in the green channel (left), the anti-GFP antibody signal was detected in the red channel (right). The system was carefully tested for overlap of these two optical channels and images were scanned separately in sequential scanning mode. The blue nuclear stain is also shown.

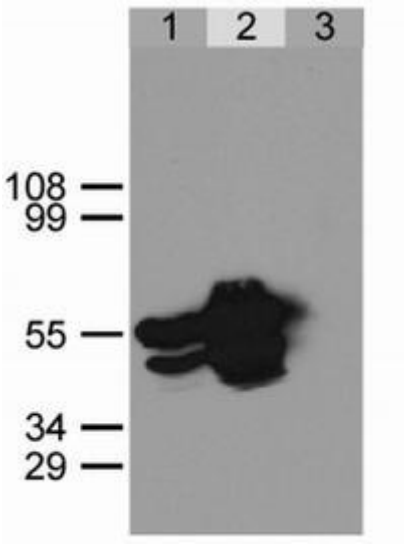

Figure 2. Immunoprecipitation of GFP-NLS from HEK293 cells using anti-GFP antibody. HEK293 cells were transfected with expression construct encoding GFP-NLS protein. Twenty hours post transfection cells were lysed in non-denaturating conditions (Lysis buffer: 20 mM Tris, pH 7.5, 100 mM NaCl, 0.5% Triton X-100, inhibitors of proteases). Aliquots of cell lysate were immunoprecipitated using a rabbit anti-GFP antibody (lane 2) or a pre-immune rabbit serum (lane 3). Immunoprecipitates together with a sample of the cell lysate (lane 1) were separated on SDS-PAGE polyacrylamide gel and immunoblotted with the anti-GFP antibody. The positions of molecular weight markers in kDa are indicated at the left.