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30-1330: Anti-GFP / Green Fluorescent Protein Polyclonal Antibody

Clonality :	Polyclonal
Application :	IP, WB, ICC
Reactivity :	Recognizes fusion proteins in all species
Format :	Purified
Isotype :	Rabbit IgG
Immunogen Information : EGFP, a native full-length protein	

Description

Green fluorescence protein (GFP) is a 27 KDa protein derived from the bioluminiscent jellyfish Aquorea victoria, emiting green light (lambda=509 nm) when excited (excitation by Blue or UV light, absorption peak lambda=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, chomophore mutations have been created. The EnhancedGFP 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.

Product Info

Amount :	0.1 mg
Purification :	Purified from rabbit serum by affinity chromatography
Storage condition :	Store at 2-8°C. Do not freeze.

Application Note

Immunoprecipitation Recommended dilution:

10-20 Ã[]µg/ Immunofluoroscence Recommended dilution:

Purified Antibody: 1-3 Ã\µg/ml

Positive control:

transfected cells

Negative control:

non-transfected cells

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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), polyclonal 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 polyclonal 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.