

nCor Green Fluorescent Protein Goat Polyclonal Antibody

GPCA-GFP

Species Cross-Reactivity

Ordering Information Web www.encorbio.com Email admin@encorbio.com Phone 352-372-7022 Fax 352-372-7066

HGNC Name: NA UniProt: Q6YGZ0 RRID: AB 2737371 Immunogen: Recombinant AcGFP protein purified from E. coli

Format: Affinity purified antibody at 1mg/mL in 50% PBS, 50% glycerol plus 5mM NaN Storage: Store at 4°C for short term, for longer term

at -20°C Recommended dilutions:

WB: 1:1,000-5,000 IF/IHC: 1:5,000

References:

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2. Shimomura, O. Structure of the chromophore of Aequorea green fluorescent protein. FEBS Lett. 104:220-2 (1979).

3. Prasher DC, et al. Primary structure of the Aequorea victoria green-fluorescent protein. Gene 111:229-33 (1992).

4. Cody CW, et al. Chemical structure of the hexapeptide chromophore of the Aeguorea green-fluorescent protein. Biochem. 32:1212-8 (1993).

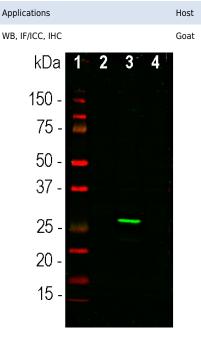
5. Chalfie M, et al. Green Fluorescent protein as a marker for gene expression. Science 263:802-5 (1994).

6. Heim R, Prasher DC, Tsien RY. Wavelength mutations and post-translational autoxidation of green fluorescent protein. PNAS 91:12501-04 (1994).

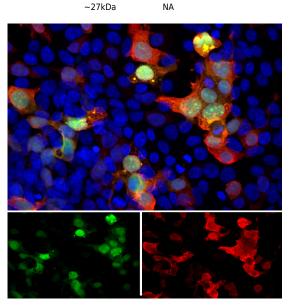
7. Ormo M, et al. Crystal structure of the Aequorea victoria green fluorescent protein. Science 273:1392-95 (1996).

8. Tsien RY. The green fluorescent protein. Annu. Rev. Biochem. 67:509-44 (1998). 9. Zacharias DA, Violin JD, Newton AC, Tsien RY. Partitioning of lipid-modified monomeric GFPs into membrane microdomains of live cells. Science 296:913-6 (2002).

10. Gurskaya NG, et al. A colourless green fluorescent protein homologue from the nonfluorescent hydromedusa Aeguorea coerulescens and its fluorescent mutants. Biochem. J. 373:403-8 (2003).



Western blot analysis of HEK293 cell lysates using goat pAb to GFP, GPCA-GFP, dilution 1:1,000, in green: [1] protein standard, [2] nontransfected control cells, [3] cells transfected with a GFP construct and [4] cells transfected with an mCherry construct. Strong band at ~27kDa corresponds to GFP protein detected only in cells



Molecular Wt.

Immunofluorescent analysis of transfected HEK293 cells with a GFPconstruct in green stained with goat pAb to GFP, GPCA-GFP, dilution 1:5,000 in red. The blue is Hoechst staining of nuclear DNA. The GPCA-GFP antibody reveals GFP protein expressed only in transfected cells, as a result transfected cells are appeared express both red and green signals and so appear an orange-golden color. Untransfected cells show neither signal.

transfected with GFP construct. This antibody does not recognize the mCherry protein.

Background:

The green fluorescent protein (GFP) is a 27kDa protein isolated originally from the jellyfish *Aequoria victoria*. It has an endogenous fluorochrome activity with excitation maximum at 395nm and emission maximum at 509nm, which is similar to that of fluorescein (1,2). The GFP gene was cloned and sequenced and the origin of the fluorochrome by autocatalytic activity of certain amino acids was discovered (3,4). Much interest in GFP was generated when it was shown that fluorescence develops rapidly when the protein is expressed and requires only molecular oxygen and no other cofactors. As a result GFP can be expressed in fluorescent form in essentially any prokaryotic or eukaryotic cell (5). GFP has been engineered to produce a vast number of variously colored mutants including blue, cyan and yellow protein derivatives, BFP, CFP and YFP (6-9). GFP and other fluorescent proteins derived from jellyfish, coral and other Cnidaria are widely used as tracers in transfection and transgenic experiments to monitor gene expression and protein localization in vivo and in in vitro. The crystal structure of GFP was determined (7) which allowed amino acid modifications to improve spectral properties and prevent multimerization (8,9). The 2008 Nobel prize in chemistry was awarded "for the discovery and development of the green fluorescent protein, GFP"

Isotype

The GPCA-GFP antibody was made against a recombinant GFP construct originating from an Aequoria species which was engineered to improve spectral properties and prevent oligomerization (10). This form of GFP, referred to as AcGFP, is 94% identical to the eGFP developed by Tsien and coworkers and is the form of GFP inserted in the Clontech/Takara pAcGFP and related expression vectors. We also supply the immunogen, PROT-AcGFP. The antibody can be used to verify the expression, size and stability of both AcGFP and eGFP fusion proteins in western blotting experiments and to amplify GFP signals in tissues of transgenic animals. We also supply mouse monoclonal antibodies and rabbit and chicken polyclonal antibodies to this protein, MCA-3B11, MCA-1F1, RPCA-GFP and CPCA-GFP.

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Abbreviation Key:

mAb—Monoclonal Antibody pAb—Polyclonal Antibody WB—Western Blot IF—Immunofluorescence ICC—Immunocytochemistry IHC—Immunohistochemistry E—ELISA Hu—Human Mo—Monkey Do—Dog Rt—Rat Ms—Mouse Co—Cow Pi—Pig Ho—Horse Ch—Chicken Dr-D. rerio Dm-D. melanogaster Sm-S. mutans Ce-C. elegans Sc-S. cerevisiae Sa-S. aureus Ec-E. coli.