

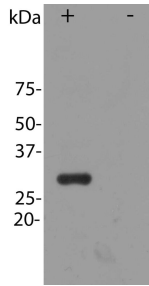
Cat. No:	AB-82367
Conjugate:	Unconjugated
Size:	100 ug
Clone:	POLY
Concentration:	1mg/mL
Host:	Rb
Isotype:	IgG
Immunogen:	Full length recombinant protein
Reactivity:	All Species
Applications:	Western Blot: 1:500 Immunofluorescence / Immunohistochemistry: 1:250
Molecular Weight:	28kDa
Purification:	Aff. Pur.

Background:

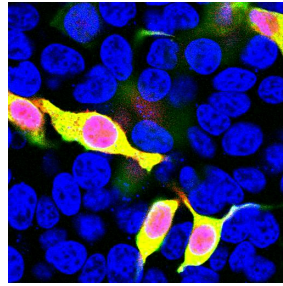
mCherry is derived from proteins originally isolated from Cnidarians (jelly fish, sea anemones and corals), and is used as a fluorescent tracer in transfection and transgenic experiments. The prototype for these fluorescent proteins is Green Fluorescent Protein (GFP), which is a ~27 kDa protein isolated originally from the jellyfish *Aequoria victoria*. GFP was the basis of the 2008 Nobel Prize in Chemistry, awarded to Osamu Shimomura, Martin Chalfie and Roger Tsien, specifically "for the discovery and development of the green fluorescent protein, GFP". GFP was shown to fluoresce on contact with molecular oxygen, requiring no other cofactors, and so can be expressed in fluorescent form in essentially any prokaryotic or eukaryotic cell. The mCherry protein is derived from DsRed, a red fluorescent protein related to GFP isolated from so-called disc corals of the genus *Discosoma*. DsRed is similar in size and properties to GFP, but, obviously, produces a red rather than a green fluorochrome. The original DsRed was engineered extensively in the Tsien lab to prevent it from forming tetramers and dimers and to modify and improve the spectral properties (1-3). Several further cycles of mutation, directed modification and evolutionary selection produced mCherry, which has an excitation maximum at 587 nm and an emission maximum at 610 nm (4). We expressed the mCherry protein sequence shown in reference 4 in bacteria, purified out the mCherry and raised this rabbit polyclonal antibody. This was affinity purified and was found to stain a band of the expected size in HEK293 cells transfected with the pFin-EF1-mCherry vector designed to express mCherry which was obtained from Clontech. As shown below, the antibody does not stain any protein band in untransfected HEK293 cells.

Form: Liquid

Storage: Shipped on ice. Store at 4°C. For long term storage, leave frozen at -20°C. Avoid freeze / thaw cycles



Blot of HEK293 cells transfected with pFin-EF1-mCherry vector, in the lane marked "+". HEK293 cells which were not transfected with this vector show no protein band in lane marked "



HEK293 cells transfected in the same way and viewed in the confocal microscope. Most HEK293 cells are not transfected so only the nucleus of these cells can be visualized with a blue DNA stain. Cells which are transfected with mCherry are red. Staining with mCherry is shown in Green. Green antibody staining is only seen in cells which express mCherry, as expected, and the superimposition of the green and red signals results in an orange signal. Interestingly, stronger mCherry staining is seen in the nucleus, possibly due to degradation of some mCherry molecules to release the low molecular weight mCherry fluorochrome.

References

1. Baird GS, Zacharias DA, Tsien RY. Biochemistry, mutagenesis, and oligomerization of DsRed, a red fluorescent protein from coral. *Proc Natl Acad Sci U S A.* 97:11984-9 (2000).
2. Gross LA, Baird GS, Hoffman RC, Baldrige KK, Tsien RY. The structure of the chromophore within DsRed, a red fluorescent protein from coral. *Proc Natl Acad Sci U S A.* 97:11990-5 (2000).
3. Heikal AA, Hess ST, Baird GS, Tsien RY, Webb WW. Molecular spectroscopy and dynamics of intrinsically fluorescent proteins: coral red (dsRed) and yellow (Citrine). *Proc Natl Acad Sci U S A.* 97:11996-2001 (2000).
4. Shaner NC, Campbell RE, Steinbach PA, Giepmans BN, Palmer AE, Tsien RY. Improved monomeric red, orange and yellow fluorescent proteins derived from *Discosoma* sp. red fluorescent protein. *Nature Biotechnology* 22:1567-1572 (2004).

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