abcam

Product datasheet

Anti-Rhodopsin antibody [1D4] ab5417

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Overview

Product name Anti-Rhodopsin antibody [1D4]

Description Mouse monoclonal [1D4] to Rhodopsin

Host species Mouse

Specificity ab5417 detects Rhodopsin from human and bovine retinal samples. Data from Yin J et al., 2012

(PMID 22743318) indicates that in Zebrafish ab5417 appears to recognize Red Opsin rather than

Rhodopsin.

Tested applications Suitable for: ELISA, IHC-FoFr, IP, WB, IHC-Fr, IHC-P, ICC/IF

Species reactivity Reacts with: Mouse, Rat, Cow, Human, Zebrafish, Amphibian

Predicted to work with: Rabbit

Immunogen Tissue, cells or virus corresponding to Bovine Rhodopsin. Bleached bovine ROS [rod outer

segment] disk membranes

Epitope The epitope for this antibody has been localized to the C-terminal nine amino acids of bovine

rhodopsin known as the 1D4 epitope.

Positive control WB: HL60 whole cell lysate IHC-P: Human and mouse retinal tissue

General notes

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.

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

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -

80°C. Avoid freeze / thaw cycle.

Storage buffer Preservative: 0.05% Sodium azide

Constituents: 99% PBS, 0.1% BSA

Purity Protein G purified

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Primary antibody notes

Vision involves the conversion of light into electrochemical signals that are processed by the retina and subsequently sent to and interpreted by the brain. The process of converting light to an electrochemical signal begins when the membrane-bound protein, rhodopsin, absorbs light within the retina. Photoexcitation of rhodopsin causes the cytoplasmic surface of the protein to become catalytically active. In the active state, rhodopsin activates transducin, a GTP binding protein. Once activated, transducin promotes the hydrolysis of cGMP by phosphodiesterase (PDE). The decrease of intracellular cGMP concentrations causes the ion channels within the outer segment of the rod or cone to close, thus causing membrane hyperpolarization and, eventually, signal transmission. Rhodopsin's activity is believed to be shut off by its phosphorylation followed by binding of the soluble protein arrestin.

Clonality Monoclonal

Clone number 1D4 lsotype lgG1

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab5417 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	
ELISA		Use at an assay dependent concentration.	
IHC-FoFr		Use at an assay dependent concentration. PubMed: 19587120	
IP		Use at an assay dependent concentration.	
WB	★★★★★ (2)	1/100 - 1/1000. Detects a band of approximately 40 kDa.	
IHC-Fr	**** <u>(1)</u>	Use at an assay dependent concentration. PubMed: 22743318	
IHC-P		1/100 - 1/1000.	
ICC/IF	**** <u>(1)</u>	Use at an assay dependent concentration.	

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Function Photoreceptor required for image-forming vision at low light intensity. Required for photoreceptor

cell viability after birth. Light-induced isomerization of 11-cis to all-trans retinal triggers a conformational change leading to G-protein activation and release of all-trans retinal.

Tissue specificity Rod shaped photoreceptor cells which mediates vision in dim light.

Involvement in disease Retinitis pigmentosa 4

Night blindness, congenital stationary, autosomal dominant 1

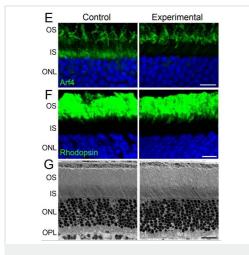
Sequence similarities Belongs to the G-protein coupled receptor 1 family. Opsin subfamily.

Post-translational Phosphorylated on some or all of the serine and threonine residues present in the C-terminal

modifications region.

Contains one covalently linked retinal chromophore.

Images



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Rhodopsin antibody
[1D4] (ab5417)

Pearring et al PLoS Genet. 2017 Apr 14;13(4):e1006740. doi: 10.1371/journal.pgen.1006740. eCollection 2017 Apr. Fig 3. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

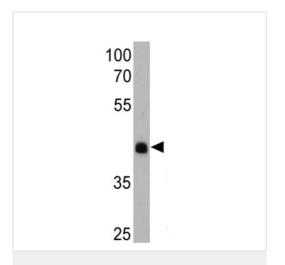
Deletion of Arf4 mice from the retina does not disrupt rhodopsin localization or photoreceptor morphology.

E. Arf4 immunostaining in $Arf4^{flox}/CagCreER$ experimental and control retinal cross-sections. Image of the photoreceptor IS where the biosynthetic membranes are localized. Eyes were collected at P34. Scale bar = 10 μ m.

F. Rhodopsin immunostaining in $Arf4^{flox}/CagCreER$ experimental and control retinal cross-sections. Eyes were collected at P34. Scale bar = 10 μ m.

G. Comparative analysis of photoreceptor morphology in $Arf4^{flox}/CagCreER$ experimental and control retinal cross-sections. Eyes were collected at P41. Scale bar = 20 μ m.

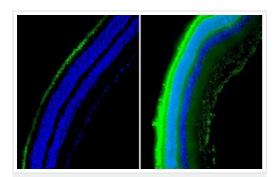
OS = outer segment, IS = inner segment, ONL = outer nuclear layer, OPL = outer plexiform layer.



Western blot - Anti-Rhodopsin antibody [1D4] (ab5417)

Anti-Rhodopsin antibody [1D4] (ab5417) at 1/500 dilution + HL60 (Human promyelocytic leukemia cell line) cell lysate at 25 µg

Observed band size: 40 kDa

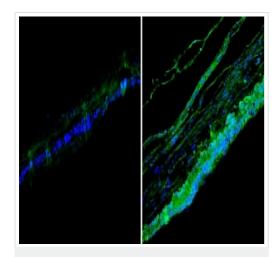


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Rhodopsin antibody
[1D4] (ab5417)

Immunohistochemical analysis of formalin-fixed mouse retinal tissue, labeling rhodopsin with ab5417 at a 1:50 dilution in 3% BSA-PBS solution and incubated at 4°C overnight in a high humidity environment.

A DyLight[®] 488 secondary antibody was used (green) incubated at room temperature in the dark. The tissue was counterstained with DAPI against DNA, showing nuclear compartments. Prior to staining the formalin-fixed tissue was permeabilized with 0.1% Triton X-100 in TBS for between 5 and 10 minutes, then blocked with 3% BSA-PBS for 30 minutes at room temperature.

The left image is a negative control with only the secondary antibody and the right image is in the presence of ab5417 and the secondary.

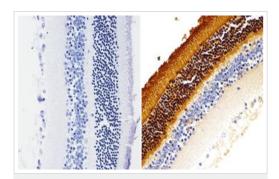


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Rhodopsin antibody
[1D4] (ab5417)

Immunohistochemical analysis of formalin-fixed human retinal tissue, labeling rhodopsin with ab5417 at a 1:50 dilution in 3% BSA-PBS solution and incubated at 4°C overnight in a high humidity environment.

A DyLight[®] 488 secondary antibody was used (green) incubated at room temperature in the dark. The tissue was counterstained with DAPI against DNA, showing nuclear compartments. Prior to staining the formalin-fixed tissue was permeabilized with 0.1% Triton X-100 in TBS for between 5 and 10 minutes, then blocked with 3% BSA-PBS for 30 minutes at room temperature.

The left image is a negative control with only the secondary antibody and the right image is in the presence of ab5417 and the secondary.

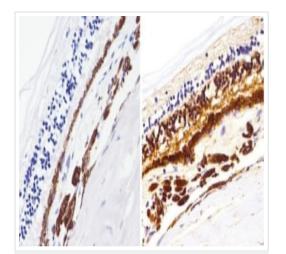


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Rhodopsin antibody
[1D4] (ab5417)

Immunohistochemical analysis of paraffin-embedded mouse retinal tissue labeling Rhodopsin with ab5417.

Secondary used was HRP conjugated. Prior preparation was initiated by antigen retrival using 10mM sodium citrate at pH 6.0, then the sample was microwaved for 8 to 15 minutes. Subsequent to retrival the retinal tissue was blocked for 15 minutes at room temperature with 3% hydrogen peroxide. The sample was then incubated with ab5417 in 3% BSA-PBS at 4°C at a dilution of 1:1000, overnight. Hematoxylin was used to counterstain the tissue.

The left side of the image is shown as a negative control and is the tissue in the absence of ab5417, the right side is in the prescence of the counterstain, ab5417 and the HRP conjugated secondary.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Rhodopsin antibody
[1D4] (ab5417)

Immunohistochemical analysis of paraffin-embedded human retinal tissue labeling Rhodopsin with ab5417.

Secondary used was HRP conjugated. Prior preparation was initiated by antigen retrieval was performed using 10mM sodium citrate (pH 6.0), microwaved for 8-15 min. Subsequent to retrival the retinal tissue was blocked in 3% H2O2-methanol for 15 min at room temperature. The sample was then incubated with ab5417 in 3% BSA-PBS at a dilution of 1:200 overnight at 4°C, overnight. Hematoxylin was used to counterstain the tissue.

The left side of the image is shown as a negative control and is the tissue in the absence of ab5417, the right side is in the prescence of the counterstain, ab5417 and the HRP conjugated secondary.

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