

Product Datasheet

RPE65 Antibody (401.8B11.3D9) NB100-355

Unit Size: 0.2 ml

Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.

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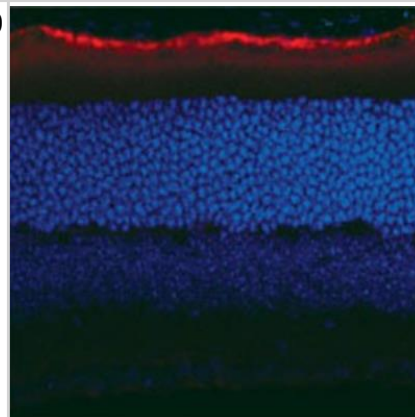
NB100-355

RPE65 Antibody (401.8B11.3D9)

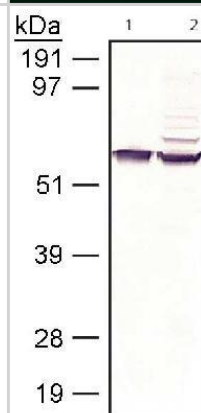
Product Information	
Unit Size	0.2 ml
Concentration	1.0 mg/ml
Storage	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	401.8B11.3D9
Preservative	0.05% Sodium Azide
Isotype	IgG1 Kappa
Purity	Protein G purified
Buffer	PBS
Target Molecular Weight	65 kDa
Product Description	
Host	Mouse
Gene ID	6121
Gene Symbol	RPE65
Species	Human, Mouse, Rat, Porcine, Bovine, Canine, Chicken, Xenopus
Reactivity Notes	Based on immunogen sequence similarity, this antibody is predicted to react with Duck/Ostrich and several other Birds species (89%), Zebrafish (74%) and Sea lamprey (71%).
Marker	Retinal Pigment Epithelium Marker
Specificity/Sensitivity	NB100-355 is specific for RPE65.
Immunogen	Bovine RPE65 microsomal membrane proteins. [UniProt# Q28175]
Product Application Details	
Applications	Western Blot, Flow Cytometry, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, CyTOF-ready, Immunocytochemistry
Recommended Dilutions	Western Blot 1:500 - 1:1000, Flow Cytometry, Immunohistochemistry 1:250, Immunocytochemistry/Immunofluorescence 1:10-1:500, Immunohistochemistry-Paraffin 1:250-1:500, Immunohistochemistry-Frozen 1:250, Immunocytochemistry 1:250, CyTOF-ready
Application Notes	For Western blot, this antibody has been validated in lysates of bovine RPE membrane and COS7 cells transfected with human RPE65, and in both samples, the antibody recognized a band at ~65 kDa, representing RPE65 protein. Note:- Hamel et al. have reported that in cultured bovine RPE cells, the levels of RPE65 are undetectable in WB after day 14, however, the mRNA levels are detectable by Northern for at least 7 weeks (PMID: 8340400). The observed molecular weight of the protein may vary from the listed predicted molecular weight due to post translational modifications, post translation cleavages, relative charges, and other experimental factors.

Images

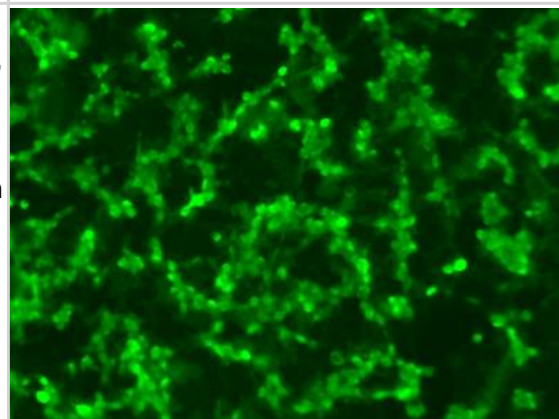
Immunohistochemistry-Frozen: RPE65 Antibody (401.8B11.3D9) [NB100-355] - Staining of RPE65 in a cryosection of mouse retina tissue using RPE65 antibody (clone 401.8B11.3D9).



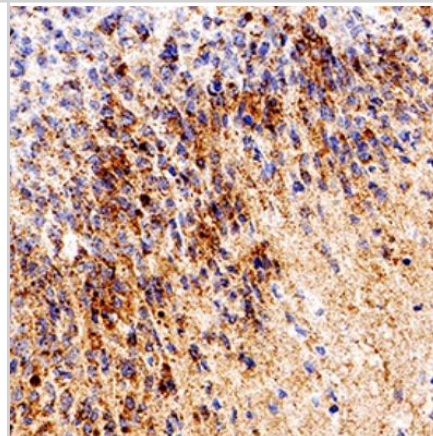
Western Blot: RPE65 Antibody (401.8B11.3D9) [NB100-355] - WB analysis of RPE65 in 20ug lysate of COS7 cells expressing recombinant Human RPE65 (Lane 1) and 5ug of Bovine retinal pigment epithelium membrane fraction (Lane 2). Blot processed for detection with alkaline phosphatase conjugated goat-anti Mouse IgG secondary antibody and NBT/BCIP substrate.



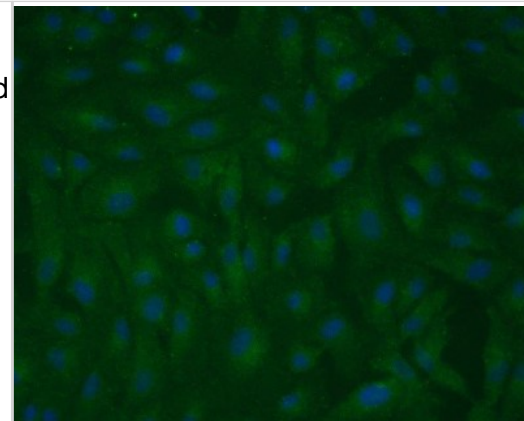
Immunocytochemistry/Immunofluorescence: RPE65 Antibody (401.8B11.3D9) [NB100-355] - ICC-IF analysis of cultured ARPE19 cells, a spontaneously arising human retinal pigment epithelia cell line - 10 minutes fixation in 4% PFA, 10 minutes permeabilization in PBS containing 0.2% Triton X-100 (PBS-T), 1 hour blocking in 10% normal goat serum containing 1% BSA in PBS-T, 1:100 primary antibody dilution in PBS, ON 4C incubation.



Immunohistochemistry-Paraffin: RPE65 Antibody (401.8B11.3D9) [NB100-355] - IHC analysis of formalin fixed paraffin-embedded (FFPE) human glioblastoma using Histone H3 [pS10] antibody at 1:500 on a Bond Rx autostainer (Leica Biosystems). The assay involved 20 minutes of heat induced antigen retrieval (HIER) using 10mM sodium citrate buffer (pH 6.0) and endogenous peroxidase quenching with peroxide block. The sections were incubated with primary antibody for 30 minutes and Bond Polymer Refine Detection (Leica Biosystems) with DAB was used for signal development followed by counterstaining with hematoxylin. Whole slide scanning and capturing of representative images was performed using Aperio AT2 (Leica Biosystems). Cytoplasmic staining of RPE65 was observed. Staining was performed by Histowiz.



Immunocytochemistry: RPE65 Antibody (401.8B11.3D9) [NB100-355] - ARPE-19 monolayer on glass substrate. Fixative Methanol Blocking Goat Serum. Dilution 1:250. Incubation 1h RT. This image was submitted via customer Review.



Publications

Sun J, Huang P, Liang J et al. Cooperation of Rel family members in regulating Ab1-40-mediated pro-inflammatory cytokine secretion by retinal pigment epithelial cells *Cell Death Dis.* 2017 Oct 12 [PMID: 29022897] (ICC/IF, Mouse)

Li Y, Zhang Y, Xu Y et al. Patient-specific mutations impair BESTROPHIN1's essential role in mediating Ca(2+)-dependent Cl(-) currents in human RPE. *Elife.* 2017 Oct 24 [PMID: 29063836] (WB, Human)

Steinfeld J, Steinfeld I, Bausch A et al. BMP-induced reprogramming of the retina into RPE requires WNT signalling in the developing chick optic cup. *Biol Open* May 25 2017 12:00AM [PMID: 28546339] (IHC, Chicken)

Chowers G, Cohen M, Marks-Ohana D et al. Course of Sodium Iodate-Induced Retinal Degeneration in Albino and Pigmented Mice. *Invest. Ophthalmol. Vis. Sci.* Apr 1 2017 12:00AM [PMID: 28418497] (IHC, Mouse)

Zheng W, Meng Q, Wang H et al. IGF-1-Mediated Survival from Induced Death of Human Primary Cultured Retinal Pigment Epithelial Cells Is Mediated by an Akt-Dependent Signaling Pathway. *Mol. Neurobiol.* Feb 25 2017 12:00AM [PMID: 28238097] (Human)

Lyzogubov VV, Bora PS, Wu X et al. The Complement Regulatory Protein CD46 Deficient Mouse Spontaneously Develops Dry-Type Age-Related Macular Degeneration-Like Phenotype. *Am. J. Respir. Cell Mol. Biol.* 2016 Jun 11 [PMID: 27295359] (ICC/IF)

Details:

This citation used the Alexa Fluor 647 version of this antibody.

Zhou TE, Rivera JC, Bhosle VK et al. Choroidal Involution Is Associated with a Progressive Degeneration of the Outer Retinal Function in a Model of Retinopathy of Prematurity: Early Role for IL-1B. *Am. J. Pathol.* Oct 18 2016 12:00AM [PMID: 27768863] (IHC)

Di Lauro S, Rodriguez-Crespo D, Gayoso MJ et al. A novel coculture model of porcine central neuroretina explants and retinal pigment epithelium cells. *Mol Vis* 2016 Mar 25 [PMID: 27081295]

Iwabe S, Ying GS, Aguirre GD, Beltran WA. Assessment of visual function and retinal structure following acute light exposure in the light sensitive T4R rhodopsin mutant dog. *Exp. Eye Res.* 2016 Apr 13 [PMID: 27085210] (IHC-Fr, Canine)

Chen M, Rajapakse D, Fraczek M et al. Retinal pigment epithelial cell multinucleation in the aging eye - a mechanism to repair damage and maintain homeostasis *Aging Cell* Feb 15 2016 12:00AM [PMID: 26875723] (ICC/IF, Mouse)

Ayala-Pena VB, Pilotti F, Volonte Y et al. Protective effects of retinoid x receptors on retina pigment epithelium cells *Biochim. Biophys. Acta* Feb 13 2016 12:00AM [PMID: 26883505] (ICC/IF, Human)

Parker J, Mitrousis N, Shoichet MS. Hydrogel for Simultaneous Tunable Growth Factor Delivery and Enhanced Viability of Encapsulated Cells in Vitro. *Biomacromolecules.* 2016 Feb 08 [PMID: 26762290] (ICC/IF, Human)

More publications at <http://www.novusbio.com/NB100-355>

Procedures

Western Blot Protocol for RPE65 Antibody (NB100-355)

Western Blot

1. Bovine RPE membrane (5ug) and recombinant human RPE65 (~20ug) expressed in COS7 cells were loaded on a NuPAGE Novex Bis-Tris 10%gel along with SeeBlue Plus2 marker.
2. Run in NuPAGE MOPS electrophoresis buffer (Invitrogen).
- 3 Samples were probed with NB 100-355 RPE65 antibody (1:5,000) followed by alkaline phosphatase-conjugated goat anti-mouse IgG incubation.
4. Visualized by NBT/BCIP substrate.





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Products Related to NB100-355

NBP2-46608	Mouse Eye Tissue Lysate (Normal)
HAF007	Goat anti-Mouse IgG Secondary Antibody [HRP (Horseradish Peroxidase)]
NB720-B	Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]
NBP1-43319-0.5mg	Mouse IgG1 Kappa Light Chain Isotype Control (P3.6.2.8.1)

Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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