abcam

Product datasheet

Anti-PAX6 antibody [EPR3352(2)] ab109233

Recombinant RabMAb

2 References 11 Images

Overview

Product name Anti-PAX6 antibody [EPR3352(2)]

Description Rabbit monoclonal [EPR3352(2)] to PAX6

Host species Rabbit

Tested applications Suitable for: IHC-P, WB, IP, mIHC

Unsuitable for: Flow Cyt or ICC/IF

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control HeLa, K562, and HepG2 whole cell lysate (ab7900) IHC-P: FFPE Rat retina normal, Mouse

retina normal, Human retina, Rat pancreas, IP: HeLa cell lysate mlHC: Human retina tissue.

General notes This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb patents**.

Properties

Form

Storage instructions Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 40% Glycerol (glycerin, glycerine), PBS, 0.05% BSA

Purity Protein A purified

Clonality Monoclonal Clone number EPR3352(2)

ΙgG Isotype

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab109233 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
IHC-P		1/2000. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB		1/1000 - 1/10000. Detects a band of approximately 47 kDa (predicted molecular weight: 47 kDa).
IP		1/50.
mIHC		Use at an assay dependent concentration.

Application notes

Is unsuitable for Flow Cyt or ICC/IF.

Target

Function

Transcription factor with important functions in the development of the eye, nose, central nervous system and pancreas. Required for the differentiation of pancreatic islet alpha cells (By similarity). Competes with PAX4 in binding to a common element in the glucagon, insulin and somatostatin promoters. Regulates specification of the ventral neuron subtypes by establishing the correct progenitor domains (By similarity). Isoform 5a appears to function as a molecular switch that specifies target genes.

Tissue specificity

Fetal eye, brain, spinal cord and olfactory epithelium. Isoform 5a is less abundant than the PAX6 shorter form.

Involvement in disease

Defects in PAX6 are the cause of aniridia (AN) [MIM:106210]. A congenital, bilateral, panocular disorder characterized by complete absence of the iris or extreme iris hypoplasia. Aniridia is not just an isolated defect in iris development but it is associated with macular and optic nerve hypoplasia, cataract, corneal changes, nystagmus. Visual acuity is generally low but is unrelated to the degree of iris hypoplasia. Glaucoma is a secondary problem causing additional visual loss over time.

Defects in PAX6 are a cause of Peters anomaly (PAN) [MIM:604229]. Peters anomaly consists of a central corneal leukoma, absence of the posterior corneal stroma and Descemet membrane, and a variable degree of iris and lenticular attachments to the central aspect of the posterior cornea.

Defects in PAX6 are a cause of foveal hypoplasia (FOVHYP) [MIM:136520]. Foveal hypoplasia can be isolated or associated with presenile cataract. Inheritance is autosomal dominant. Defects in PAX6 are a cause of keratitis hereditary (KERH) [MIM:148190]. An ocular disorder characterized by corneal opacification, recurrent stromal keratitis and vascularization. Defects in PAX6 are a cause of coloboma ocular (COLO) [MIM:120200]; also known as uveoretinal coloboma or coloboma of iris, choroid and retina. Ocular colobomas are a set of malformations resulting from abnormal morphogenesis of the optic cup and stalk, and the fusion of the fetal fissure (optic fissure). Severe colobomatous malformations may cause as much as 10% of the childhood blindness. The clinical presentation of ocular coloboma is variable. Some individuals may present with minimal defects in the anterior iris leaf without other ocular defects. More complex malformations create a combination of iris, uveoretinal and/or optic nerve defects without or with microphthalmia or even anophthalmia.

Defects in PAX6 are a cause of coloboma of optic nerve (COLON) [MIM:120430]. Defects in PAX6 are a cause of bilateral optic nerve hypoplasia (BONH) [MIM:165550]; also known as bilateral optic nerve aplasia. A congenital anomaly in which the optic disc appears abnormally small. It may be an isolated finding or part of a spectrum of anatomic and functional abnormalities that includes partial or complete agenesis of the septum pellucidum, other midline brain defects, cerebral anomalies, pituitary dysfunction, and structural abnormalities of the pituitary.

Defects in PAX6 are a cause of aniridia cerebellar ataxia and mental deficiency (ACAMD) [MIM:206700]; also known as Gillespie syndrome. A rare condition consisting of partial rudimentary iris, cerebellar impairment of the ability to perform coordinated voluntary movements, and mental retardation.

Sequence similarities Belongs to the paired homeobox family.

Contains 1 homeobox DNA-binding domain.

Contains 1 paired domain.

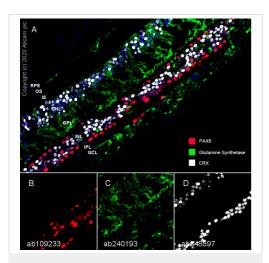
Developmental stage Expressed in the developing eye and brain.

Post-translational modifications

Ubiquitinated by TRIM11, leading to ubiquitination and proteasomal degradation.

Cellular localization Nucleus.

Images



Multiplex immunohistochemistry - Anti-PAX6 antibody [EPR3352(2)] (ab109233)

Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Human retina tissue labeling PAX6, Glutamine Synthetase and CRX with ab109233 at 1/10000 dilution, **ab240193** at 1/20000 dilution and **ab248897** at 1/1000 dilution followed by a ready to use Opal Polymer HRP Ms + Rb secondary antibody. Nuclear counter stain used was DAPI.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

Panel A: merged staining of anti-CRX (gray; Opal™690), anti-Glutamine Synthetase (green; Opal™520) and anti-PAX6 (red; Opal™570) on human retina.

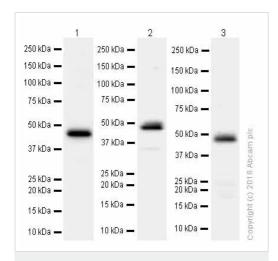
Panel B: anti-PAX6 stained on retinal progenitor cells.

Panel C: anti-Glutamine Synthetase stained on Müller glia.

Panel D: anti-CRX stained on subset cells of outer nuclear layer and inner nuclear layer.

The section was incubated in three rounds of staining: in the order of <u>ab248897</u>, <u>ab240193</u>, and ab109233 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems
BOND® RX instrument with an Opal™ 4-color kit. Image acquisition
was performed with Leica SP8 confocal microscope.



Western blot - Anti-PAX6 antibody [EPR3352(2)] (ab109233)

All lanes : Anti-PAX6 antibody [EPR3352(2)] (ab109233) at 1/1000 dilution

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates

Lane 2: Mouse retina lyates

Lane 3: Rat retina lyates

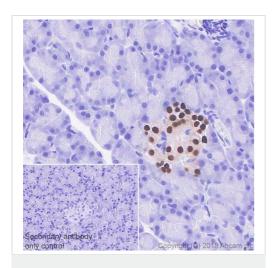
Lysates/proteins at 15 µg per lane.

Secondary

Lanes 1-2: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

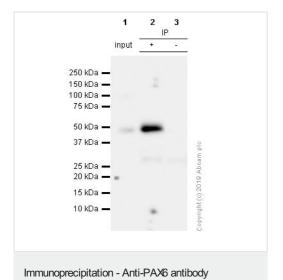
Lane 3: Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution (Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated)

Predicted band size: 47 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PAX6 antibody
[EPR3352(2)] (ab109233)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Rat pancreas tissue sections labeling PAX6 with Purified ab109233 at 1:2000 dilution (0.52 μ g/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use)was used. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



[EPR3352(2)] (ab109233)

ab109233 (purified) at 1/50 dilution ($20\mu g/ml$) immunoprecipitating PAX6 in HeLa whole cell lysate.

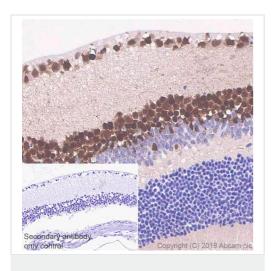
Lane 1 (input): HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate 10µg

Lane 2 (+): ab109233 & HeLa whole cell lysate
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of

ab109233 in HeLa whole cell lysate

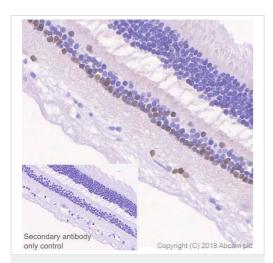
For western blotting, ab109233 at 1/500 and veriBlot for IP secondary antibody (HRP) (ab131366) was used at 1/1000 dilution.

Blocking and diluting buffer: 5% NFDM /TBST.



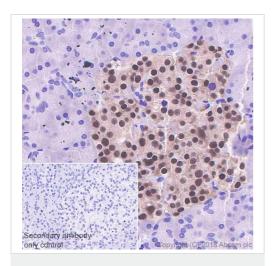
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PAX6 antibody
[EPR3352(2)] (ab109233)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Mouse retina tissue sections labeling PAX6 with Purified ab109233 at 1:2000 dilution (0.52 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PAX6 antibody
[EPR3352(2)] (ab109233)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human retina tissue sections labeling PAX6 with Purified ab109233 at 1:2000 dilution (0.52 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use)was used. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PAX6 antibody

[EPR3352(2)] (ab109233)

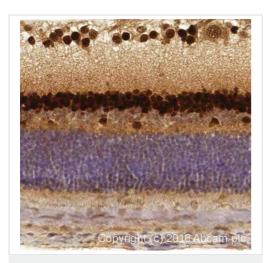
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human pancreas tissue sections labeling PAX6 with Purified ab109233 at 1:2000 dilution (0.52 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use). Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PAX6 antibody
[EPR3352(2)] (ab109233)

IHC image of Pax6 staining in normal mouse retina formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab109233, 1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

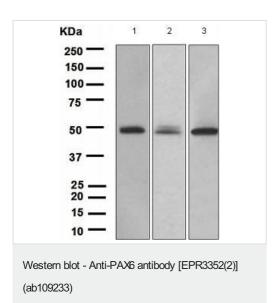


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PAX6 antibody

[EPR3352(2)] (ab109233)

IHC image of Pax6 staining in normal rat retina formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab109233, 1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



All lanes : Anti-PAX6 antibody [EPR3352(2)] (ab109233) at 1/1000 dilution

Lane 1 : HeLa cell lysate
Lane 2 : K562 cell lysate
Lane 3 : HepG2 cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: HRP labelled goat anti-rabbit at 1/2000 dilution

Predicted band size: 47 kDa **Observed band size:** 47 kDa



Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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