

Product datasheet

Anti-MERTK antibody [Y323] ab52968

KO VALIDATED Recombinant RabMAb

★★★★★ 4 Abreviews 57 References 10 Images

Overview

Product name	Anti-MERTK antibody [Y323]
Description	Rabbit monoclonal [Y323] to MERTK
Host species	Rabbit
Tested applications	Suitable for: WB, IP, IHC-P, IHC-Fr Unsuitable for: Flow Cyt or ICC/IF
Species reactivity	Reacts with: Human
Immunogen	Synthetic peptide within Human MERTK aa 1-100 (N terminal). The exact sequence is proprietary.
Positive control	WB: A172, HEK-293 and K562 cell lysate. Ild-type HAP1 whole cell lysate IHC-P: Human tonsil and lymphoma tissues. IP: MERTK IP in HEK-293 cell lysate. IHC-Fr: Human spleen tissue sections.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none">- High batch-to-batch consistency and reproducibility- Improved sensitivity and specificity- Long-term security of supply- Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p> <p>Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.</p>

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. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol, 0.05% BSA

Purity	Protein A purified
Clonality	Monoclonal
Clone number	Y323
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab52968 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

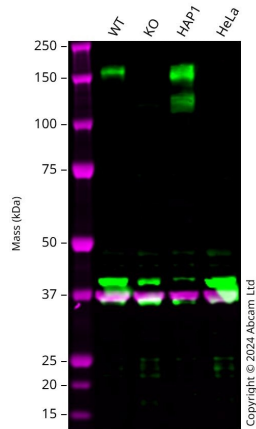
Application	Abreviews	Notes
WB	★★★★★ (1)	1/2000. Detects a band of approximately 180 kDa. For unpurified use at 1/500.
IP		1/20. For unpurified use at 1/50.
IHC-P	★★★★★ (3)	1/1000 - 1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See <u>IHC antigen retrieval protocols</u> .
IHC-Fr		Use at an assay dependent concentration.

Application notes Is unsuitable for Flow Cyt or ICC/IF.

Target

Function	In case of filovirus infection, seems to function as a cell entry factor.
Tissue specificity	Not expressed in normal B- and T-lymphocytes but is expressed in numerous neoplastic B- and T-cell lines.
Involvement in disease	Defects in MERTK are the cause of retinitis pigmentosa type 38 (RP38) [MIM:613862]. RP38 is a retinal dystrophy belonging to the group of pigmentary retinopathies. Retinitis pigmentosa is characterized by retinal pigment deposits visible on fundus examination and primary loss of rod photoreceptor cells followed by secondary loss of cone photoreceptors. Patients typically have night vision blindness and loss of midperipheral visual field. As their condition progresses, they lose their far peripheral visual field and eventually central vision as well.
Sequence similarities	Belongs to the protein kinase superfamily. Tyr protein kinase family. AXL/UFO subfamily. Contains 2 fibronectin type-III domains. Contains 2 Ig-like C2-type (immunoglobulin-like) domains. Contains 1 protein kinase domain.
Cellular localization	Membrane.

Images



Western blot - Anti-MERTK antibody [Y323] (ab52968)

All lanes : Anti-MERTK antibody [Y323] (ab52968) at 1/1000 dilution

Lane 1 : Wild-type A549 cell lysate

Lane 2 : MERTK knockout A549 cell lysate

Lane 3 : HAP1 cell lysate

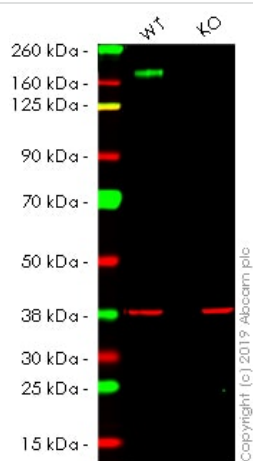
Lane 4 : HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 160 kDa

Western blot: Anti-MERTK antibody [Y323] (ab52968) staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (**ab8245**) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, ab52968 was shown to bind specifically to MERTK. A band was observed at 160 kDa in wild-type A549 cell lysates with no signal observed at this size in MERTK knockout cell line. To generate this image, wild-type and MERTK knockout A549 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.



Western blot - Anti-MERTK antibody [Y323]
(ab52968)

All lanes : Anti-MERTK antibody [Y323] (ab52968) at 1/500 dilution

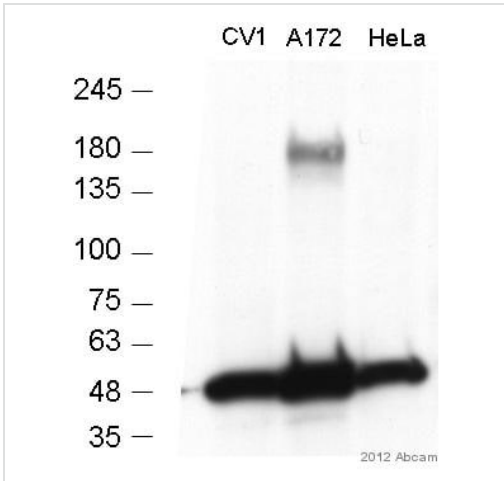
Lane 1 : Wild-type HAP1 whole cell lysate

Lane 2 : MERTK knockout HAP1 whole cell lysate

Lysates/proteins at 20 µg per lane.

Lanes 1 - 2: Merged signal (red and green). Green - ab52968 observed at 110 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab52968 was shown to specifically react with MERTK in wild-type HAP1 cells as signal was lost in MERTK knockout cells. Wild-type and MERTK knockout samples were subjected to SDS-PAGE. The membrane was blocked with 0. Ab52968 and **ab8245** (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/500 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed **ab216776** secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-MERTK antibody [Y323]
(ab52968)

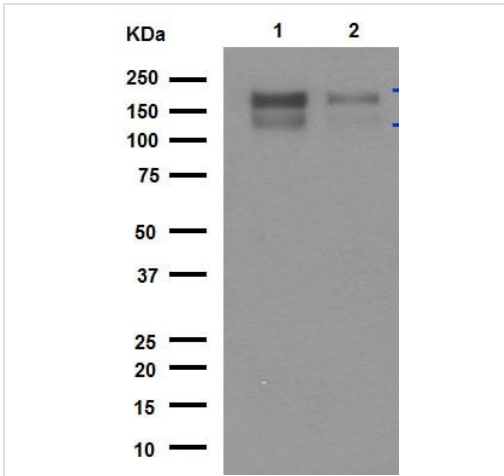
ab52968 used at a 1/200 dilution, 18 hours at 4°C, 5% BSA in Tween PBS.

Positive cell line: A172

Negative cell lines: CV1 and HeLa.

Secondary used is a goat anti-rabbit polyclonal-HRP at 1/10000 dilution.

Blocked with 5% milk for 1 hour at RT.



Western blot - Anti-MERTK antibody [Y323]
(ab52968)

All lanes : Anti-MERTK antibody [Y323] (ab52968) at 1/2000 dilution (purified)

Lane 1 : HEK-293 (Human epithelial cell line from embryonic kidney) cell lysate

Lane 2 : K562 (Human chronic myelogenous leukemia cell line from bone marrow) cell lysate

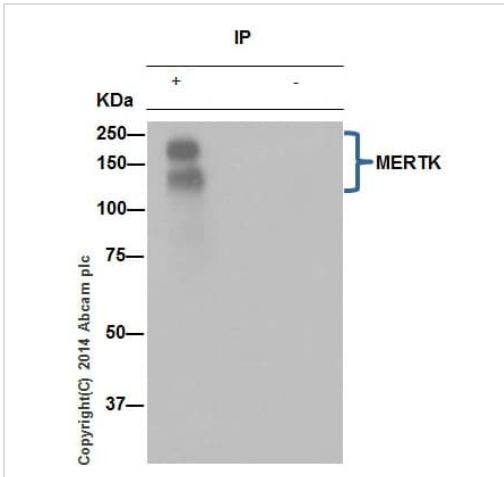
Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Peroxidase-conjugated goat anti-rabbit IgG at 1/1000 dilution

Observed band size: 140-180 kDa

Blocking/Dilution buffer and concentration: 5% NFDM/TBST.



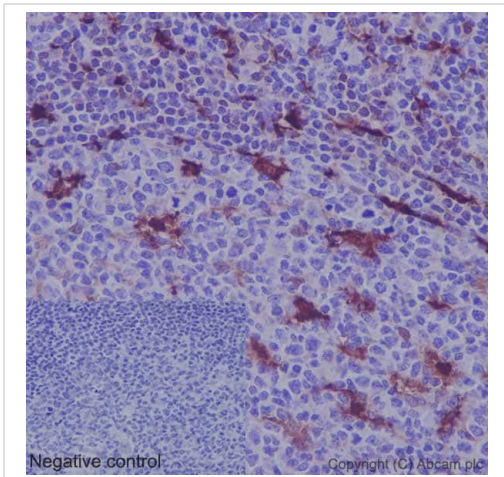
Immunoprecipitation - Anti-MERTK antibody [Y323] (ab52968)

Lane 1: ab52968 (purified) at 1/20 immunoprecipitating MERTK in HEK-293 (Human epithelial cell line from embryonic kidney) cell lysate.

Lane 2 - PBS.

For western blotting, a HRP-conjugated anti-rabbit IgG, specific to the non-reduced form of IgG was used as the secondary antibody (1/1500).

Blocking/Dilution buffer and concentration: 5% NFDm/TBST.

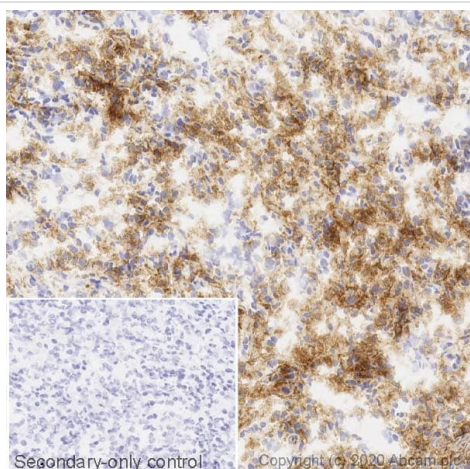


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MERTK antibody [Y323] (ab52968)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human tonsil tissue labeling MERTK with purified ab52968 at 1/500.

Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Counterstained with hematoxylin.

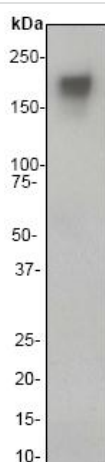
Negative control using PBS instead of primary antibody (Inset).



Immunohistochemistry (Frozen sections) - Anti-MERTK antibody [Y323] (ab52968)

IHC image of MERTK staining in a section of frozen normal human spleen performed on a Leica BOND™ system using the standard protocol. The section was fixed in 10% paraformaldehyde (10 min) prior to staining. The section was incubated with ab52968, 1/1000 dilution, 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. The inset secondary-only control image is taken from an identical assay without primary antibody.

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.



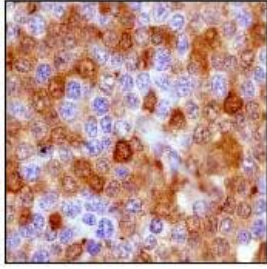
Western blot - Anti-MERTK antibody [Y323] (ab52968)

Anti-MERTK antibody [Y323] (ab52968) at 1/1000 dilution (unpurified) + HEK-293 (Human epithelial cell line from embryonic kidney) cell lysate at 10 µg

Secondary

HRP-conjugated goat anti-rabbit IgG at 1/2000 dilution

Observed band size: 180 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MERTK antibody [Y323] (ab52968)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human lymphoma tissue labeling MERTK with unpurified ab52968 at a 1/50 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Why choose a recombinant antibody?



Anti-MERTK antibody [Y323] (ab52968)

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