abcam

Product datasheet

Anti-Frizzled 6 antibody [EPR25319-149] ab290728

KO VALIDATED Recombinant RabMAb

6 Images

Overview

Product name	Anti-Frizzled 6 antibody [EPR25319-149]
Description	Rabbit monoclonal [EPR25319-149] to Frizzled 6
Host species	Rabbit
Tested applications	**Suitable for:** IHC-P, WB **Unsuitable for:** Flow Cyt,ICC/IF or IP
Species reactivity	**Reacts with:** Human **Does not react with:** Mouse, Rat
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HeLa and HCT 116 whole cell lysates. IHC-P: Human cervical carcinoma, Human endometrial carcinoma tissue, and HeLa cell pellet.
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	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR25319-149

Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab290728 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		1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		1/1000. Detects a band of approximately 79 kDa (predicted molecular weight: 79 kDa).

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

Target

Function Receptor for Wnt proteins. Most of frizzled receptors are coupled to the beta-catenin canonical signaling pathway, which leads to the activation of disheveled proteins, inhibition of GSK-3 kinase, nuclear accumulation of beta-catenin and activation of Wnt target genes. A second signaling pathway involving PKC and calcium fluxes has been seen for some family members, but it is not yet clear if it represents a distinct pathway or if it can be integrated in the canonical pathway, as PKC seems to be required for Wnt-mediated inactivation of GSK-3 kinase. Both pathways seem to involve interactions with G-proteins. May be involved in transduction and intercellular transmission of polarity information during tissue morphogenesis and/or in differentiated tissues. Together with FZD3, is involved in the neural tube closure and plays a role in the regulation of the establishment of planar cell polarity (PCP), particularly in the orientation of asymmetric bundles of stereocilia on the apical faces of a subset of auditory and vestibular sensory cells located in the inner ear.

Tissue specificity Detected in adult heart, brain, placenta, lung, liver, skeletal muscle, kidney, pancreas, thymus, prostate, testis, ovary, small intestine and colon. In the fetus, expressed in brain, lung, liver and kidney.

Involvement in disease Nail disorder, non-syndromic congenital, 10
Rare non-synonymous variants in FZD6 may contribute to neural tube defects, congenital malformations of the central nervous system and adjacent structures related to defective neural tube closure during the first trimester of pregnancy.

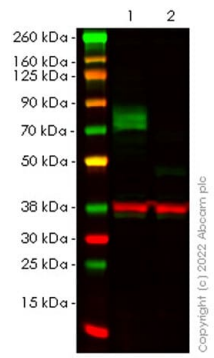
Sequence similarities Belongs to the G-protein coupled receptor Fz/Smo family.
Contains 1 FZ (frizzled) domain.

Domain Lys-Thr-X-X-X-Trp motif interacts with the PDZ doman of Dvl (Disheveled) family members and is involved in the activation of the Wnt/beta-catenin signaling pathway.
The FZ domain is involved in binding with Wnt ligands.

Post-translational modifications Ubiquitinated by ZNRF3, leading to its degradation by the proteasome.

Cellular localization Membrane. Cell membrane. Cell surface. Apical cell membrane. Cytoplasmic vesicle membrane. Colocalizes with FZD3 at the apical face of cells (By similarity).

Images



Western blot - Anti-Frizzled 6 antibody [EPR25319-149] (ab290728)

All lanes : Anti-Frizzled 6 antibody [EPR25319-149] (ab290728) at 1/1000 dilution

Lane 1 : Wild-type HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate
Lane 2 : FZD6 knockout HeLa (**ab265280**) whole cell lysate

Lysates/proteins at 20 µg per lane.

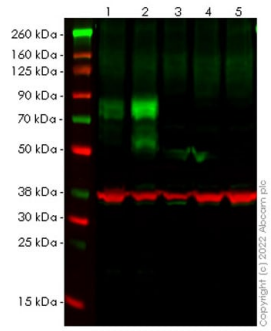
Secondary
All lanes : Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) at 1/10000 dilution

Predicted band size: 79 kDa
Observed band size: 79 kDa

Intercept® (TBS) Blocking Buffer diluted with an equal volume of 0.1% TBS was used as a blocking and diluting buffer and concentration.

Samples are non-boiled as boiling may cause protein aggregates. Lysates/proteins at 20 µg per lane.
Performed under reducing conditions.
False colour image of Western blot: Anti-FZD6 antibody [EPR25319-149] staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody 6C5 loading control staining at 1/20000 dilution, shown in red.
In Western blot, ab290728 was shown to bind specifically to FZD6. A band was observed at 79 kDa in wild-type Hela cell lysates with no signal observed at this size in FZD6 knockout cell line **ab265280** (knockout cell lysate **ab258874**). To generate this image, wild-type and FZD6 knockout Hela cell lysates were analyzed. First, samples were run on an SDS-PAGE gel then transferred onto an immobilon-FL PVDF membrane. Membranes were blocked in Intercept® (TBS) Blocking Buffer diluted with an equal volume of 0.1% TBS 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 (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) at 1/10000 dilution.



Western blot - Anti-Frizzled 6 antibody [EPR25319-149] (ab290728)

All lanes : Anti-Frizzled 6 antibody [EPR25319-149] (ab290728) at 1/1000 dilution

Lane 1 : HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate
Lane 2 : HCT 116 (human colorectal carcinoma epithelial cell) whole cell lysate
Lane 3 : K562 (human chronic myelogenous leukemia lymphoblast) whole cell lysate
Lane 4 : Daudi (human Burkitt's lymphoma lymphoblast) whole cell lysate
Lane 5 : Raji (human Burkitt's lymphoma B lymphocyte) whole cell lysate

Lysates/proteins at 20 µg per lane.

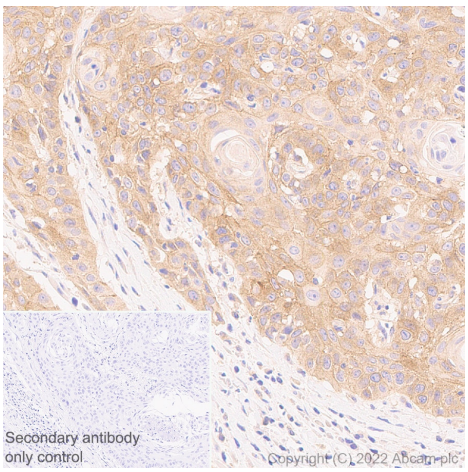
Secondary
All lanes : Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) at 1/10000 dilution

Performed under reducing conditions.

Predicted band size: 79 kDa
Observed band size: 79 kDa

Low expression: Daudi, Raji, K562 (PMID: 9480858).
Samples are non-boiled as boiling may cause protein aggregates.
Lysates/proteins at 20 µg per lane.
Performed under reducing conditions.
False colour image of Western blot: Anti-FZD6 antibody [EPR25319-149] staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody 6C5 loading control staining at 1/20000 dilution, shown in red.
First, samples were run on an SDS-PAGE gel then transferred onto an immobilon-FL PVDF membrane. Membranes were blocked in Intercept® (TBS) Blocking Buffer diluted with an equal volume of 0.1% TBS 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 (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) at 1/10000 dilution.

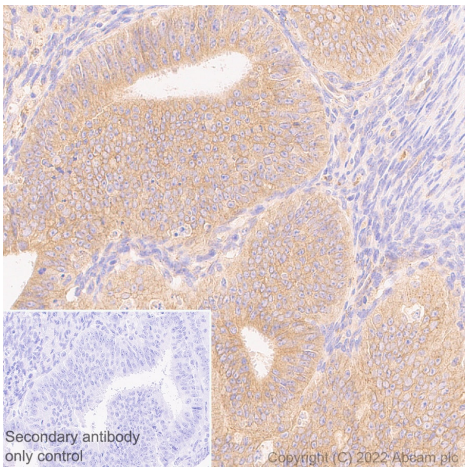


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Frizzled 6 antibody [EPR25319-149] (ab290728)

Immunohistochemical analysis of paraffin-embedded human cervical carcinoma tissue labelling Frizzled 6 with ab290728 at 1/100, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Cytoplasmic staining on human cervical carcinoma.
The section was incubated with ab290728 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody LeicaDS9800 (Bond™ Polymer Refine Detection).

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

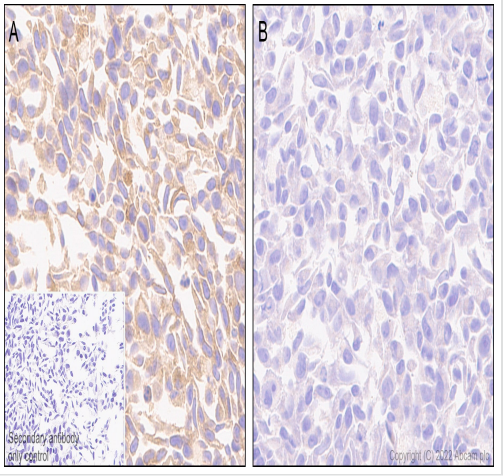


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Frizzled 6 antibody [EPR25319-149] (ab290728)

Immunohistochemical analysis of paraffin-embedded human endometrial carcinoma tissue labelling Frizzled 6 with ab290728 at 1/100, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Cytoplasmic staining on human endometrial carcinoma.
The section was incubated with ab290728 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody LeicaDS9800 (Bond™ Polymer Refine Detection).

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



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Frizzled 6 antibody [EPR25319-149] (ab290728)

Immunohistochemical analysis of paraffin-embedded (A) Wild-type HeLa (human cervix adenocarcinoma epithelial cell) cell pellet (B) FZD6 knockout HeLa (**ab265280**) cell pellet labelling Frizzled 6 with ab290728 at 1/200, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Cytoplasmic staining on human endometrial carcinoma.

The section was incubated with ab290728 for 30 mins at room temperature. Cytoplasmic staining on (A) Wild-type HeLa (human cervix adenocarcinoma epithelial cell) cell pellet (**ab255928**), no staining on (B) FZD6 knockout HeLa (**ab265280**) cell pellet.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody LeicaDS9800 (Bond™ Polymer Refine Detection).

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

Anti-Frizzled 6 antibody [EPR25319-149] (ab290728)

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