

**Product datasheet** 

# Anti-CD133 antibody [EPR20980-45] - BSA and Azide free ab226486

KO VALIDATED Recombinant RobMAb

4 Images

Overview	
Product name	Anti-CD133 antibody [EPR20980-45] - BSA and Azide free
Description	Rabbit monoclonal [EPR20980-45] to CD133 - BSA and Azide free
Host species	Rabbit
Tested applications	Suitable for: WB, IHC-P
Species reactivity	Reacts with: Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	IHC-P: Human pancreas tissue. WB: HAP1 and Caco-2 cell lysates.
General notes	ab226486 is the carrier-free version of ab226355.
	Our <b><u>carrier-free</u></b> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.
	This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.
	Use our <b>conjugation kits</b> for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.
	This product is compatible with the Maxpar <sup>®</sup> Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar <sup>®</sup> is a trademark of Fluidigm Canada Inc.
	<ul> <li>This product is a recombinant monoclonal antibody, which offers several advantages including:</li> <li>High batch-to-batch consistency and reproducibility</li> <li>Improved sensitivity and specificity</li> <li>Long-term security of supply</li> <li>Animal-free production</li> <li>For more information <u>see here</u>.</li> <li>Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <u>RabMAb<sup>®</sup> patents</u>.</li> </ul>

## Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR20980-45
lsotype	lgG

# Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab226486 in the following tested applications.

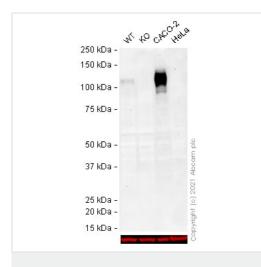
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Predicted molecular weight: 97 kDa.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Target	
Function	May play a role in cell differentiation, proliferation and apoptosis (PubMed:24556617). Binds cholesterol in cholesterol-containing plasma membrane microdomains and may play a role in the organization of the apical plasma membrane in epithelial cells. During early retinal development acts as a key regulator of disk morphogenesis. Involved in regulation of MAPK and Akt signaling pathways. In neuroblastoma cells suppresses cell differentiation such as neurite outgrowth in a RET-dependent manner (PubMed:20818439).
Tissue specificity	Isoform 1 is selectively expressed on CD34 hematopoietic stem and progenitor cells in adult and fetal bone marrow, fetal liver, cord blood and adult peripheral blood. Isoform 1 is not detected on other blood cells. Isoform 1 is also expressed in a number of non-lymphoid tissues including retina, pancreas, placenta, kidney, liver, lung, brain and heart. Found in saliva within small membrane particles. Isoform 2 is predominantly expressed in fetal liver, skeletal muscle, kidney, and heart as well as adult pancreas, kidney, liver, lung, and placenta. Isoform 2 is highly expressed in fetal liver, low in bone marrow, and barely detectable in peripheral blood. Isoform 2 is expressed on hematopoietic stem cells and in epidermal basal cells (at protein level). Expressed in adult retina by rod and cone photoreceptor cells (at protein level).
Involvement in disease	Retinitis pigmentosa 41 Cone-rod dystrophy 12 Stargardt disease 4 Retinal macular dystrophy 2

Sequence similarities	Belongs to the prominin family.
Post-translational modifications	lsoform 1 and isoform 2 are glycosylated. Acetylation at Lys-225, Lys-257 and Lys-264 by NAT8 and NAT8B may control PROM1 protein expression and its function in cell apoptosis.
Cellular localization	Apical cell membrane. Cell projection, microvillus membrane. Cell projection, cilium, photoreceptor outer segment. Endoplasmic reticulum. Endoplasmic reticulum-Golgi intermediate compartment. Found in extracellular membrane particles in various body fluids such as cerebrospinal fluid, saliva, seminal fluid and urine.

#### Images



Western blot - Anti-CD133 antibody [EPR20980-45]

- BSA and Azide free (ab226486)

All lanes : Anti-CD133 antibody [EPR20980-45] (<u>ab226355</u>) at 1/1000 dilution

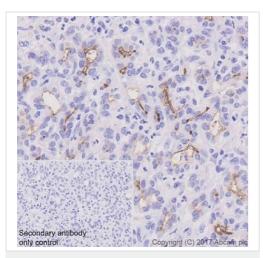
Lane 1 : Wild-type HAP1 cell lysate at 40 µg Lane 2 : PROM1 knockout HAP1 cell lysate at 40 µg Lane 3 : Caco-2 cell lysate at 20 µg Lane 4 : HeLa cell lysate at 20 µg

Performed under reducing conditions.

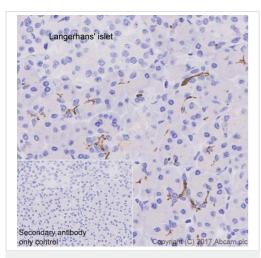
Predicted band size: 97 kDa Observed band size: 110 kDa

Exposure time: 20 minutes

ab226355 was shown to react with CD133 in wild-type HAP1 cells in western blot. Loss of signal was observed when PROM1 knockout sample was used. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween<sup>®</sup>) before incubation with <u>ab226355</u> overnight at 4°C at a 1 in 1000 dilution. Blots were incubated with HRP conjugated Goat anti-Rabbit (H+L) secondary antibody at 1/5000 for 1 hour at room temperature before development with ECL reagent and imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD133 antibody [EPR20980-45] - BSA and Azide free (ab226486)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD133 antibody [EPR20980-45] - BSA and Azide free (ab226486)

Immunohistochemical analysis of paraffin-embedded human pancreatic cancer tissue labeling CD133 with <u>ab226355</u> at 1/1000 dilution, followed by a Goat Anti-Rabbit IgG H&L (HRP), ready to use. Apical/endoluminal staining on ducts of human pancreatic cancer (PMID: 18261235) is observed. Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a Goat Anti-Rabbit IgG H&L (HRP), ready to use.

Perform heat mediated antigen retrieval using <u>ab93684</u> (Tris/EDTA buffer, pH 9.0).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab226355</u>).

Immunohistochemical analysis of paraffin-embedded human pancreas tissue labeling CD133 with <u>ab226355</u> at 1/1000 dilution, followed by a Goat Anti-Rabbit IgG H&L (HRP), ready to use. Apical/endoluminal staining on ducts of human pancreas, and negative on Langerhans cells of the islets (PMID: 18261235) is observed. Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a Goat Anti-Rabbit IgG H&L (HRP), ready to use.

Perform heat mediated antigen retrieval using <u>ab93684</u> (Tris/EDTA buffer, pH 9.0).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab226355</u>).



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