

Product datasheet

Anti-CD166 antibody [EPR2759(2)] α b109215

KO **VALIDATED** Recombinant RabMAb

★★★★☆ **3 Abreviews** **22 References** **11 Images**

Overview

Product name	Anti-CD166 antibody [EPR2759(2)]
Description	Rabbit monoclonal [EPR2759(2)] to CD166
Host species	Rabbit
Tested applications	Suitable for: WB, IP, IHC-P, Flow Cyt, ICC/IF
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: SH-SY5Y, HuT-78, HT1080, Daudi and HeLa whole cell lysate (ab150035). Mouse and rat brain tissue lysates; Wild-type HAP1 whole cell lysate. IHC-P: Human liver and prostatic adenocarcinoma tissues. ICC/IF: THP-1 cells. Flow Cyt: HuT-78 cells. IP: SH-SY5Y cell lysate.
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>

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. Stable for 12 months at -20°C.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 40% Glycerol, 0.05% BSA, 59% PBS
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR2759(2)

Isotype

IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab109215 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)	1/10000 - 1/20000. Detects a band of approximately 100-105 kDa (predicted molecular weight: 65 kDa). For unpurified use at 1/1000 - 1/10000.
IP	★★★★★ (1)	1/30. For unpurified use at 1/10 - 1/100.
IHC-P	★★★★★ (1)	1/50. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols . For unpurified use at 1/100 - 1/250.
Flow Cyt		1/90. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		1/50 - 1/250.

Target

Function

Cell adhesion molecule that binds to CD6. Involved in neurite extension by neurons via heterophilic and homophilic interactions. May play a role in the binding of T- and B-cells to activated leukocytes, as well as in interactions between cells of the nervous system.

Tissue specificity

Spleen, placenta, liver, and weakly in liver. Expressed by activated T-cells, B-cells, monocytes and thymic epithelial cells. Expressed by neurons in the brain. Restricted expression in tumor cell lines. Preferentially expressed in highly metastasizing melanoma cell lines.

Sequence similarities

Contains 3 Ig-like C2-type (immunoglobulin-like) domains.
Contains 2 Ig-like V-type (immunoglobulin-like) domains.

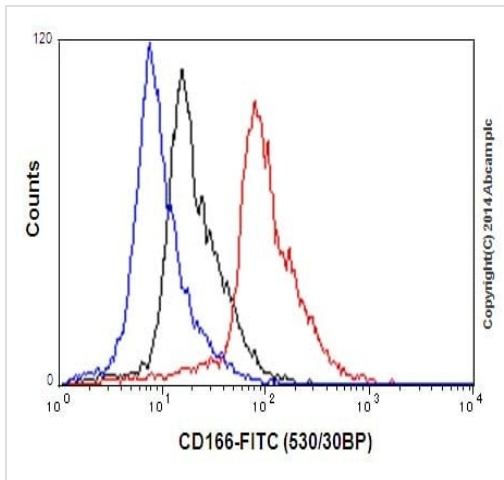
Domain

The CD6 binding site is located in the N-terminal Ig-like domain.

Cellular localization

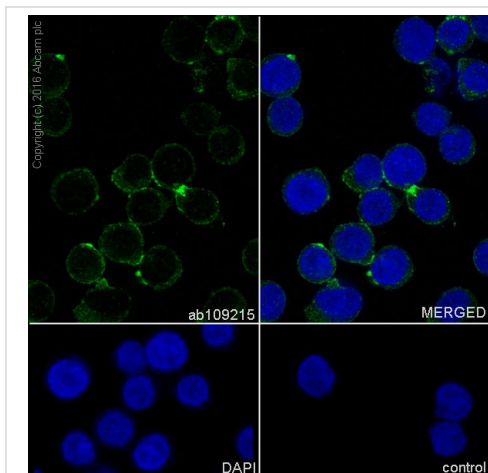
Membrane.

Images



Flow Cytometry - Anti-CD166 antibody [EPR2759(2)]
(ab109215)

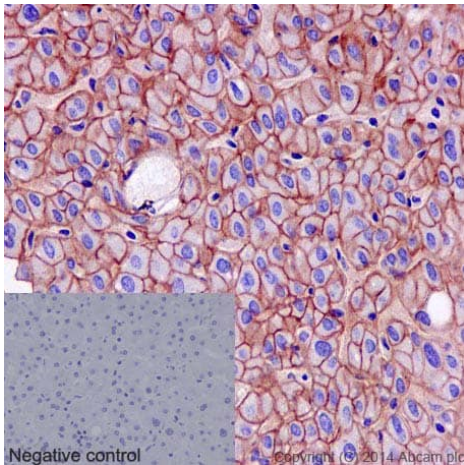
Flow Cytometry analysis of HuT-78 cells labelling CD166 with purified ab109215 at 1/90 (red). Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



Immunocytochemistry/ Immunofluorescence - Anti-
CD166 antibody [EPR2759(2)] (ab109215)

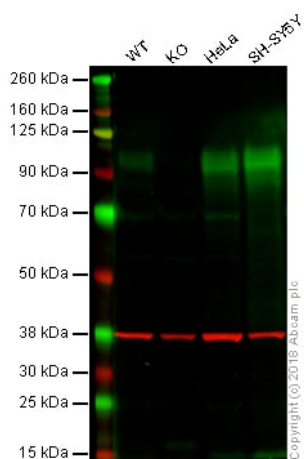
Immunocytochemistry/Immunofluorescence analysis of THP-1 (human monocytic leukemia cell line) cells labelling CD166 (green) with purified ab109215 at 1/250. Cells were fixed with 100% methanol. **ab150077**, Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. DAPI (blue) was used as a nuclear counterstain.

Secondary Only Control: PBS was used instead of the primary antibody as the negative control.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD166 antibody [EPR2759(2)] (ab109215)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human liver tissue labelling CD166 with purified ab109215 at 1/50. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. A prediluted HRP-polymer conjugated anti-rabbit IgG was used as the secondary antibody. Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Western blot - Anti-CD166 antibody [EPR2759(2)] (ab109215)

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)

Lane 2: CD166 knockout HAP1 whole cell lysate (20 µg)

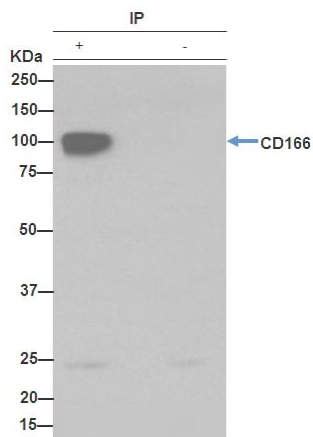
Lane 3: HeLa whole cell lysate (20 µg)

Lane 4: SH-SY5Y whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab109215 observed at 100 kDa. Red - loading control, [ab9484](#), observed at 37 kDa.

ab109215 was shown to specifically react with CD166 in wild-type HAP1 cells as signal was lost in CD166 knockout cells. Wild-type and CD166 knockout samples were subjected to SDS-PAGE.

Ab109215 and [ab9484](#) (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed [ab216772](#) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed [ab216777](#) secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

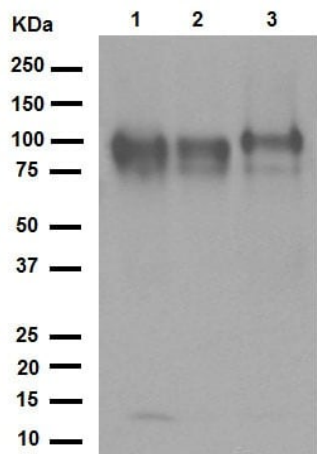


Immunoprecipitation - Anti-CD166 antibody
[EPR2759(2)] (ab109215)

ab109215 (purified) at 1/30 immunoprecipitating CD166 in SH-SY5Y (human neuroblastoma cell line from bone marrow) cell lysate (Lane 1). 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 buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Western blot - Anti-CD166 antibody [EPR2759(2)]
(ab109215)

All lanes : Anti-CD166 antibody [EPR2759(2)] (ab109215) at 1/10000 dilution (purified)

Lane 1 : SH-SY5Y (human neuroblastoma cell line from bone marrow) cell lysate

Lane 2 : HuT-78 cell lysate

Lane 3 : HT-1080 (human fibrosarcoma cell line) cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

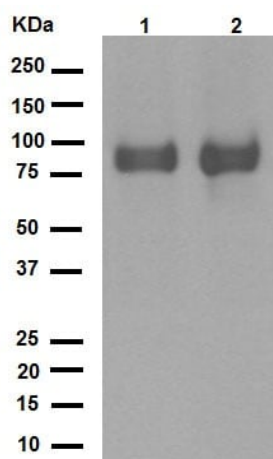
All lanes : Peroxidase conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 65 kDa

Observed band size: 100-105 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Western blot - Anti-CD166 antibody [EPR2759(2)] (ab109215)

All lanes : Anti-CD166 antibody [EPR2759(2)] (ab109215) at 1/10000 dilution (purified)

Lane 1 : Mouse brain tissue lysate

Lane 2 : Rat brain tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

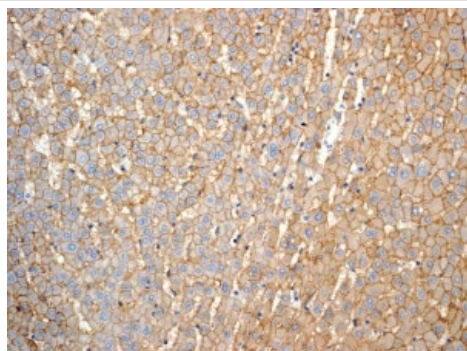
All lanes : Peroxidase conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 65 kDa

Observed band size: 100-105 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

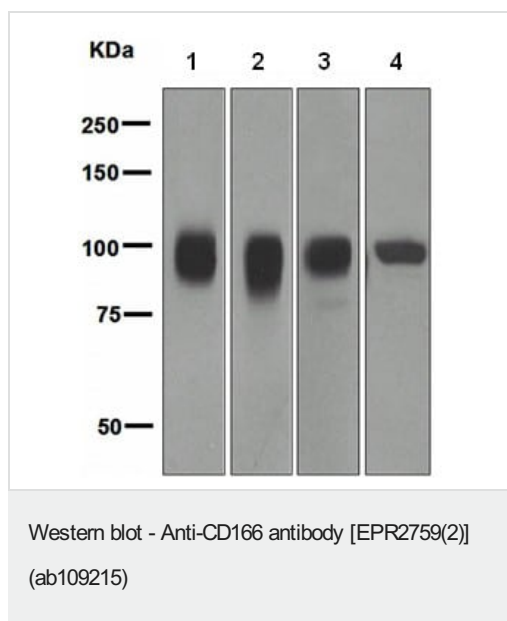
Diluting buffer and concentration: 5% NFDM /TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD166 antibody [EPR2759(2)] (ab109215)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human liver tissue labelling CD166 with unpurified ab109215 at 1/100.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



All lanes : Anti-CD166 antibody [EPR2759(2)] (ab109215) at 1/1000 dilution (unpurified)

Lane 1 : SH-SY5Y (human neuroblastoma cell line from bone marrow) cell lysate

Lane 2 : HuT-78 cell lysate

Lane 3 : HT1080 (human fibrosarcoma cell line) cell lysate

Lane 4 : Daudi (human Burkitt's lymphoma cell line) cell lysate

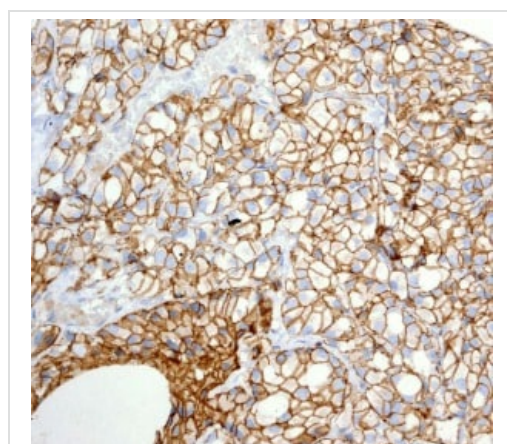
Lysates/proteins at 10 µg per lane.

Secondary

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

Predicted band size: 65 kDa

Observed band size: 100-105 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human prostatic adenocarcinoma tissue labelling CD166 with unpurified ab109215 at 1/100.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-CD166 antibody [EPR2759(2)] (ab109215)

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