# abcam

## Product datasheet

## Anti-PEG10/EDR antibody [EPR20051] ab215035





#### 2 References 11 Images

## Overview

**Product name** Anti-PEG10/EDR antibody [EPR20051]

Rabbit monoclonal [EPR20051] to PEG10/EDR **Description** 

**Host species** Rabbit

**Tested applications** Suitable for: Flow Cyt (Intra), ICC/IF, IP, IHC-P, WB

Species reactivity Reacts with: Human

**Immunogen** Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: HEK 293, HeLa and HepG2 whole cell lysates; Human testis lysate. IHC-P: Human placenta,

liver cancer and testis tissues. ICC/IF: HepG2 cells. Flow Cyt (intra): HepG2 cells. IP: HepG2

whole cell lysate.

**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: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

**Purity** Protein A purified

Clonality Monoclonal Clone number EPR20051

**Isotype** IgG

## **Applications**

The Abpromise guarantee

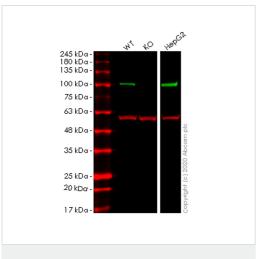
Our <u>Abpromise guarantee</u> covers the use of ab215035 in the following tested applications.

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

Application	Abreviews	Notes
Flow Cyt (Intra)		1/500.
ICC/IF		1/500.
IP		1/30.
IHC-P		1/500. 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 100 kDa (predicted molecular weight: 30,80 kDa).

Target		
Function	Prevents apoptosis in hepatocellular carcinoma (HCC) cells through interaction with SIAH1, a mediator of apoptosis. May also have a role in cell growth promotion and hepatoma formation. Inhibits the TGF-beta signaling by interacting with the TGF-beta receptor ALK1. When overexpressed, induces the formation of cellular extension, such as filipodia in association with ALK1. Involved at the immediate early stage of adipocyte differentiation (By similarity). May bind to the 5'-GCCTGTCTTT-3' DNA sequence of the MB1 domain in the myelin basic protein (MBP) promoter.	
Tissue specificity	Expressed in the cytotrophoblast layer but not in the overlying syncytiotrophoblast of the placenta. Expressed in prostate and breast carcinomas but not in normal breast and prostate epithelial cells. Expressed in the HepG2 cell line (at protein level). Expressed in brain, liver, spleen, kidney, thymus, lung, ovary, testis, reactive lymph node, skeletal muscle, adipose tissue and placenta. Expressed in pancreatic and hepatocellular carcinomas (HCC).	
Sequence similarities	Contains 1 CCHC-type zinc finger.	
Developmental stage	Expressed in placenta during the first trimester of gestation (at protein level). In placenta, down-regulated at early hypoxic phase, and highly activated at 11-12 week of gestation.	
Post-translational modifications	Isoform RF1/RF2 undergoes proteolytic cleavage.	
Cellular localization	Nucleus. Cytoplasm. Detected predominantly in the cytoplasm of breast and prostate carcinomas, in hepatocellular carcinoma (HCC) and B-cell chronic lymphocytic leukemia (B-CLL) cells and in the HepG2 cell line. Colocalized with ALK1.	

## **Images**



Western blot - Anti-PEG10/EDR antibody [EPR20051] (ab215035)

**All lanes :** Anti-PEG10/EDR antibody [EPR20051] (ab215035) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: PEG10 knockout HeLa cell lysate

Lane 3: HepG2 cell lysate

Lysates/proteins at 20 µg per lane.

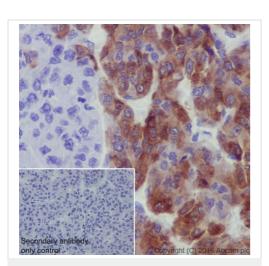
## **Secondary**

All lanes: Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

**Predicted band size:** 30,80 kDa **Observed band size:** 100 kDa

**Lanes 1-4:** Merged signal (red and green). Green - ab215035 observed at 100 kDa. Red - loading control <u>ab7291</u> observed at 50 kDa.

ab215035 Anti-PEG10/EDR antibody [EPR20051] was shown to specifically react with PEG10/EDR in wild-type HeLa cells. Loss of signal was observed when knockout cell line <a href="mailto:ab265925">ab265925</a> (knockout cell lysate <a href="mailto:ab258103">ab258103</a>) was used. Wild-type and PEG10/EDR knockout samples were subjected to SDS-PAGE. ab215035 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (<a href="mailto:ab7291">ab7291</a>) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<a href="mailto:ab216776">ab216773</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

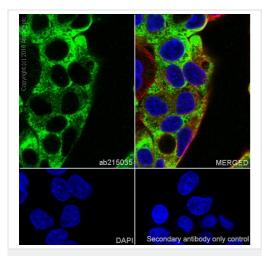


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PEG10/EDR antibody
[EPR20051] (ab215035)

Immunohistochemical analysis of paraffin-embedded human liver cancer tissue labeling PEG10/EDR with ab215035 at 1/500 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) at 1/500 dilution. Cytoplasmic staining on part of the cells in human liver cancer is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) at 1/500 dilution.

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

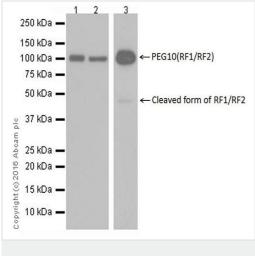


Immunocytochemistry/ Immunofluorescence - Anti-PEG10/EDR antibody [EPR20051] (ab215035)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HepG2 (Human liver hepatocellular carcinoma cell line) cells labeling PEG10/EDR with ab215035 at 1/500 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining on HepG2 cell line.

The nuclear counter stain is DAPI (blue). Tubulin is detected with <a href="mailto:ab195889"><u>ab195889</u></a> (Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor<sup>®</sup> 594)) at 1/200 dilution (red).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit lgG (Alexa Fluor<sup>®</sup> 488) (ab150077) at 1/1000 dilution.



Western blot - Anti-PEG10/EDR antibody [EPR20051] (ab215035) **Lanes 1-2**: Anti-PEG10/EDR antibody [EPR20051] (ab215035) at 1/2000 dilution

**Lane 3**: Anti-PEG10/EDR antibody [EPR20051] (ab215035) at 1/10000 dilution

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

**Lane 2**: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

**Lane 3 :** HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate

Lysates/proteins at 10 µg per lane.

## **Secondary**

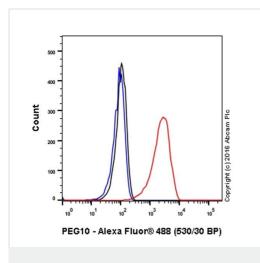
**All lanes :** Goat Anti-Rabbit  $\lg G \ H\&L \ (HRP) \ (\underline{ab97051})$  at 1/100000 dilution

**Predicted band size:** 30,80 kDa **Observed band size:** 100 kDa

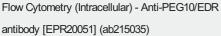
Exposure time: 3 minutes

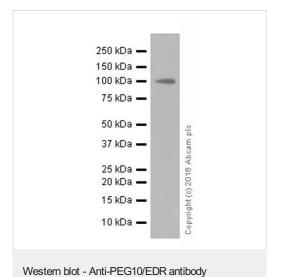
Blocking/Dilution buffer: 5% NFDM/TBST.

The expression of PEG10/EDR in HepG2 cells is consistent with what has been described in the literature PMID: 17942406.



Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed HepG2 (Human liver hepatocellular carcinoma cell line) cells labeling PEG10/EDR with ab215035 at 1/500 dilution (red) compared with a rabbit monoclonal lgG isotype control (ab172730; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit lgG (Alexa Fluor<sup>®</sup> 488) at 1/2000 dilution was used as the secondary antibody.





[EPR20051] (ab215035)

Anti-PEG10/EDR antibody [EPR20051] (ab215035) at 1/1000 dilution + Human testis lysate at 10 µg

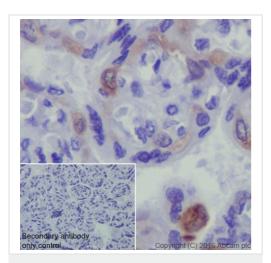
## **Secondary**

Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/100000 dilution

**Predicted band size:** 30,80 kDa **Observed band size:** 100 kDa

Exposure time: 1 minute

Blocking/Dilution buffer: 5% NFDM/TBST.

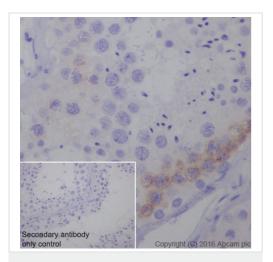


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PEG10/EDR antibody [EPR20051] (ab215035)

Immunohistochemical analysis of paraffin-embedded human placenta tissue labeling PEG10/EDR with ab215035 at 1/500 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) at 1/500 dilution. Cytoplasmic staining on some cells in human placenta is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) at 1/500 dilution.

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

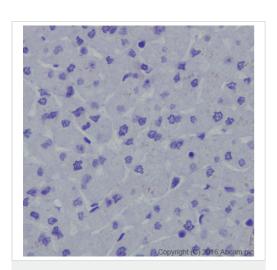


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PEG10/EDR antibody
[EPR20051] (ab215035)

Immunohistochemical analysis of paraffin-embedded human testis tissue labeling PEG10/EDR with ab215035 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) at 1/500 dilution. Cytoplasmic staining on the germ cells in human testis is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) at 1/500 dilution.

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



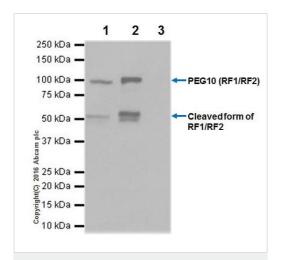
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PEG10/EDR antibody [EPR20051] (ab215035)

Immunohistochemical analysis of paraffin-embedded human liver tissue labeling PEG10/EDR with ab215035 at 1/500 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) at 1/500 dilution.

Negative staining on normal cells in human liver.

Counter stained with Hematoxylin.

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



Immunoprecipitation - Anti-PEG10/EDR antibody [EPR20051] (ab215035)

PEG10/EDR was immunoprecipitated from 0.35 mg of HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate with ab215035 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab215035 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10,000 dilution

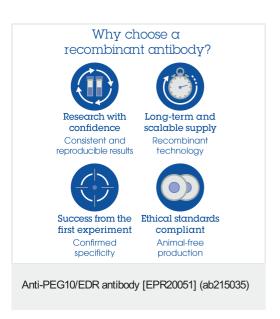
Lane 1: HepG2 whole cell lysate, 10 µg (Input).

Lane 2: ab215035 IP in HepG2 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab215035 in HepG2 whole cell lysate.

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

Exposure time: 3 seconds.



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