abcam

Product datasheet

Anti-Smadl antibody [EPR5522] ab126761





RabMAb

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Overview

Product name Anti-Smad1 antibody [EPR5522]

Description Rabbit monoclonal [EPR5522] to Smad1

Host species Rabbit

Tested applications Suitable for: WB, IHC-P, Flow Cyt (Intra)

Unsuitable for: IP

Species reactivity Reacts with: Human

Immunogen Synthetic peptide within Human Smad1 aa 200-300. The exact sequence is proprietary.

Positive control WB: HeLa, HT1080, HUVEC, A673, HepG2, and 293T cell lysates. IHC-P: Human colon tissue.

Flow Cyt (intra): HeLa cells

General notesThis 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**.

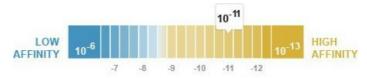
Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Stable for 12 months at -20°C.

Dissociation constant (K_D) $K_D = 2.56 \times 10^{-11} M$



Learn more about K_D

Storage buffer pH: 7.20

1

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.5% BSA

Purity Protein A purified

ClonalityMonoclonalClone numberEPR5522

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab126761 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		1/1000 - 1/10000. Detects a band of approximately 58 kDa (predicted molecular weight: 52 kDa).
IHC-P		1/50 - 1/100. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
Flow Cyt (Intra)		1/400. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.

Application notes Is unsuitable for IP.

Target

Function Transcriptional modulator activated by BMP (bone morphogenetic proteins) type 1 receptor

kinase. SMAD1 is a receptor-regulated SMAD (R-SMAD). SMAD1/OAZ1/PSMB4 complex

mediates the degradation of the CREBBP/EP300 repressor SNIP1.

Tissue specificity Ubiquitous. Highest expression seen in the heart and skeletal muscle.

Sequence similarities Belongs to the dwarfin/SMAD family.

Contains 1 MH1 (MAD homology 1) domain. Contains 1 MH2 (MAD homology 2) domain.

Post-translational

Phosphorylated on serine by BMP type 1 receptor kinase.

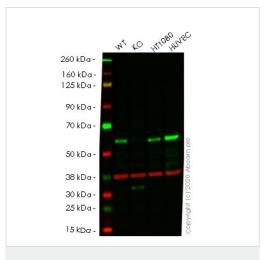
modifications

 $\label{thm:continuous} \mbox{Ubiquitin-mediated proteolysis by SMAD-specific E3 ubiquitin ligase SMURF1}.$

Cellular localization Cytoplasm. Nucleus. Cytoplasmic in the absence of ligand. Migrates to the nucleus when

complexed with SMAD4. Co-localizes with LEMD3 at the nucleus inner membrane.

Images



Western blot - Anti-Smad1 antibody [EPR5522] (ab126761)

All lanes : Anti-Smad1 antibody [EPR5522] (ab126761) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: SMAD1 knockout HeLa cell lysate

Lane 3 : HT1080 cell lysate

Lane 4 : Huvec cell lysate

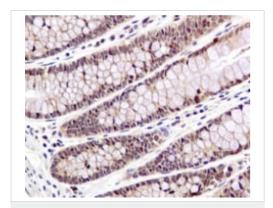
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 52 kDa Observed band size: 52 kDa

Lanes 1-4: Merged signal (red and green). Green - ab126761 observed at 52 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.

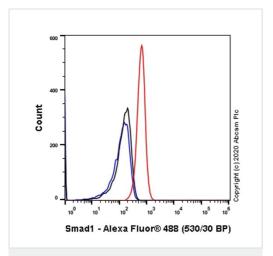
ab126761 was shown to react with Smad1 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab265400 (knockout cell lysate ab257686) was used. Wild-type HeLa and SMAD1 knockout HeLa cell lysates were subjected to SDS-PAGE. ab126761 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at a 1 in 1000 Dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Smad1 antibody
[EPR5522] (ab126761)

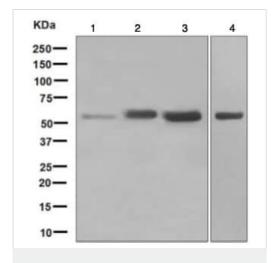
ab126761, at 1/50 dilution, staining Smad1 in paraffin-embedded Human colon tissue by Immunohistochemistry.

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



Flow Cytometry (Intracellular) - Anti-Smad1 antibody [EPR5522] (ab126761)

Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Smad1 with purified ab126761 at 1/400 dilution (1 µg/mL) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor® 488, <u>ab150077</u>) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal lgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Western blot - Anti-Smad1 antibody [EPR5522] (ab126761)

All lanes : Anti-Smad1 antibody [EPR5522] (ab126761) at 1/1000 dilution

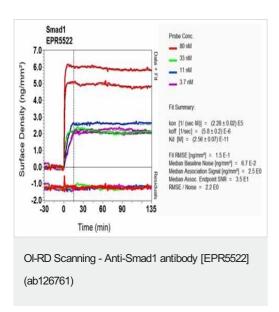
Lane 1 : HeLa cell lysate
Lane 2 : A673 cell lysate
Lane 3 : HepG2 cell lysate
Lane 4 : 293T cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

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

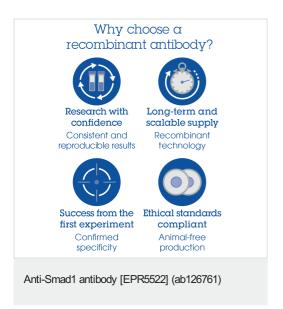
Predicted band size: 52 kDa



Equilibrium disassociation constant (K_D)

Click here to learn more about K_D

Learn more about K_D



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