

Product datasheet

Alexa Fluor® 647 Anti-Smad2 antibody [EP567Y] ab196538

Recombinant RabMAb

★★★★★ <u>1 Abreviews</u> 3 Images

Overview	
Product name	Alexa Fluor® 647 Anti-Smad2 antibody [EP567Y]
Description	Alexa Fluor® 647 Rabbit monoclonal [EP567Y] to Smad2
Host species	Rabbit
Conjugation	Alexa Fluor® 647. Ex: 652nm, Em: 668nm
Tested applications	Suitable for: Flow Cyt (Intra), ICC/IF
Species reactivity	Reacts with: Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	ICC/IF: Jurkat cells. Flow Cyt (intra): Jurkat cells.
General notes	Our RabMAb [®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <u>RabMAb[®] patents</u> .
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Properties

Form Storage instructions Liquid

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

	Avoid freeze / thaw cycle. Store In the Dark.
Storage buffer	pH: 7.40
	Preservative: 0.02% Sodium azide
	Constituents: PBS, 30% Glycerol (glycerin, glycerine), 1% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP567Y
lsotype	lgG

Applications

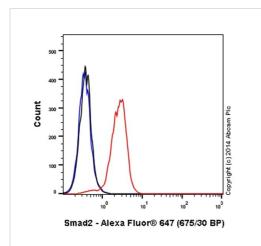
The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab196538 in the following tested applications.

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. <u>ab199093</u> - Rabbit monoclonal IgG (Alexa Fluor® 647), is suitable for use as an isotype control with this antibody.
ICC/IF		1/50.

Target	
Function	Receptor-regulated SMAD (R-SMAD) that is an intracellular signal transducer and transcriptional modulator activated by TGF-beta (transforming growth factor) and activin type 1 receptor kinases. Binds the TRE element in the promoter region of many genes that are regulated by TGF-beta and, on formation of the SMAD2/SMAD4 complex, activates transcription. May act as a tumor suppressor in colorectal carcinoma.
Tissue specificity	Expressed at high levels in skeletal muscle, heart and placenta.
Sequence similarities	Belongs to the dwarfin/SMAD family. Contains 1 MH1 (MAD homology 1) domain. Contains 1 MH2 (MAD homology 2) domain.
Post-translational modifications	Phosphorylated on one or several of Thr-220, Ser-245, Ser-250, and Ser-255. In response to TGF-beta, phosphorylated on Ser-465/467 by TGF-beta and activin type 1 receptor kinases. Able to interact with SMURF2 when phosphorylated on Ser-465/467, recruiting other proteins, such as SNON, for degradation. In response to decorin, the naturally occurring inhibitor of TGF-beta signaling, phosphorylated on Ser-240 by CaMK2. Phosphorylated by MAPK3 upon EGF stimulation; which increases transcriptional activity and stability, and is blocked by calmodulin. In response to TGF-beta, ubiquitinated by NEDD4L; which promotes its degradation. Acetylated on Lys-19 by coactivators in response to TGF-beta signaling, which increases transcriptional activity. Isoform short: Acetylation increases DNA binding activity in vitro and enhances its association with target promoters in vivo. Acetylation in the nucleus by EP300 is enhanced by TGF-beta.
Cellular localization	Cytoplasm. Nucleus. Cytoplasmic and nuclear in the absence of TGF-beta. On TGF-beta stimulation, migrates to the nucleus when complexed with SMAD4. On dephosphorylation by

Images



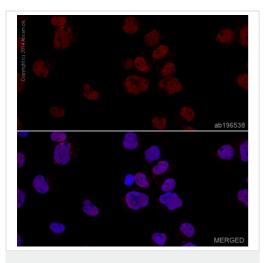
Flow Cytometry (Intracellular) - Alexa Fluor® 647 Anti-Smad2 antibody [EP567Y] (ab196538) Overlay histogram showing Jurkat cells stained with ab196538 (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab196538, 1/500 dilution) for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) Alexa Fluor® 647 used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5,000 events were collected using a solid-state 25mW red diode laser (635 nm) and 675/30 bandpass filter.

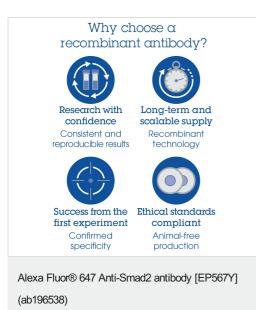
This antibody gave a positive signal in Jurkat cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

ab196538 staining Smad2 in Jurkat cells. The cells were fixed with 4% formaldehyde (10 min), permeabilised in 0.1% Triton X-100 for 5 minutes and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated with ab196538 at 1/50 dilution (shown in red) overnight at +4°C. Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Immunocytochemistry/ Immunofluorescence - Alexa Fluor® 647 Anti-Smad2 antibody [EP567Y] (ab196538)



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