abcam

Product datasheet

Alexa Fluor® 647 Anti-SF2 antibody [EPR8239] ab197878

Recombinant

RabMAb

3 Images

Overview

Product name Alexa Fluor® 647 Anti-SF2 antibody [EPR8239]

Description Alexa Fluor® 647 Rabbit monoclonal [EPR8239] to SF2

Host species Rabbit

Conjugation Alexa Fluor® 647. Ex: 652nm, Em: 668nm

Tested applications Suitable for: ICC/IF, Flow Cyt (Intra)

Species reactivity Reacts with: Human

Predicted to work with: Mouse, Rat

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control ICC/IF: HepG2 cells. Flow Cyt (intra): HepG2 cells.

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**.

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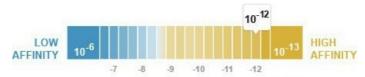
Properties

Form Liquid

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

Avoid freeze / thaw cycle. Store In the Dark.

Dissociation constant (K_D) $K_D = 8.00 \times 10^{-12} M$



Learn more about K_D

Storage buffer pH: 7.40

Preservative: 0.02% Sodium azide

Constituents: 30% Glycerol (glycerin, glycerine), 1% BSA, PBS

Purity Protein A purified

ClonalityMonoclonalClone numberEPR8239

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab197878 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
ICC/IF		1/100. This product gave a positive signal in HepG2 cells fixed with 4% formaldehyde (10 min)
Flow Cyt (Intra)		1/50. ab199093 - Rabbit monoclonal IgG (Alexa Fluor® 647), is suitable for use as an isotype control with this antibody.

Target

Function

Plays a role in preventing exon skipping, ensuring the accuracy of splicing and regulating alternative splicing. Interacts with other spliceosomal components, via the RS domains, to form a bridge between the 5'- and 3'-splice site binding components, U1 snRNP and U2AF. Can stimulate binding of U1 snRNP to a 5'-splice site-containing pre-mRNA. Binds to purine-rich RNA sequences, either the octamer, 5'-RGAAGAAC-3' (r=A or G) or the decamers,

AGGACAGACC/AGGACGAAGC. Binds preferentially to the 5'-CGAGGCG-3' motif in vitro. Three copies of the octamer constitute a powerful splicing enhancer in vitro, the ASF/SF2 splicing enhancer (ASE) which can specifically activate ASE-dependent splicing. Isoform ASF-2 and isoform ASF-3 act as splicing repressors.

Sequence similarities

Belongs to the splicing factor SR family.

Contains 2 RRM (RNA recognition motif) domains.

Domain

The RRM 2 domain plays an important role in governing both the binding mode and the phosphorylation mechanism of the RS domain by SRPK1. RS domain and RRM 2 are uniquely positioned to initiate a highly directional (C-terminus to N-terminus) phosphorylation reaction in which the RS domain slides through an extended electronegative channel separating the docking groove of SRPK1 and the active site. RRM 2 binds toward the periphery of the active site and guides the directional phosphorylation mechanism. Both the RS domain and an RRM domain are required for nucleocytoplasmic shuttling.

Post-translational modifications

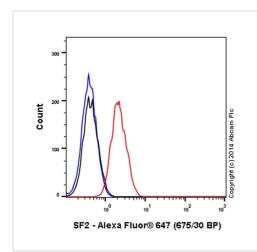
Phosphorylated by CLK1, CLK2, CLK3 and CLK4. Phosphorylated by SRPK1 at multiple serines in its RS domain via a directional (C-terminal to N-terminal) and a dual-track mechanism incorporating both processive phosphorylation (in which the kinase stays attached to the substrate after each round of phosphorylation) and distributive phosphorylation steps (in which the kinase and substrate dissociate after each phosphorylation event). The RS domain of SRSF1 binds to a docking groove in the large lobe of the kinase domain of SRPK1 and this induces certain structural changes in SRPK1 and/or RRM 2 domain of SRSF1, allowing RRM 2 to bind the kinase and initiate phosphorylation. The cycles continue for several phosphorylation steps in a processive manner (steps 1-8) until the last few phosphorylation steps (approximately steps 9-12). During that time, a mechanical stress induces the unfolding of the beta-4 motif in RRM 2, which then docks at the docking groove of SRPK1. This also signals RRM 2 to begin to dissociate, which facilitates SRSF1 dissociation after phosphorylation is completed.

Arg-97 is dimethylated, probably to asymmetric dimethylarginine.

Cellular localization

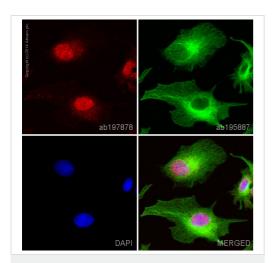
Cytoplasm. Nucleus speckle. In nuclear speckles. Shuttles between the nucleus and the cytoplasm.

Images



Flow Cytometry (Intracellular) - Alexa Fluor® 647 Anti-SF2 antibody [EPR8239] (ab197878) Overlay histogram showing HepG2 cells stained with ab197878 (red line). The cells were fixed with 80% methanol (5 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 (ab197878, 1/50 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.

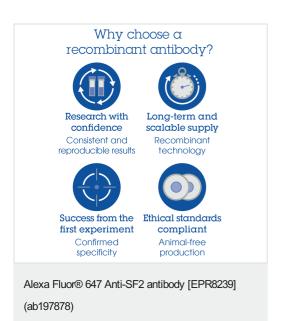
Acquisition of >5,000 events were collected using a solid-state 25mW red diode laser (635 nm) and 675/30 bandpass filter.



Immunocytochemistry/ Immunofluorescence - Alexa Fluor® 647 Anti-SF2 antibody [EPR8239] (ab197878)

ab197878 staining SF2 in HepG2 cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab197878 at a 1/100 dilution (shown in red) and ab195887, Mouse monoclonal to alpha Tubulin (Alexa Fluor[®] 488), at a 1/250 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

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



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