

Product datasheet

Anti-NFAT1 antibody [EPR24658-149] - BSA and Azide free ab283659

Recombinant RabMAb

3 Images

Overview		
Product name	Anti-NFAT1 antibody [EPR24658-149] - BSA and Azide free	
Description	Rabbit monoclonal [EPR24658-149] to NFAT1 - BSA and Azide free	
Host species	Rabbit	
Tested applications	Suitable for: WB Unsuitable for: ChIP,Flow Cyt (Intra),ICC/IF,IHC-P or IP	
Species reactivity	Reacts with: Mouse, Human	
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.	
Positive control	WB: Ramos, Raji, Daudi and EL4 whole cell lysates.	
General notes	ab283659 is the carrier-free version of ab283649 .	
	Our <u>carrier-free</u> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.	
	This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.	
	Use our <u>conjugation kits</u> for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.	
	This product is compatible with the Maxpar [®] Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar [®] is a trademark of Fluidigm Canada Inc.	
	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 <u>see here</u> .	
	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> .	

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C.
Storage buffer	pH: 7.2 Constituent: 100% PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR24658-149
Isotype	lgG

Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab283659 in the following tested applications.

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

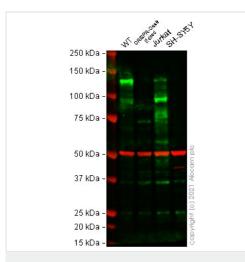
Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Predicted molecular weight: 14 kDa.

Application notes

Is unsuitable for ChIP,Flow Cyt (Intra),ICC/IF,IHC-P or IP.

Target		
Function	Plays a role in the inducible expression of cytokine genes in T-cells, especially in the induction of the IL-2, IL-3, IL-4, TNF-alpha or GM-CSF.	
Tissue specificity	Expressed in thymus, spleen, heart, testis, brain, placenta, muscle and pancreas.	
Sequence similarities	Contains 1 RHD (Rel-like) domain.	
Domain	Rel Similarity Domain (RSD) allows DNA-binding and cooperative interactions with AP1 factors.	
Post-translational modifications	In resting cells, phosphorylated by NFATC-kinase on at least 18 sites in the 99-363 region. Upon cell stimulation, all these sites except Ser-243 are dephosphorylated by calcineurin. Dephosphorylation induces a conformational change that simultaneously exposes an NLS and masks an NES, which results in nuclear localization. Simultaneously, Ser-53 or Ser-56 is phosphorylated; which is required for full transcriptional activity.	
Cellular localization	Cytoplasm. Nucleus. Cytoplasmic for the phosphorylated form and nuclear after activation that is controlled by calcineurin-mediated dephosphorylation. Rapid nuclear exit of NFATC is thought to be one mechanism by which cells distinguish between sustained and transient calcium signals. The subcellular localization of NFATC plays a key role in the regulation of gene transcription.	

Images



Western blot - Anti-NFAT1 antibody [EPR24658-149] - BSA and Azide free (ab283659)

All lanes : Anti-NFAT1 antibody [EPR24658-149] (<u>ab283649</u>) at 1/1000 dilution

Lane 1 : Wild-type Raji cell lysate Lane 2 : NFATC2 CRISPR-Cas9 edited Raji cell lysate Lane 3 : Jurkat cell lysate Lane 4 : SH-SY5Y cell lysate

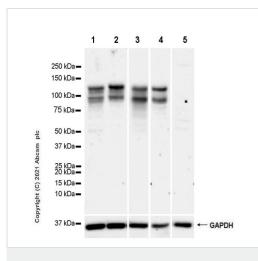
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 14 kDa Observed band size: 100 kDa

This data was developed using <u>ab283649</u>, the same antibody clone in a different buffer formulation.

False colour image of Western blot: Anti-NFAT1 antibody [EPR24658-149] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (ab7291) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab283649 was shown to bind specifically to NFAT1. A band was observed at 100 kDa in wild-type Raji cell lysates with no signal observed at this size in NFATC2 CRISPR-Cas9 edited cell line ab280906 (CRISPR-Cas9 edited cell lysate ab282940). The band observed in the CRISPR-Cas9 edited lysate lane below 100 kDa is likely to represent a truncated form of NFAT1. This has not been investigated further and the functional properties of the gene product have not been determined. To generate this image, wildtype and NFATC2 CRISPR-Cas9 edited Raji cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDve[®] 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye[®] 680RD) preabsorbed (ab216776) at 1/20000 dilution.



Western blot - Anti-CGRP-I antibody [EPR24658-149] - BSA and Azide free (ab283659) All lanes : Anti-NFAT1 antibody [EPR24658-149] (<u>ab283649</u>) at 1/1000 dilution

Lane 1 : Ramos (human Burkitt's lymphoma B lymphocyte), whole cell lysate

Lane 2 : Raji (human Burkitts lymphoma B lymphocyte), whole cell lysate

Lane 3 : Daudi (human Burkitts lymphoma lymphoblast), whole cell lysate

Lane 4 : EL4 (mouse lymphoma T lymphocyte), whole cell lysate Lane 5 : HeLa (human cervix adenocarcinoma epithelial cell), whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution (Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated)

Predicted band size: 14 kDa Observed band size: 100, 140 kDa

This data was developed using <u>ab283649</u>, the same antibody clone in a different buffer formulation.

Blocking and diluting buffer and concentration: 5% NFDM/TBST

The molecular weight observed is consistent with what has been described in the literature (PMID:21078663, PMID:25696812).

Negative control: Hela (PMID:21078663)

Exposure time: Lane 1, 2, 4, 5: 3 minutes ; Lane 3: 92 seconds



Anti-CGRP-I antibody [EPR24658-149] - BSA and

Azide free (ab283659)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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