abcam

Product datasheet

Anti-IRF3 (phospho S386) antibody [EPR2346] ab76493



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Overview

Product name Anti-IRF3 (phospho S386) antibody [EPR2346]

Description Rabbit monoclonal [EPR2346] to IRF3 (phospho S386)

Host species Rabbit

Tested applications Suitable for: WB, Dot blot

Species reactivity Reacts with: Human

Immunogen Synthetic peptide corresponding to Human IRF3 (phospho S386).

Database link: Q14653

Positive control WB: MCF7 cells treated with Calyculin A.

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**.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

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.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

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

Purity Protein A purified

Clonality Monoclonal

1

Clone number **EPR2346**

Isotype lgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab76493 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	★★★★ <u>(1)</u>	1/1000 - 1/10000. Predicted molecular weight: 47 kDa.
Dot blot		1/1000.

Target

Function

Mediates interferon-stimulated response element (ISRE) promoter activation. Functions as a molecular switch for antiviral activity. DsRNA generated during the course of an viral infection leads to IRF3 phosphorylation on the C-terminal serine/threonine cluster. This induces a conformational change, leading to its dimerization, nuclear localization and association with CREB binding protein (CREBBP) to form dsRNA-activated factor 1 (DRAF1), a complex which activates the transcription of genes under the control of ISRE. The complex binds to the IE and PRDIII regions on the IFN-alpha and IFN-beta promoters respectively. IRF-3 does not have any transcription activation domains.

Tissue specificity

Expressed constitutively in a variety of tissues.

Sequence similarities

Belongs to the IRF family.

Contains 1 IRF tryptophan pentad repeat DNA-binding domain.

Post-translational modifications

Constitutively phosphorylated on many serines residues. C-terminal serine/threonine cluster is phosphorylated in response of induction by IKBKE and TBK1. Ser-385 and Ser-386 may be specifically phosphorylated in response to induction. An alternate model propose that the five serine/threonine residues between 396 and 405 are phosphorylated in response to a viral infection. Phosphorylation, and subsequent activation of IRF3 is inhibited by vaccinia virus protein E3.

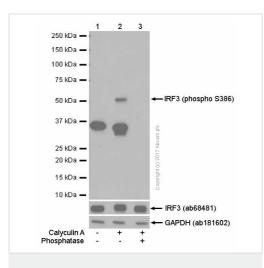
Ubiquitinated; ubiquitination involves RBCK1 leading to proteasomal degradation. Polyubiquitinated; ubiquitination involves TRIM21 leading to proteasomal degradation. ISGylated by HERC5 resulting in sustained IRF3 activation and in the inhibition of IRF3 ubiquitination by disrupting PIN1 binding. The phosphorylation state of IRF3 does not alter

ISGylation.

Cellular localization

Cytoplasm. Nucleus. Shuttles between cytoplasmic and nuclear compartments, with export being the prevailing effect. When activated, IRF3 interaction with CREBBP prevents its export to the cytoplasm.

Images



Western blot - Anti-IRF3 (phospho S386) antibody [EPR2346] (ab76493)

All lanes : Anti-IRF3 (phospho S386) antibody [EPR2346] (ab76493) at 0.2 μg/ml (purified)

Lane 1 : MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysates

Lane 2 : MCF7 (Human breast adenocarcinoma epithelial cell) treated with calyculin A for 45 minutes whole cell lysates

Lane 3: MCF7 (Human breast adenocarcinoma epithelial cell) treated with calyculin A for 45 minutes whole cell lysates. Then the membrane was incubated with phosphatase

Lysates/proteins at 15 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 47 kDa

Blocking and diluting buffer: 5% NFDM/TBST

We are unsure to define the extra band at 37KD.

1 2 2 50 kDa — 150 kDa — 150 kDa — 175 kDa — 18F3 (phospho S386)

37 kDa — 18F3 ab76409

Western blot - Anti-IRF3 (phospho S386) antibody [EPR2346] (ab76493)

All lanes : Anti-IRF3 (phospho S386) antibody [EPR2346] (ab76493) at 1/20000 dilution (Unpurified)

Lane 1: MCF7 cell lysate - treated with Calyculin A

Lane 2: MCF7 cell lysate - untreated

Lysates/proteins at 20 µg per lane.

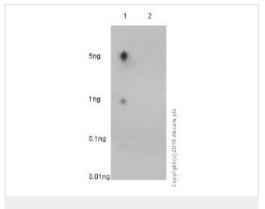
Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

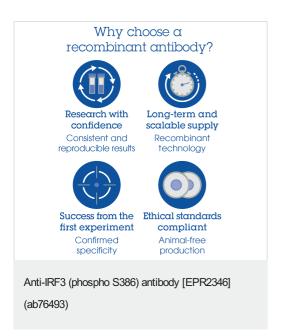
Predicted band size: 47 kDa

Blocking buffer - 5% NFDM/TBST

Diluting buffer - 1% BSA



Dot Blot - Anti-IRF3 (phospho S386) antibody [EPR2346] (ab76493) Dot blot analysis of IRF3 single phospho peptide pS386 (lane 1) and IRF3 non-phospho peptide (lane 2) with unpurified ab76493 at 1/1000. Blocking and diluting buffer was 5% NFDM/TBST. The secondary antibody used was ab97051 Peroxidase conjugated Goat Anti-Rabbit IgG, (H+L) at 1/100,000.



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