abcam

Product datasheet

Anti-TIFA (phospho T9) antibody [EPR19853] ab214815

Recombinant RabMAb

2 References 5 Images

Overview

Product name Anti-TIFA (phospho T9) antibody [EPR19853]

Description Rabbit monoclonal [EPR19853] to TIFA (phospho T9)

Host species Rabbit

Tested applications Suitable for: WB, Dot blot, IP

Species reactivity Reacts with: Human

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

Positive control WB: HeLa transfected with 3×Flag-tagged TIFA expression vector for 24 hours, and then infected

> with S. flexneri 2457T for 4 hours, whole cell lysate. HEK-293T transfected with GFP-tagged TIFA expression vector for 24 hours, and then infected with B. cenocepacia J2315 for 4 hours, whole cell lysate. IP: HEK-293T transfected with GFP-tagged TIFA expression vector for 24 hours, and

then infected with B. cenocepacia J2315 for 4 hours whole cell lysate.

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**.

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 long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

Purity Protein A purified

Clonality Monoclonal

Clone number EPR19853

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab214815 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. Predicted molecular weight: 21 kDa.
Dot blot		1/1000.
IP		1/30.

Target

Function Adapter protein which mediates the IRAK1 and TRAF6 interaction following IL-1 stimulation,

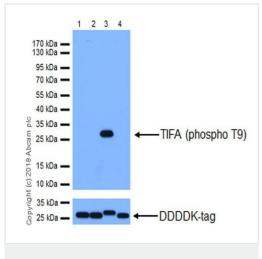
resulting in the downstream activation of NF-kappa-B and AP-1 pathways. Induces the

oligomerization and polyubiquitination of TRAF6, which leads to the activation of TAK1 and IKK

through a proteasome-independent mechanism.

Sequence similarities Contains 1 FHA domain.

Images



Western blot - Anti-TIFA (phospho T9) antibody [EPR19853] (ab214815)

All lanes : Anti-TIFA (phospho T9) antibody [EPR19853] (ab214815) at 1/1000 dilution

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) transfected with 3×Flag-tagged TIFA expression vector for 28 hours, whole cell lysate

Lane 2: HeLa transfected with 3×Flag-tagged TIFA T9A mutant expression vector for 28 hours, whole cell lysate

Lane 3 : HeLa transfected with 3×Flag-tagged TIFA expression vector for 24 hours, and then infected with S. flexneri 2457T for 4 hours, whole cell lysate

Lane 4: HeLa transfected with 3×Flag-tagged TIFAT9A mutant expression vector for 24 hours, and then infected with S. flexneri 2457T for 4 hours, whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes: Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at

1/5000 dilution

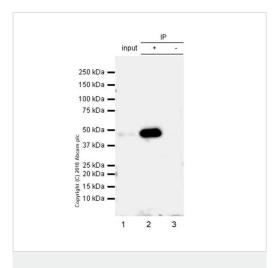
Predicted band size: 21 kDa **Observed band size:** 26 kDa

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% milk/TBST.

The image was kindly provided by our collaborator Dr Feng Shao's lab, NIBS.

S. flexneri infection induces an innate immune response (PMID: 28222186).



Immunoprecipitation - Anti-TIFA (phospho T9) antibody [EPR19853] (ab214815)

TIFA (phospho T9) was immunoprecipitated from 0.35 mg HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate with ab214815 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab214815 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/5,000 dilution

Lane 1: HEK-293T transfected with GFP-tagged TIFA expression vector for 24 hours, and then infected with *B. cenocepacia* J2315 for 4 hours whole cell lysate 10 μ g (Input).

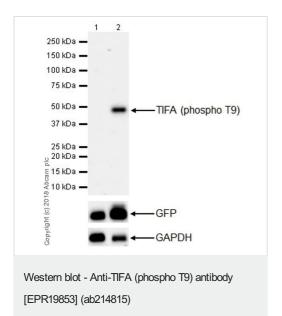
Lane 2: ab214815 IP in HEK-293T transfected with GFP-tagged TIFA expression vector for 24 hours, and then infected with *B. cenocepacia* J2315 for 4 hours whole cell lysate (+).

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab214815 in HEK-293T transfected with GFP-tagged TIFA expression vector for 24 hours, and then infected with *B. cenocepacia* J2315 for 4 hours whole cell lysate (-).

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.

The lysates were kindly provided by our collaborator Dr Feng Shao's lab, NIBS.



All lanes : Anti-TIFA (phospho T9) antibody [EPR19853] (ab214815) at 1/5000 dilution

Lane 1: HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) transfected with GFP-tagged TIFA expression vector, whole cell lysate

Lane 2: HEK-293T transfected with GFP-tagged TIFA expression vector for 24 hours, and then infected with B. cenocepacia J2315 for 4 hours, 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

Predicted band size: 21 kDa **Observed band size:** 47 kDa

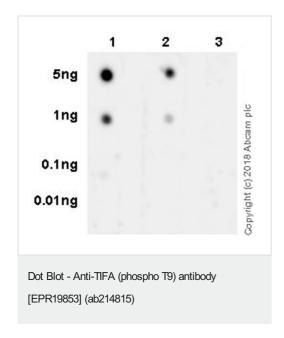
Exposure time: 103 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

The band observed represents GFP-tagged TIFA pT9.

The lysates were kindly provided by our collaborator Dr Feng Shao's lab, NIBS.

B. cenocepacia J2315 infection elicits an inflammatory response. (PMID: 27133449)



Dot blot analysis of TIFA (phospho T9) labeled with ab214815 at 1/1000 dilution.

Lane 1: TIFA (phospho T9) peptide (aa3-12).

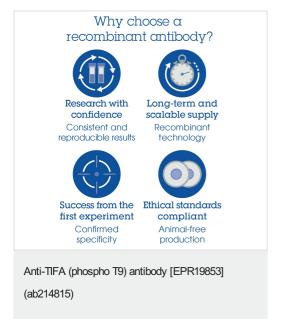
Lane 2: TIFA (phospho T9) peptide (aa5-15).

Lane 3: TIFA non-phospho peptide (aa3-15).

Secondary used was Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ab97051) at 1/100000 dilution.

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 10 seconds.



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