# abcam

### Product datasheet

## Anti-G3BP antibody [EPR13985(B)] ab181149



Recombinant RabMAb

8 References 8 Images

#### Overview

**Product name** Anti-G3BP antibody [EPR13985(B)]

**Description** Rabbit monoclonal [EPR13985(B)] to G3BP

**Host species** Rabbit

Suitable for: WB, ICC/IF, IP, Flow Cyt, IHC-P **Tested applications** 

Species reactivity Reacts with: Human

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

Positive control WB: Wild-type A431, Jurkat and HEK-293 whole cell lysates.

**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.

Preservative: 0.01% Sodium azide Storage buffer

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

**Purity** Protein A purified

Clonality Monoclonal Clone number EPR13985(B)

Isotype lgG

#### The Abpromise guarantee

Our Abpromise quarantee covers the use of ab181149 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		1/10000 - 1/50000. Detects a band of approximately 68 kDa (predicted molecular weight: 52 kDa).
ICC/IF		1/250 - 1/500.
IP		1/20 - 1/40.
Flow Cyt		1/20. <u>ab172730</u> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
IHC-P		1/50 - 1/100.

#### **Target**

**Function** 

May be a regulated effector of stress granule assembly. Phosphorylation-dependent sequencespecific endoribonuclease in vitro. Cleaves exclusively between cytosine and adenine and cleaves MYC mRNA preferentially at the 3'-UTR. ATP- and magnesium-dependent helicase. Unwinds preferentially partial DNA and RNA duplexes having a 17 bp annealed portion and either a hanging 3' tail or hanging tails at both 5'- and 3'-ends. Unwinds DNA/DNA, RNA/DNA, and RNA/RNA substrates with comparable efficiency. Acts unidirectionally by moving in the 5' to 3' direction along the bound single-stranded DNA.

Tissue specificity

Ubiquitous.

Sequence similarities

Contains 1 NTF2 domain.

Contains 1 RRM (RNA recognition motif) domain.

**Domain** 

The NTF2 domain mediates multimerization.

Post-translational modifications

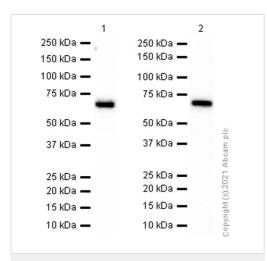
Phosphorylated exclusively on serine residues. Hyperphosphorylated in quiescent fibroblasts. Hypophosphorylation leads to a decrease in endoribonuclease activity (By similarity). RASA1dependent phosphorylation of Ser-149 induces a conformational change that prevents selfassociation. Dephosphorylation after HRAS activation is required for stress granule assembly. Ser-149 phosphorylation induces partial nuclear localization.

Arg-435 is dimethylated, probably to asymmetric dimethylarginine.

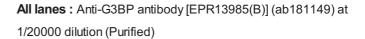
**Cellular localization** 

Cytoplasm. Cytoplasm > cytosol. Cell membrane. Nucleus. Cytoplasmic in proliferating cells, can be recruited to the plasma membrane in exponentially growing cells (By similarity). Cytosolic and partially nuclear in resting cells. Recruited to stress granules (SGs) upon either arsenite or high temperature treatment. Recruitment to SGs is influenced by HRAS.

#### **Images**



Western blot - Anti-G3BP antibody [EPR13985(B)] (ab181149)



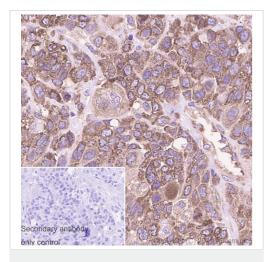
Lane 1 : Ramos (Human Burkitt's lymphoma B lymphocyte) whole cell lysate

Lane 2 : Raji (Human Burkitt's lymphoma B lymphocyte) whole cell lysate

#### **Secondary**

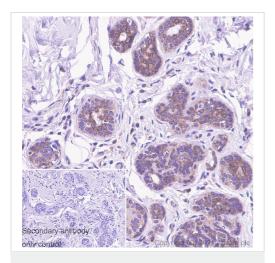
**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 52 kDa



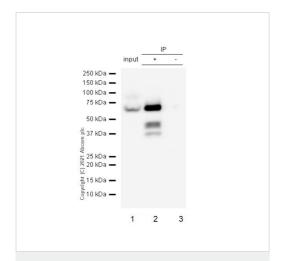
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-G3BP antibody
[EPR13985(B)] (ab181149)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human breast carcinoma tissue sections labeling G3BP with Purified ab181149 at 1:300 (0.4 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-G3BP antibody
[EPR13985(B)] (ab181149)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human breast tissue sections labeling G3BP with Purified ab181149 at 1:300 (0.4 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.



Immunoprecipitation - Anti-G3BP antibody [EPR13985(B)] (ab181149)

Purified ab181149 at 1:20 dilution  $(0.6\mu g)$  immunoprecipitating G3BP in Ramos whole cell lysate.

Lane 1 (input): Ramos (Human Burkitt's lymphoma B lymphocyte) whole cell lysate 10  $\mu g$ .

Lane 2 (+): ab181149 + Ramos whole cell lysate.

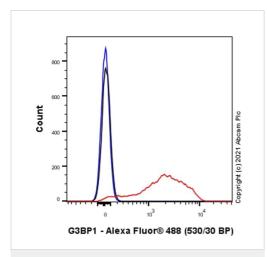
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab181149 in Ramos whole cell lysate.

VeriBlot for IP Detection Reagent (HRP)(<u>ab131366</u>) (1:1000 dilution) was used for Western blotting.

Blocking Buffer and concentration: 5% NFDM/TBST.

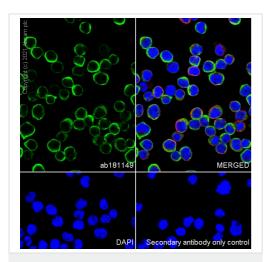
Diluting buffer and concentration: 5% NFDM/TBST.

Observed band size: kDa



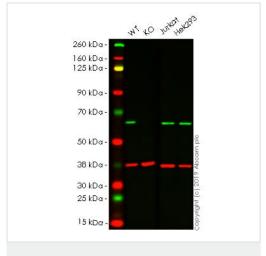
Flow Cytometry - Anti-G3BP antibody [EPR13985(B)] (ab181149)

Flow Cytometry analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labelling G3BP with Purified ab181149 at 1:20 dilution (5 µg/ml) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor® 488, **ab150081**) secondary antibody was used at 1:2000. Isotype control - Rabbit monoclonal lgG (Black). Unlabelled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunocytochemistry/ Immunofluorescence - Anti-G3BP antibody [EPR13985(B)] (ab181149)

Immunocytochemistry analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labeling G3BP with Purified ab181149 at 1:50 dilution (2.4  $\mu$ g/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A]+H21:L21 - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5  $\mu$ g/ml). Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1:1000 (2  $\mu$ g/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Western blot - Anti-G3BP antibody [EPR13985(B)] (ab181149)

**All lanes**: Anti-G3BP antibody [EPR13985(B)] (ab181149) at 1/10000 dilution

**Lane 1 :** Wild-type A-431 (Human epidermoid carcinoma cell line) whole cell lysate

Lane 2: G3BP1 knockout A-431 (Human epidermoid carcinoma cell line) whole cell lysate

Lane 3 : Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

**Lane 4 :** HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

Lysates/proteins at 20 µg per lane.

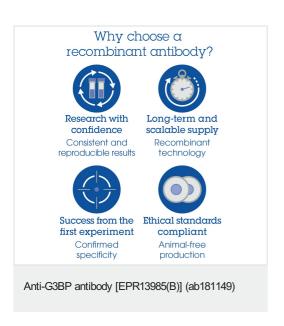
Performed under reducing conditions.

Predicted band size: 52 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab181149 observed at 68 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab181149 was shown to specifically react with G3BP1 in wild-type A431 cells as signal was lost in G3BP1 knockout cells. Wild-type and G3BP1 knockout samples were subjected to SDS-PAGE. The membrane was blocked with 3% Milk. Ab181149 and <a href="mailto:ab8245">ab8245</a> (Mouse anti-GAPDH loading control) were incubated overnight at

4°C at 1/10000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye<sup>®</sup> 800CW) preabsorbed <u>ab216773</u> and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> 680RD) preabsorbed <u>ab216776</u> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



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