**Product datasheet**

**Anti-G3BP antibody [EPR13986(B)] ab181150**

**6 References  13 Images**

### Overview

**Product name**  
Anti-G3BP antibody [EPR13986(B)]

**Description**  
Rabbit monoclonal [EPR13986(B)] to G3BP

**Host species**  
Rabbit

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

**Species reactivity**  
Reacts with: Mouse, Rat, Human

**Immunogen**  
Synthetic peptide within Human G3BP aa 400 to the C-terminus. The exact sequence is proprietary.  
Database link: Q13283

**Positive control**  

**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.

**We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.**

### Properties

**Form**  
Liquid

**Storage instructions**  
**Storage buffer**
- pH: 7.20
- Preservative: 0.01% Sodium azide
- Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

**Purity**
- Protein A purified

**Clonality**
- Monoclonal

**Clone number**
- EPR13986(B)

**Isotype**
- IgG

### Applications

Our [Abpromise guarantee](#) covers the use of **ab181150** in the following tested applications.

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

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>WB</td>
<td>1/1000 - 1/10000. Detects a band of approximately 68 kDa (predicted molecular weight: 52 kDa).</td>
<td></td>
</tr>
<tr>
<td>IHC-P</td>
<td>1/50 - 1/100. Perform heat mediated antigen retrieval using Citrate buffer, pH 6.0.</td>
<td></td>
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<tr>
<td>ICC/IF</td>
<td>1/500. For unpurified format use at 1/50 - 1/100 dilution.</td>
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</tr>
<tr>
<td>IP</td>
<td>1/20.</td>
<td></td>
</tr>
<tr>
<td>Flow Cyt</td>
<td>1/30.</td>
<td></td>
</tr>
<tr>
<td>IHC-P</td>
<td>1/50. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.</td>
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</tr>
</tbody>
</table>

### Target

**Function**
- May be a regulated effector of stress granule assembly. Phosphorylation-dependent sequence-specific endoribonuclease in vitro. Cleave exclusively between cytosine and adenine and cleave 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). RASA1-dependent phosphorylation of Ser-149 induces a conformational change that prevents self-association. 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**

All lanes: Anti-G3BP antibody [EPR13986(B)] (ab181150) at 1/1000 dilution

Lane 1: Wild-type A431 whole cell lysate
Lane 2: G3BP1 knockout A431 whole cell lysate
Lane 3: Jurkat whole cell lysate
Lane 4: HEK-293 whole cell lysate

Lysates/proteins at 20 µg per lane.

**Predicted band size:** 52 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab181150 observed at 68 kDa. Red - loading control, ab8245, observed at 37 kDa.

ab181150 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. Ab181150 and ab8245 (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human hepatocellular cancer tissue sections labeling G3BP with purified ab181150 at 1/50 dilution (6.86 µg/ml). Heat mediated antigen retrieval was performed using Citrate buffer, pH 6.0. ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

**Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-G3BP antibody [EPR13986(B)] (ab181150)**

**Western blot - Anti-G3BP antibody [EPR13986(B)] (ab181150)**

**All lanes**: Anti-G3BP antibody [EPR13986(B)] (ab181150) at 1/10000 dilution (Purified)

**Lane 1**: HEK-293 (Human embryonic kidney epithelial cell) whole cell lysates

**Lane 2**: Ramos (Human Burkitt's lymphoma B lymphocyte) whole cell lysates

**Lane 3**: PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysates

**Lane 4**: NIH/3T3 (Mouse embryonic fibroblast) whole cell lysates

Lysates/proteins at 20 µg per lane.

**Secondary**

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

**Predicted band size**: 52 kDa

**Observed band size**: 68 kDa

**why is the actual band size different from the predicted?**
Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling G3BP with purified ab181150 at 1/500 dilution (0.7 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at 1/200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1/1000 (2 µg/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**

<table>
<thead>
<tr>
<th>Lanes</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Jurkat cell lysate with NFDM/TBST</td>
</tr>
<tr>
<td>2</td>
<td>Ramos cell lysate with NFDM/TBST</td>
</tr>
</tbody>
</table>

Lysates/proteins at 20 µg per lane.

Blocking peptides at 5 % per lane.

**Secondary**

All lanes: Peroxidase conjugated Goat anti-Rabbit IgG (H+L) at 1/1000 dilution

**Predicted band size:** 52 kDa

**Observed band size:** 68 kDa why is the actual band size different from the predicted?
Immunoprecipitation - Anti-G3BP antibody [EPR13986(B)] (ab181150)

ab181150 (purified) at 1/20 dilution (2ug) immunoprecipitating G3BP in Ramos whole cell lysate. Ramos (Human Burkitt's lymphoma B lymphocyte) whole cell lysate 10ug
Lane 2 (+): ab181150 & Ramos whole cell lysate
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab181150 in Ramos whole cell lysate
For western blotting, VeriBlot for IP secondary antibody (HRP) (ab131366) was used at 1/1000 dilution.
Blocking and diluting buffer: 5% NFDM/TBST.

Flow Cytometry - Anti-G3BP antibody [EPR13986(B)] (ab181150)

Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling G3BP with purified ab181150 at 1/30 dilution (10 µg/ml) (red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-G3BP antibody [EPR13986(B)] (ab181150)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Rat kidney tissue sections labeling G3BP with purified ab181150 at 1/50 dilution (6.86 µg/ml). Heat mediated antigen retrieval was performed using Citrate buffer, pH 6.0. ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Mouse kidney tissue sections labeling G3BP with purified ab181150 at 1/50 dilution (6.86 µg/ml). Heat mediated antigen retrieval was performed using Citrate buffer, pH 6.0. ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

Immunofluorescent analysis of 4% paraformaldehyde fixed 293 cells staining G3BP using ab181150 (unpurified) at 1/100 dilution, and Alexa Fluor®555 stained Goat anti Rabbit IgG at 1/200 dilution as a secondary antibody (red). Dapi counterstain (blue)

**Western blot**

**All lanes**: Anti-G3BP antibody [EPR13986(B)] (ab181150) at 1/1000 dilution (unpurified)

**Lane 1**: Hela cell lysate with NFDM/TBST

**Lane 2**: 293 cell lysate with NFDM/TBST

Lysates/proteins at 20 µg per lane.

Blocking peptides at 5 % per lane.

**Secondary**

**All lanes**: Peroxidase conjugated Goat anti-Rabbit IgG (H+L) at 1/1000 dilution

**Predicted band size**: 52 kDa
**Observed band size:** 68 kDa why is the actual band size different from the predicted?

Immunohistochemical analysis of paraffin-embedded Human colon tissue staining G3BP using ab181150 (unpurified) at 1/100 dilution, and prediluted HRP Polymer for Rabbit IgG as a secondary antibody with Hematoxylin counterstain.

Western blot analysis of G3BP in immunoprecipitation pellets from Ramos cell lysate, using ab181151 at a 1/50 dilution. Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated was used as a secondary antibody at 1/1000 dilution. Blocking/dilution buffer and concentration: 5% NFDM/TBST

Anti-G3BP antibody [EPR13986(B)] (ab181150) at 1/50 dilution (unpurified) + Immunoprecipitate from Ramos cell lysate using ab181150 with NFDM/TBST at 5 %

**Secondary**
Peroxidase conjugated Goat anti-Rabbit IgG (H+L) at 1/1000 dilution

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