abcam

Product datasheet

Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade - BSA and Azide free ab273882





18 Images

Overview

Product name Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade - BSA and Azide free

Description Rabbit monoclonal [EPR23796-74] to RBBP7 - ChIP Grade - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: IHC-P, WB, Flow Cyt (Intra), ICC/IF, ChIP-sequencing, IP

Species reactivity Reacts with: Mouse, Rat, Human

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

WB: HAP1, HeLa, HepG2, C6, Jurkat, MCF7, SH-SY5Y, LNCaP and F9 whole cell lysates; Positive control

> Mouse brain, heart, liver, and spleen tissue lysates; Rat heart, kidney and spleen tissue lysates. ICC/IF: HAP1, HeLa and NIH/3T3 cells. IHC-P: Human lung cancer tissue; Mouse lung tissue; Rat lung tissue. Flow Cyt (intra): HAP1, HeLa and NIH/3T3 cells. IP: HeLa and NIH/3T3 whole cell

lysates. ChIP-Seq: Chromatin from HeLa cells.

General notes ab273882 is the carrier-free version of ab259957.

> Our carrier-free 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 cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our **conjugation kits** 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 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. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal
Clone number EPR23796-74

Isotype IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab273882 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
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		Use at an assay dependent concentration. Detects a band of approximately 48 kDa (predicted molecular weight: 48 kDa).
Flow Cyt (Intra)		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.
ChIP-sequencing		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.

Target

Function

Core histone-binding subunit that may target chromatin remodeling factors, histone acetyltransferases and histone deacetylases to their histone substrates in a manner that is regulated by nucleosomal DNA. Component of several complexes which regulate chromatin metabolism. These include the type B histone acetyltransferase (HAT) complex, which is required for chromatin assembly following DNA replication; the core histone deacetylase (HDAC) complex, which promotes histone deacetylation and consequent transcriptional repression; the nucleosome remodeling and histone deacetylase complex (the NuRD complex), which promotes

transcriptional repression by histone deacetylation and nucleosome remodeling; and the PRC2/EED-EZH2 complex, which promotes repression of homeotic genes during development; and the NURF (nucleosome remodeling factor) complex.

Sequence similarities

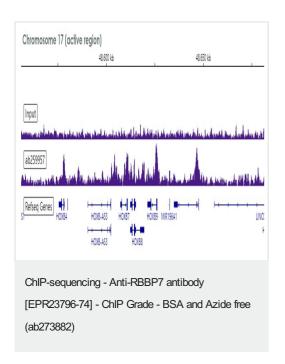
Belongs to the WD repeat RBAP46/RBAP48/MSI1 family.

Contains 7 WD repeats.

Cellular localization

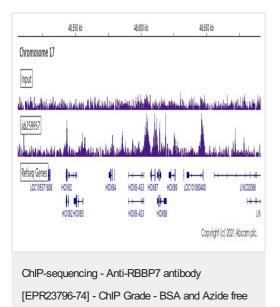
Nucleus.

Images



Chromatin was prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 10^7 HeLa cells and 8 μ g of <u>ab259957</u> [EPR23796-74]. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads.

Additional screenshots of mapped reads can be downloaded <u>here</u>.

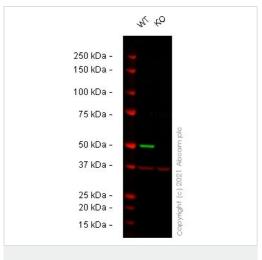


(ab273882)

Chromatin was prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 10^7 HeLa cells and 3 μ g of <u>ab259957</u> [EPR23796-74]. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads.

At time of publication of this image, ChIP-seq was not widely characterised in HeLa for this antibody. For any questions, please contact Abcam Technical Support.

Additional screenshots of mapped reads can be downloaded **here**.



Western blot - Anti-RBBP7 antibody [EPR23796-74]
- ChIP Grade - BSA and Azide free (ab273882)

All lanes : Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade (ab259957) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: RBBP7 knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

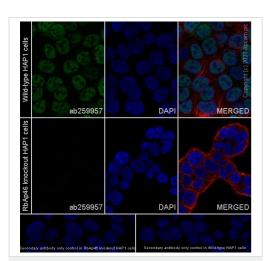
Performed under reducing conditions.

Predicted band size: 48 kDa **Observed band size:** 50 kDa

This data was developed using the same antibody clone in a different buffer formulation (ab259957).

Lanes 1 - 2: Merged signal (red and green). Green - <u>ab259957</u> observed at 50 kDa. Red - loading control <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab259957 was shown to react with RBBP7 in wild-type HeLa cells in Western blot with loss of signal observed in RBBP7 knockout cell line ab264677 (RBBP7 knockout cell lysate ab258628). Wild-type HeLa and RBBP7 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with ab259957 and ab8245 (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated 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 in 20000 dilution for 1 h at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade -BSA and Azide free (ab273882)



Western blot - Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade - BSA and Azide free (ab273882)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized RBBP7 (RbAp46) KO HAP1 cells labelling RBBP7 with ab259957 at 1/500 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing no staining in RBBP7 (RbAp46) KO HAP1 cells. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is <u>ab150077</u> Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) at 1/1000 (2 ug/ml) dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab259957).

All lanes : Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade (ab259957) at 1/1000 dilution

Lane 1: Jurkat (human t cell leukemia t lymphocyte) whole cell lysate

Lane 2 : MCF7 (human breast adenocarcinoma epithelial cell) whole cell lysate

Lane 3: SH-SY5Y (human neuroblastoma epithelial cell) whole cell lysate

Lane 4 : LNCaP (human prostate carcinoma epithelial cell) whole cell lysate

Lane 5: F9 (mouse embryonal carcinoma epithelial cell) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 48 kDa Observed band size: 48 kDa

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

Exposure times: 8 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab259957).

1 2 3 4

250 kDa —

150 kDa —

100 kDa —

75 kDa —

50 kDa —

91d weep op occ 20 kDa —

115 kDa —

Western blot - Anti-RBBP7 antibody [EPR23796-74]

- ChIP Grade - BSA and Azide free (ab273882)

All lanes : Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade (ab259957) at 1/2000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: RBBP7 (RbAp46) knockout HAP1 whole cell lysate

Lane 3: HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 4 : HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 48 kDa **Observed band size:** 48 kDa

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

<u>ab259957</u> was shown to specifically react with RBBP7 in wild-type HAP1 cells as signal was lost in RBBP7 (RbAp46) knockout cells.

Wild-type and RBBP7 (RbAp46) knockout samples were subjected to SDS-PAGE. <u>ab259957</u> and <u>ab181602</u> (Rabbit anti-GAPDH loading control) were incubated 1 hour at room temperature at 1/2000 dilution and 1/200,000 dilution respectively. Blots were developed with Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (<u>ab97051</u>) secondary antibody at 1/50,000 dilution for 1 hour at room temperature before imaging. The blot was developed on a BIO-RAD® ChemiDoc[™] MP instrument using the ECL

technique.

Exposure time: 10 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab259957).

Immunohistochemical analysis of paraffin-embedded Mouse lung tissue labeling RBBP7 with ab**ab259957** at 1/2000 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Nuclear staining in mouse lung (PMID: 19655816). The section was incubated with **ab259957** for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

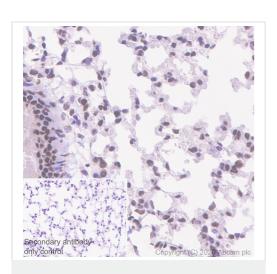
Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab259957).

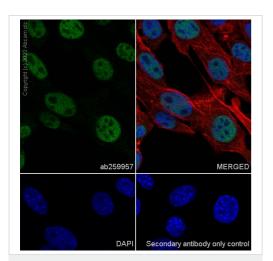
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized NIH/3T3 cells labelling RBBP7 with ab259957 at 1/500 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing mainly nuclear staining in NIH/3T3 cell line. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor[®] 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is <u>ab150077</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor[®] 488) at 1/1000 (2 ug/ml) dilution.

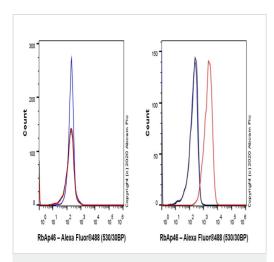


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBBP7 antibody

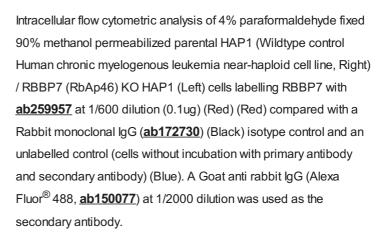
[EPR23796-74] - ChIP Grade - BSA and Azide free (ab273882)



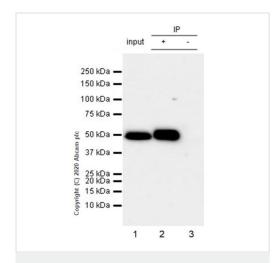
Immunocytochemistry/ Immunofluorescence - Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade -BSA and Azide free (ab273882)



Flow Cytometry (Intracellular) - Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade - BSA and Azide free (ab273882)



This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab259957).



Immunoprecipitation - Anti-RBBP7 antibody
[EPR23796-74] - ChIP Grade - BSA and Azide free
(ab273882)

RBBP7 was immunoprecipitated from 0.35 mg NIH/3T3 (mouse embryonic fibroblast) whole cell lysate with <u>ab259957</u> at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using <u>ab259957</u> at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP)(<u>ab131366</u>) was used at 1/5000 dilution.

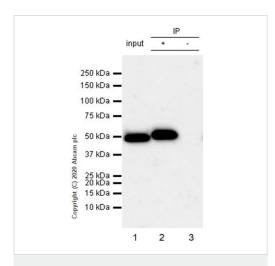
Lane 1: NIH/3T3 (mouse embryonic fibroblast) whole cell lysate 10 ug

Lane 2: abab259957 IP in NIH/3T3 whole cell lysate

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of <u>ab259957</u> in NIH/3T3 whole cell lysate

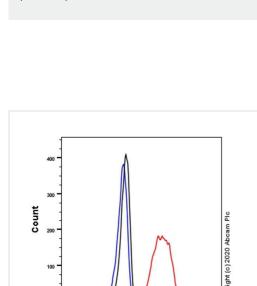
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 24 seconds.



Immunoprecipitation - Anti-RBBP7 antibody

[EPR23796-74] - ChIP Grade - BSA and Azide free
(ab273882)



Flow Cytometry (Intracellular) - Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade - BSA and Azide free (ab273882)

RbAp46 - Alexa Fluor®488 (530/30BP)

RBBP7 was immunoprecipitated from 0.35 mg HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate with <u>ab259957</u> at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using <u>ab259957</u> at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP)(<u>ab131366</u>) was used at 1/5000 dilution.

Lane 1: HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate 10 ug

Lane 2: abab259957 IP in HeLa whole cell lysate

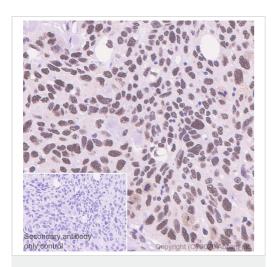
Lane 3: Rabbit monoclonal $\lg G$ (<u>ab172730</u>) instead of <u>ab259957</u> in HeLa whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 24 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab259957</u>).

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized NIH/3T3 (Mouse embryonic fibroblast) cells labelling RBBP7 (RbAp46) with <u>ab259957</u> at 1/600 dilution (0.1ug) (Red) compared with a Rabbit monoclonal IgG (<u>ab172730</u>) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor[®] 488, <u>ab150077</u>) at 1/2000 dilution was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBBP7 antibody

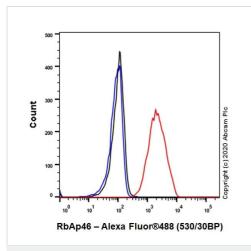
[EPR23796-74] - ChIP Grade - BSA and Azide free (ab273882)

Immunohistochemical analysis of paraffin-embedded Human lung cancer tissue labeling RBBP7 with ab<u>ab259957</u> at 1/2000 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Nuclear staining in human lung cancer (PMID: 19655816).The section was incubated with <u>ab259957</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

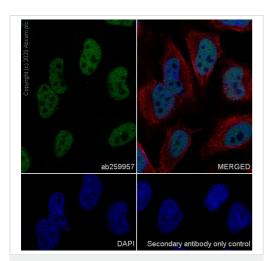
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab259957).



Flow Cytometry (Intracellular) - Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade - BSA and Azide free (ab273882)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized Hela (Human cervix adenocarcinoma epithelial cell) cells labelling RBBP7 (RbAp46) with <u>ab259957</u> at 1/600 dilution (0.1ug) (Red) compared with a Rabbit monoclonal lgG (<u>ab172730</u>) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit lgG (Alexa Fluor[®] 488, <u>ab150077</u>) at 1/2000 dilution was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade -BSA and Azide free (ab273882)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBBP7 antibody

[EPR23796-74] - ChIP Grade - BSA and Azide free (ab273882)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized HeLa cells labelling RBBP7 with ab259957 at 1/500 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing mainly nuclear staining in HeLa cell line. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor[®] 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

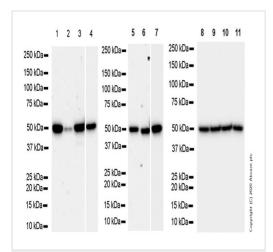
Secondary antibody only control: Secondary antibody is <u>ab150077</u> Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) at 1/1000 (2 ug/ml) dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab259957).

Immunohistochemical analysis of paraffin-embedded Rat lung tissue labeling RBBP7 with ab<u>ab259957</u> at 1/2000 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Nuclear staining in rat lung (PMID: 19655816). The section was incubated with <u>ab259957</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



Western blot - Anti-RBBP7 antibody [EPR23796-74]

- ChIP Grade - BSA and Azide free (ab273882)

All lanes : Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade (ab259957) at 1/1000 dilution

Lane 1: Mouse brain tissue lysate

Lane 2: Mouse heart tissue lysate

Lane 3: Mouse liver tissue lysate

Lane 4: Mouse spleen tissue lysate

Lane 5: Rat heart tissue lysate

Lane 6: Rat kidney tissue lysate

Lane 7: Rat spleen tissue lysate

Lane 8: C6 (rat glial tumor glial cell) whole cell lysate

Lane 9: RAW 264.7 (mouse abelson murine leukemia virus-

induced tumor macrophage) whole cell lysate

Lane 10 : PC-12 (rat adrenal gland pheochromocytoma) whole cell

lysate

Lane 11: NIH/3T3 (mouse embryonic fibroblast) whole cell lysate

Lysates/proteins at 20 µg per lane.

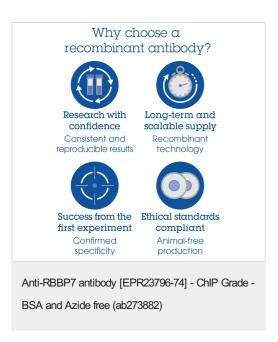
Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 48 kDa **Observed band size:** 48 kDa

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

Exposure times: Lanes 1-3: 3 minutes; Lane 4: 3 seconds; Lanes 5-6: 3 minutes; Lane 7-11: 3 seconds.



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

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit https://www.abcam.com/abpromise or contact our technical team.

Terms and conditions

• Guarantee only valid for products bought direct from Abcam or one of our authorized distributors