abcam

Product datasheet

Anti-PBR antibody [EPR5384] - BSA and Azide free ab213654

KO 評価済 リコンビナント RabMAb

画像数 18

製品の概要

製品名	Anti-PBR antibody [EPR5384] - BSA and Azide free
製品の詳細	Rabbit monoclonal [EPR5384] to PBR - BSA and Azide free
由来種	Rabbit
特異性	IHC results on rat tissues (such as liver and kidney) showed weak cytoplasmic and nuclear staining. However, other customer feedback suggests that this antibody works well in rat. Due to the inconclusive nature of these results, we do not currently guarantee this antibody in rat. Please contact our Scientific support team for more information.
アプリケーション	**適用あり:** IHC-Fr, IHC-P, WB, IP, ICC/IF, Flow Cyt (Intra)
種交差性	**交差種:** Mouse, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. (Peptide available as **ab170987**)
ポジティブ・コントロール	WB: HCT116, HAP1, HeLa, U-87MG, 293T, A431, RAW264.7, and NIH3T3 cell lysates; IHC-P: Human bladder carcinoma and colon tissue; ICC/IF: U-87MG cells, HeLa cells, TSPO-HAP1 cells; Flow Cyt (intra): HepG2 and U-87MG cells; IP: A431 and U-87MG cell lysate; IHC-Fr: Mouse kidney and adrenal gland tissue; IHC-P: Human bladder carcinoma and Mouse hypothalamus tissue.
特記事項	ab213654 is the carrier-free version of **ab109497**. 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 cell-based 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**.

Rat: We have preliminary internal testing data to indicate this antibody may not react with this species. Please contact us for more information.

製品の特性

製品の状態	Liquid
保存方法	Shipped at 4°C. Store at +4°C. Do Not Freeze.
バッファー	pH: 7.2 Constituent: PBS
キャリア・フリー	はい
精製度	Protein A purified
ポリ/モノ	モノクローナル
クローン名	EPR5384
アイソタイプ	IgG

アプリケーション

The Abpromise guarantee **Abpromise保証は、** **次のテスト済みアプリケーションにおけるab213654の使用に適用されます**

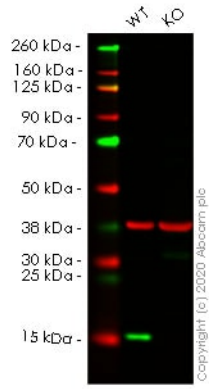
アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
IHC-Fr		Use at an assay dependent concentration. Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**.
WB		Use at an assay dependent concentration. Predicted molecular weight: 19 kDa.
IP		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab199376** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

ターゲット情報

機能	Responsible for the manifestation of peripheral-type benzodiazepine recognition sites and is most likely to comprise binding domains for benzodiazepines and isoquinoline carboxamides. May play a role in the transport of porphyrins and heme. Plays a role in the transport of cholesterol across mitochondrial membranes in steroidogenic cells.
組織特異性	Found in many tissue types. Expressed at the highest levels under normal conditions in tissues that synthesize steroids.
配列類似性	Belongs to the TspO/BZRP family.
細胞内局在	Mitochondrion membrane.

画像



Western blot - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

All lanes : Anti-PBR antibody [EPR5384] (**ab109497**) at 1/1000 dilution

Lane 1 : Wild-type HCT116 cell lysate
Lane 2 : TSPO knockout HCT116 cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

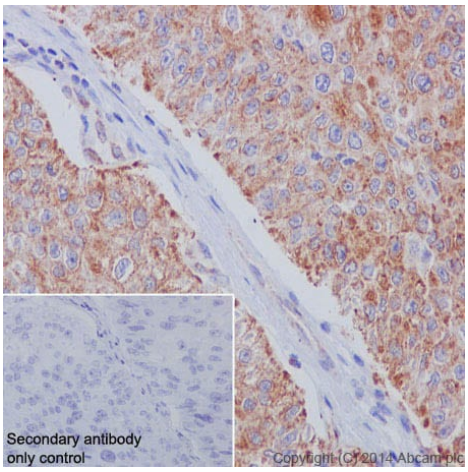
Predicted band size: 19 kDa
Observed band size: 17 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab109497**).

Lanes 1- 2: Merged signal (red and green). Green - **ab109497** observed at 17 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab109497 was shown to react with PBR in wild-type HCT116 cells in western blot. Loss of signal was observed when knockout cell line **ab266878** (knockout cell lysate **ab257067**) was used. Wild-type HCT116 and TSPO knockout HCT116 cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. **ab109497** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (**ab216776**) secondary

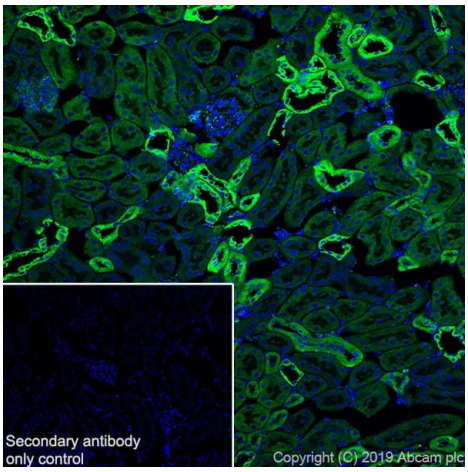
antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human bladder carcinoma tissue labelling PBR with purified **ab109497** at 1/100. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

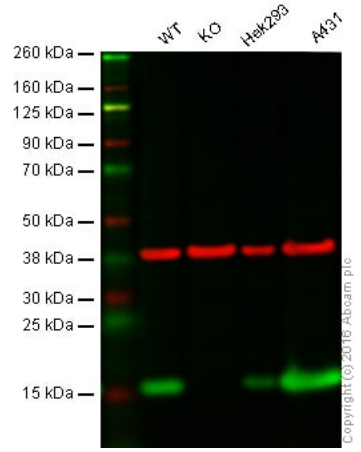
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).



Immunohistochemistry (Frozen sections) - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

Immunohistochemistry (Frozen sections) analysis of mouse kidney tissue sections labeling PBR with Purified **ab109497** at 1/250 (3.8 µg/ml).Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. DAPI was used as a counterstain.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).



Western blot - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

All lanes : Anti-PBR antibody [EPR5384] (**ab109497**) at 1/10000 dilution

Lane 1 : Wild-type HAP1 cell lysate
Lane 2 : PBR knockout HAP1 cell lysate
Lane 3 : HEK293 cell lysate
Lane 4 : A431 cell lysate

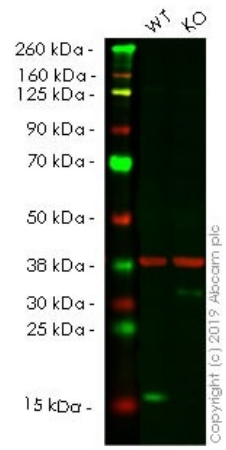
Lysates/proteins at 20 µg per lane.

Predicted band size: 19 kDa

This data was produced with **ab109497**, the same antibody in a diffrent forulation with BSA and Azide.

Lanes 1 - 4: Merged signal (red and green). Green - **ab109497** observed at 15 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab109497 was shown to specifically react with PBR in wild type HAP1 cells. No band was observed when PBR knockout samples were used. Wild-type and PBR knockout samples were subjected to SDS-PAGE. Ab109497 and **ab8245** (loading control to GAPDH) were diluted at 1/10000 and 1/10000 dilution respectively and incubated overnight at 4C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

All lanes : Anti-PBR antibody [EPR5384] (**ab109497**) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : TSPO (PBR) knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

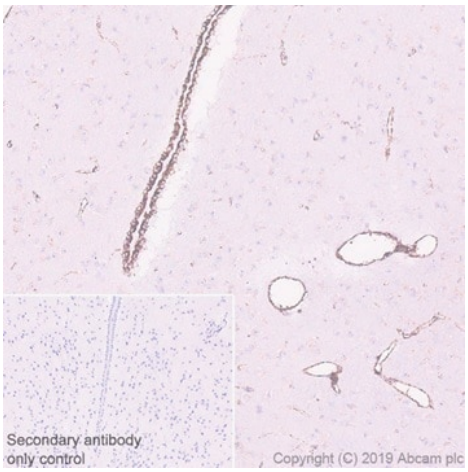
Performed under reducing conditions.

Predicted band size: 19 kDa
Observed band size: 15 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab109497**).

Lanes 1- 2: Merged signal (red and green). Green - **ab109497** observed at 15 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab109497 was shown to react with PBR in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab264942** (knockout cell lysate **ab257066**) was used. Wild-type HeLa and TSPO knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. **ab109497** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

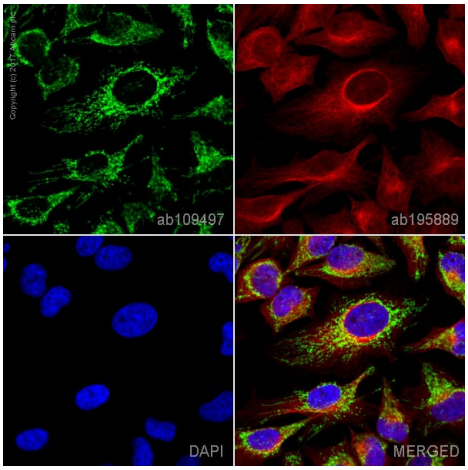


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

Immunohistochemistry (formalin/PFA-fixed paraffin-embedded sections) analysis of mouse hypothalamus tissue labelling PBR with **ab109497** at 1/1000 dilution (1.03 mg/ml). Heat mediated antigen retrieval was performed using Tris/EDTA buffer, pH 9.0 (**ab93684**). A goat anti-rabbit IgG (H+L) (HRP) was used as the secondary antibody (concentration: ready to use). A secondary-antibody-only control was also performed. Counterstained with hematoxylin.

Positive staining was observed on ependymal cells and endothelial cells in mouse hypothalamus.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).

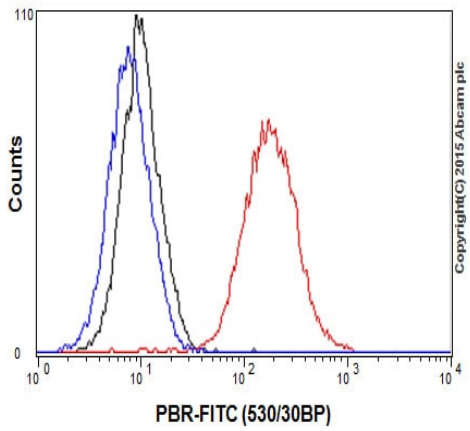


Immunocytochemistry/ Immunofluorescence - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

ab109497 staining PBR in HeLa. The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with **ab109497** at 1µg/ml and **ab195889** at 1/250 dilution (shown in pseudocolour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit IgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

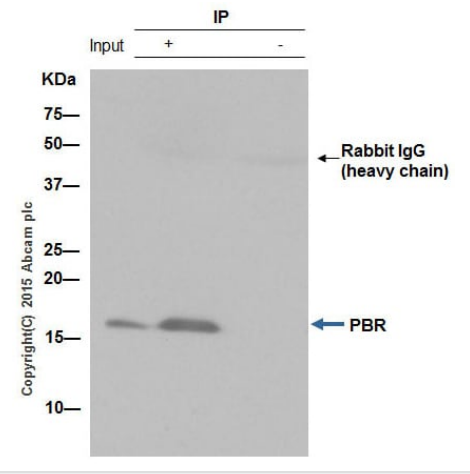
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).



Flow Cytometry (Intracellular) - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

Intracellular Flow Cytometry analysis of U87-MG cells labelling PBR with purified **ab109497** at 1/150 (red). Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).



Immunoprecipitation - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

ab109497 (purified) at 1/60 immunoprecipitating PBR in A431 whole cell lysate.

Lane 1 (input): A431 whole cell lysate (10µg)

Lane 2 (+): **ab109497** + A431 whole cell lysate (10µg).

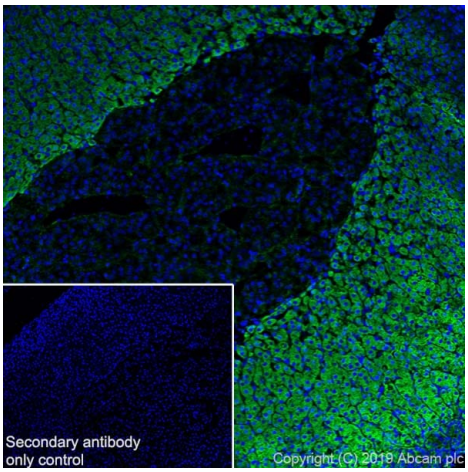
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of **ab109497** in A431 whole cell lysate.

For western blotting, VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/1500 dilution.

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

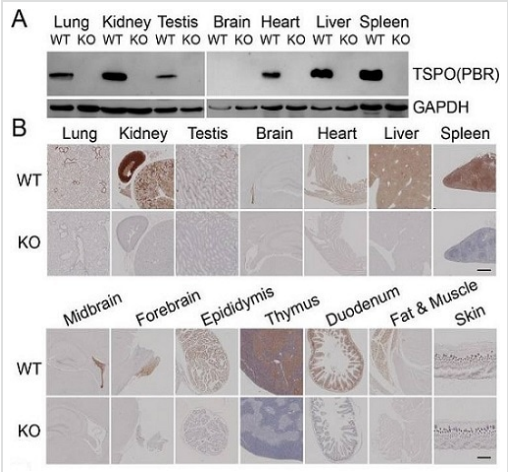
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).



Immunohistochemistry (Frozen sections) - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

Immunohistochemistry (Frozen sections) analysis of mouse adrenal gland tissue sections labeling PBR with Purified **ab109497** at 1/250 (3.8 µg/ml).Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. DAPI was used as a counterstain.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

Wang H. et al. PLoS One. 2016 Dec 1;11(12):e0167307. doi: 10.1371/journal.pone.0167307. eCollection 2016.

TSPO (PBR) expression was abolished in global KO mice without pathological changes

TSPO expression in different tissues from WT and KO mice was detected by western blotting (A) and IHC (B). Scale Bars, 100µm.

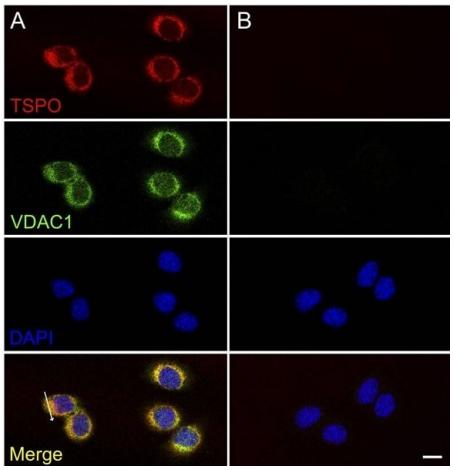
4% PFA-fixed tissue sections were blocked with 5% goat serum and incubated overnight at 4°C with **ab109497** at 1/1500 dilution. DAB staining.

Note: TSPO and PBR are alternative names for the same target.

(Adapted from Figure 3 of Wang et al)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

Morohaku K. et al. PLoS One. 2013 Sep 5;8(9):e74509. doi: 10.1371/journal.pone.0074509. eCollection 2013.

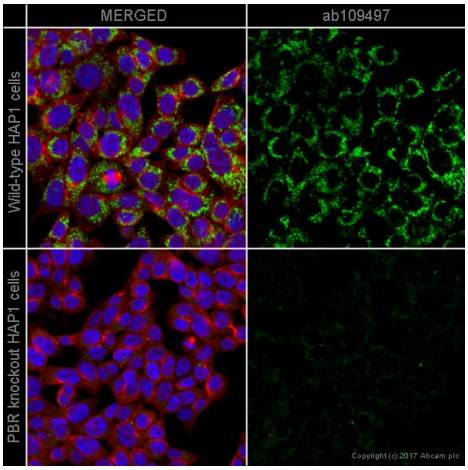
TSPO (PBR) expression is localized to the mitochondria

(A) Panel shows confocal images of TSPO (red), VDAC1 (green) and nuclear counterstain (blue) in MA-10 Leydig cells. (B) Negative control panel. Colocalization of TSPO to the mitochondrial protein VDAC1 validates the specific localization of TSPO. Scale bar 20 µm.

Cells were fixed with 4% formaldehyde and permeabilized using 0.1% Triton X-100. Cells were then blocked using 5% normal goat serum and incubated with **ab109497** at 1/200 dilution.

Note: TSPO and PBR are alternative names for the same target.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).

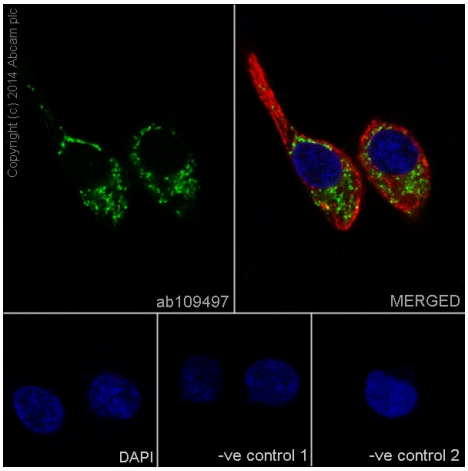


Immunocytochemistry/ Immunofluorescence - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

ab109497 staining PBR in wild-type HAP1 cells (top panel) and PBR knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with **ab109497** at 1µg/ml and **ab195889** at 1/250 dilution (shown in pseudocolour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit IgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).



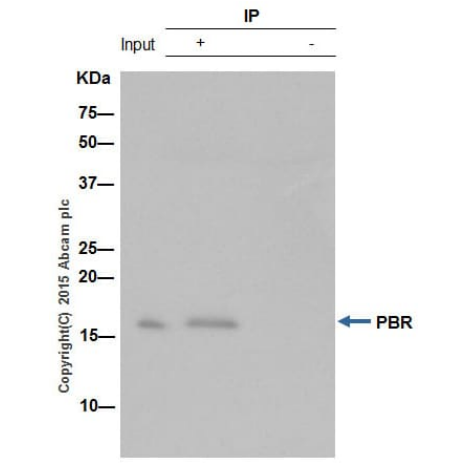
Immunocytochemistry/ Immunofluorescence - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

Immunocytochemistry/Immunofluorescence analysis of U87-MG cells labelling PBR with purified **ab109497** at 1/100. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. **ab7291**, a mouse anti-tubulin (1/1000) and **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500) were also used.

Control 1: primary antibody (1/100) and secondary antibody, **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500).

Control 2: **ab7291** (1/1000) and secondary antibody, **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).



Immunoprecipitation - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

ab109497 (purified) at 1/60 immunoprecipitating PBR in U87-MG whole cell lysate.

Lane 1 (input): U87-MG whole cell lysate (10µg)

Lane 2 (+): **ab109497** + U87-MG whole cell lysate (10µg).

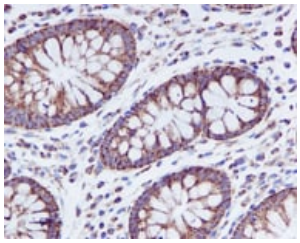
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of **ab109497** in U87-MG whole cell lysate.

For western blotting, VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/1500 dilution.

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).

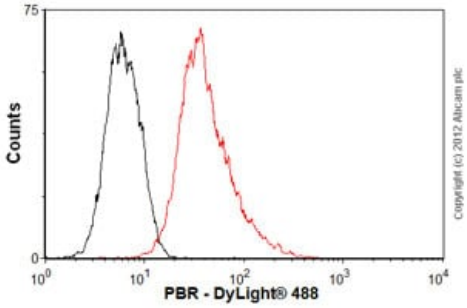


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon tissue labelling PBR with unpurified **ab109497** at a dilution of 1/100.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).

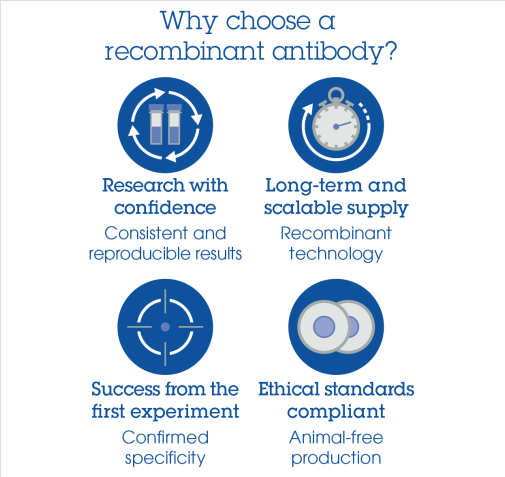
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-PBR antibody [EPR5384] - BSA and Azide free (ab213654)

Overlay histogram showing HepG2 cells stained with unpurified **ab109497** (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions. The cells were then incubated with the antibody (unpurified **ab109497**, 1/50 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (**ab96899**) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109497**).



Anti-PBR antibody [EPR5384] - BSA and Azide free

(ab213654)

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- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
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- 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.co.jp/abpromise** or contact our technical team.

Terms and conditions

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