# abcam

### Product datasheet

# Anti-EBP50/NHERF-1 antibody [EPR5562] - BSA and Azide free ab247858



リコンビナント

RabMAb

#### 画像数 6

#### 製品の概要

特記事項

製品名 Anti-EBP50/NHERF-1 antibody [EPR5562] - BSA and Azide free

製品の詳細 Rabbit monoclonal [EPR5562] to EBP50/NHERF-1 - BSA and Azide free

由来種 Rabbit

アプリケーション 適用あり: WB, IHC-P

適用なし: Flow Cyt,ICC/IF or IP

種交差性 交差種: Rat, Human

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

ポジティブ・コントロール HCT116, HepG2, 293T, Jurkat, C6, PC-12 and MCF-7 cell lysates; Human kidney tissue

ab247858 is the carrier-free version of ab109430.

Our <u>carrier-free</u> 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<sup>®</sup> Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar<sup>®</sup> 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<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

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Mouse: 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** EPR5562

アイソタイプ IgG

#### アプリケーション

**The Abpromise guarantee** <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab247858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
WB		Use at an assay dependent concentration. Predicted molecular weight: 39 kDa.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.  Antigen retrieval is recommended.

追加情報

Is unsuitable for Flow Cyt,ICC/IF or IP.

#### ターゲット情報

機能 Scaffold protein that connects plasma membrane proteins with members of the

ezrin/moesin/radixin family and thereby helps to link them to the actin cytoskeleton and to regulate their surface expression. Necessary for recycling of internalized ADRB2. Was first known to play a role in the regulation of the activity and subcellular location of SLC9A3. Necessary for cAMP-mediated phosphorylation and inhibition of SLC9A3. May enhance Wnt signaling. May participate

in HTR4 targeting to microvilli (By similarity). Interacts with MCC.

組織特異性 Detected in liver, kidney, pancreas, prostate, spleen, small intestine and placenta, in particular in

the syncytiotrophoblast.

関連疾患 Defects in SLC9A3R1 are the cause of hypophosphatemic nephrolithiasis/osteoporosis type 2

(NPHLOP2) [MIM:612287]. Hypophosphatemia results from idiopathic renal phosphate loss. It contributes to the pathogenesis of hypophosphatemic urolithiasis (formation of urinary calculi) as

well to that of hypophosphatemic osteoporosis (bone demineralization).

## 配列類似性 翻訳後修飾

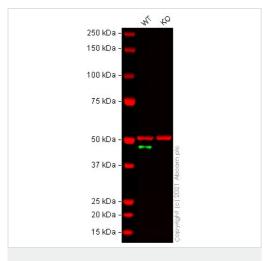
細胞内局在

Contains 2 PDZ (DHR) domains.

Phosphorylated on serine residues.

Cytoplasm. Apical cell membrane. Endomembrane system. Cell projection > filopodium. Cell projection > ruffle. Cell projection > microvillus. Translocates from the cytoplasm to the apical cell membrane in a PODXL-dependent manner (By similarity). Colocalizes with actin in microvilli-rich apical regions of the syncytiotrophoblast. Found in microvilli, ruffling membrane and filopodia of HeLa cells. Present in lipid rafts of T-cells.

#### 画像



Western blot - Anti-EBP50/NHERF-1 antibody [EPR5562] - BSA and Azide free (ab247858)

**All lanes :** Anti-EBP50/NHERF-1 antibody [EPR5562] (<u>ab109430</u>) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

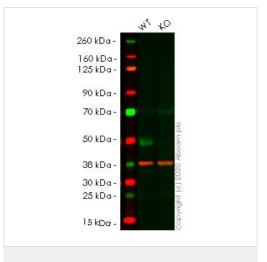
Lane 2: SLC9A3R1 knockout HeLa cell lysate

Performed under reducing conditions.

**Predicted band size:** 39 kDa **Observed band size:** 46 kDa

This data was developed using the same antibody clone in a different buffer formulation (<u>ab109430</u>).

False colour image of Western blot: Anti-EBP50/NHERF-1 antibody [EPR5562] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (ab7291) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab109430 was shown to bind specifically to EBP50/NHERF-1. A band was observed at 46 kDa in wild-type HeLa cell lysates with no signal observed at this size in SLC9A3R1 knockout cell line ab264914 (knockout cell lysate ab257280). To generate this image, wild-type and SLC9A3R1 knockout HeLa cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> 680RD) preabsorbed (**ab216776**) at 1/20000 dilution.



Western blot - Anti-EBP50/NHERF-1 antibody [EPR5562] - BSA and Azide free (ab247858)

**All lanes :** Anti-EBP50/NHERF-1 antibody [EPR5562] (**ab109430**) at 1/1000 dilution

Lane 1: Wild-type HCT116 cell lysate

Lane 2: SLC9A3R1 knockout HCT116 cell lysate

Lysates/proteins at 20 µg per lane.

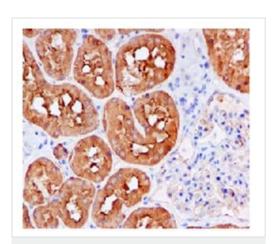
Performed under reducing conditions.

Predicted band size: 39 kDa Observed band size: 48 kDa

This data was developed using the same antibody clone in a different buffer formulation (<u>ab109430</u>).

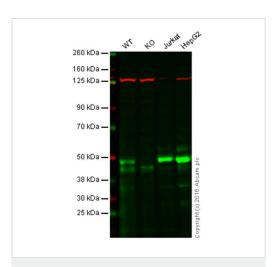
**Lanes 1-2:** Merged signal (red and green). Green - <u>ab109430</u> observed at 48 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) observed at 37 kDa.

ab109430 was shown to react with EBP50/NHERF-1 in wild-type HCT116 cells in western blot. Loss of signal was observed when knockout cell line ab266876 (knockout cell lysate ab257281) was used. Wild-type HCT116 and SLC9A3R1 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. ab109430 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 lgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG 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 paraffinembedded sections) - Anti-EBP50/NHERF-1 antibody [EPR5562] - BSA and Azide free (ab247858)

This data was developed using <u>ab109430</u>, the same antibody clone in a different buffer formulation.lmmunohistochemical analysis of EBP50/NHERF-1 in paraffin-embedded Human kidney tissue using <u>ab109430</u> at 1/100 dilution. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-EBP50/NHERF-1 antibody [EPR5562] - BSA and Azide free (ab247858) This data was developed using <u>ab109430</u>, the same antibody clone in a different buffer formulation.

Lane 1: Wild-type HAP1 cell lysate (20 µg)

Lane 2: EBP50/NHERF-1 knockout HAP1 cell lysate (20 µg)

Lane 3: Jurkat cell lysate (20 µg)

Lane 4: HepG2 cell lysate (20 µg)

**Lanes 1 - 4:** Merged signal (red and green). Green - <u>ab109430</u> observed at 48 kDa. Red - loading control, <u>ab18058</u>, observed at 124 kDa.

<u>ab109430</u> was shown to specifically recognize EBP50/NHERF-1 in wild-type HAP1 cells along with additional cross-reactive bands. No band was observed when EBP50/NHERF-1 knockout samples were usexamined. Wild-type and EBP50 knockout samples were subjected to SDS-PAGE. <u>ab109430</u> and <u>ab18058</u> (loading control to Vinculin) were diluted at 1/500 and 1/10000 respectively and incubated overnight at 4°C. Blots were developed withGoat anti-Rabbit IgG H&L (IRDye<sup>®</sup> 800CW) preadsorbed (<u>ab216773</u>) and

Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> 680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.

[EPR5562] - BSA and Azide free (ab247858)

**All lanes :** Anti-EBP50/NHERF-1 antibody [EPR5562] (<u>ab109430</u>) at 1/1000 dilution

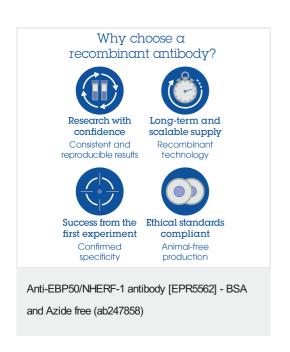
Lane 1 : HepG2 cell lysate
Lane 2 : 293T cell lysate
Lane 3 : Jurkat cell lysate
Lane 4 : C6 cell lysate
Lane 5 : PC12 cell lysate

Lane 6 : MCF7 cell lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 39 kDa

This data was developed using <u>ab109430</u>, the same antibody clone in a different buffer formulation.



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