abcam

Product datasheet

Anti-ENPP2/ATX antibody [1F8] ab77104



10 References 4 Images

Overview

Product name Anti-ENPP2/ATX antibody [1F8]

Description Mouse monoclonal [1F8] to ENPP2/ATX

Host species Mouse

Tested applications Suitable for: IHC-P, WB

Unsuitable for: ICC

Species reactivity Reacts with: Human

Predicted to work with: Mouse

Immunogen Recombinant full length protein corresponding to Human ENPP2/ATX.

Positive control WB: HEK293 whole cell lysate and in the following human tissue lysates: kidney; placenta; ovary;

small intestine. IHC: human tonsil paraffin sections.

General notes

This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or

conjugation for your experiments, please contact orders@abcam.com.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Storage instructions Shipped at 4°C. Upon delivery aliquot and store at -20°C or -80°C. Avoid repeated freeze / thaw

cycles.

Storage buffer pH: 7.40

Preservative: 0.02% Sodium azide Constituents: PBS, 6.97% L-Arginine

Purity Protein G purified

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Clonality Monoclonal

Clone number 1F8 lsotype lqG1

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab77104 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 a concentration of 1 µg/ml.
WB		Use a concentration of 1 µg/ml. Detects a band of approximately 100 kDa (predicted molecular weight: 99 kDa).

Application notes Is unsuitable for ICC.

Target

Function

Hydrolyzes lysophospholipids to produce lysophosphatidic acid (LPA) in extracellular fluids. Major substrate is lysophosphatidylcholine. Also can act on sphingosylphosphphorylcholine producing sphingosine-1-phosphate, a modulator of cell motility. Can hydrolyze, in vitro, bis-pNPP, to some extent pNP-TMP, and barely ATP. Involved in several motility-related processes such as angiogenesis and neurite outgrowth. Acts as an angiogenic factor by stimulating migration of smooth muscle cells and microtubule formation. Stimulates migration of melanoma cells, probably via a pertussis toxin-sensitive G protein. May have a role in induction of parturition. Possible involvement in cell proliferation and adipose tissue development. Tumor cell motility-stimulating factor.

Tissue specificity

Predominantly expressed in brain, placenta, ovary, and small intestine. Expressed in a number of carcinomas such as hepatocellular and prostate carcinoma, neuroblastoma and non-small-cell lung cancer. Expressed in body fluids such as plasma, cerebral spinal fluid (CSF), saliva, follicular and amniotic fluids. Not detected in leukocytes. Isoform 1 is more highly expressed in peripheral tissues than in the central nervous system (CNS). Adipocytes only express isoform 1. Isoform 3 is more highly expressed in the brain than in peripheral tissues.

Sequence similarities

Belongs to the nucleotide pyrophosphatase/phosphodiesterase family.

Contains 2 SMB (somatomedin-B) domains.

Post-translational modifications

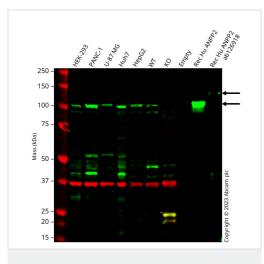
N-glycosylation, but not furin-cleavage, plays a critical role on secretion and on lysoPLD activity.

Cellular localization

Secreted. Secreted by most body fluids including serum and CSF. Also by adipocytes and ...

numerous cancer cells.

Images



Western blot - Anti-ENPP2/ATX antibody [1F8] (ab77104)

All lanes : Anti-ENPP2/ATX antibody [1F8] (ab77104) at 1/1000 dilution

Lane 1: HEK-293 cell lysate at 20 μg **Lane 2**: PANC-1 cell lysate at 20 μg

Lane 3 : U-87 MG cell lysate at 20 μg

Lane 4 : Huh7 cell lysate at 20 μg

Lane 5: HepG2 cell lysate at 20 µg

Lane 6 : Wild Type HeLa cell lysate at 20 μg

Lane 7: ENPP2 knockout HeLa cell lysate at 20 µg

Lane 8: Empty cell lysate at 0 µg

Lane 9: Recombinant Human ENPP-2/Autotaxin Protein, CF cell

lysate at 0.5 µg

Lane 10 : Recombinant Human ENPP2/ATX Protein ab126918

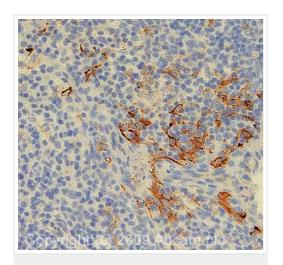
cell lysate at 0.5 µg

Performed under reducing conditions.

Predicted band size: 99 kDa

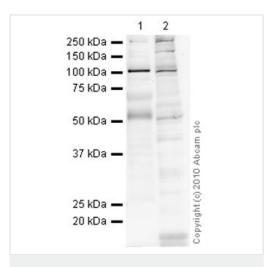
Observed band size: 105 kDa

Anti-ENPP2 antibody [1F8] (ab77104) staining at 1/1000 dilution, shown in green; Rabbit Anti-GAPDH antibody [EPR16891] (ab181602) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab77104 was shown to bind specifically to ENPP2. A band was observed at 105 kDa in wild-type HEK-293 cell lysates with no signal observed at this size in ENPP2 knockout cell line. To generate this image, wild-type and ENPP2 knockout HEK-293 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) 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-Mouse lgG H&L 800CW and Goat anti-Rabbit lgG H&L 680RD at 1/20000 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ENPP2/ATX antibody [1F8] (ab77104)

IHC image of ENPP2/ATX staining in Human Tonsil FFPE section, performed on a BondTM system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab77104, 1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX



Western blot - Anti-ENPP2/ATX antibody [1F8] (ab77104)

All lanes: Anti-ENPP2/ATX antibody [1F8] (ab77104) at 10 μg/ml

 $\textbf{Lane 1:} \ \text{Human kidney tissue lysate - total protein } (\underline{\textbf{ab30203}})$

Lane 2: HEK293 (Human embryonic kidney cell line) Whole Cell Lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat polyclonal to Mouse IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Developed using the ECL technique.

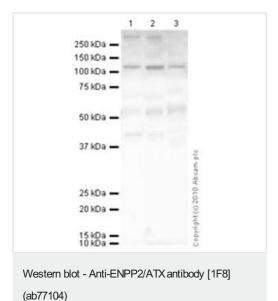
Performed under reducing conditions.

Predicted band size: 99 kDa **Observed band size:** 100 kDa

Additional bands at: 180 kDa, 250 kDa, 50 kDa, 65 kDa. We are

unsure as to the identity of these extra bands.

Exposure time: 20 minutes



All lanes: Anti-ENPP2/ATX antibody [1F8] (ab77104) at 10 μg/ml

Lane 1: Human placenta tissue lysate - total protein (ab29745)

Lane 2: Human ovary tissue lysate - total protein (ab30222)

Lane 3: Human small intestine tissue lysate - total protein

(ab29276)

Lysates/proteins at 20 µg per lane.

Secondary

All lanes: Mouse IgG - H&L - Pre-Adsorbed (HRP) at 1/3000

dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 99 kDa **Observed band size:** 110 kDa

Additional bands at: 300 kDa, 40 kDa, 55 kDa. We are unsure as

to the identity of these extra bands.

Exposure time: 20 minutes

ENPP2/ATX contains three potential glycosylation sites (SwissProt), which might explain its migration at a higher molecular weight than predicted.

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