

Product datasheet

Anti-ENPP2/ATX antibody [1F8] ab77104

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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, ICC/IF
Species reactivity	Reacts with: Mouse, Human
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. ICC/IF: HeLa cells.
General notes	<p>This antibody clone is manufactured by Abcam.</p> <p>If you require this antibody in a particular buffer formulation or a particular conjugate for your experiments, please contact orders@abcam.com or you can find further information here.</p> <p>This product was previously labelled as ENPP2</p>

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	<p>pH: 7.40</p> <p>Preservative: 0.02% Sodium azide</p> <p>Constituents: PBS, 6.97% L-Arginine</p>
Purity	IgG fraction
Clonality	Monoclonal
Clone number	1F8
Isotype	IgG1

Applications

Our [Abpromise guarantee](#) covers the use of **ab77104** in the following tested applications.

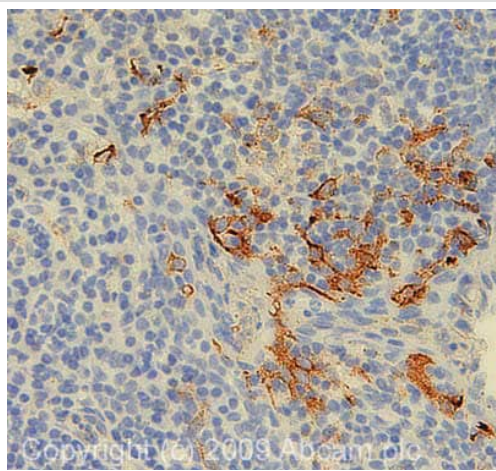
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

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).
ICC/IF		Use at an assay dependent concentration.

Target

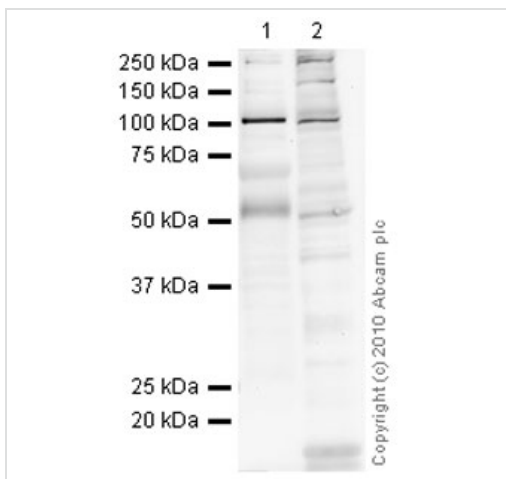
Function	Hydrolyzes lysophospholipids to produce lysophosphatidic acid (LPA) in extracellular fluids. Major substrate is lysophosphatidylcholine. Also can act on sphingosylphosphorylcholine producing sphingosine-1-phosphate, a modulator of cell motility. Can hydrolyze, in vitro, bis-pNPP, to some extent 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



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded 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

Lane 1 : Human kidney tissue lysate - total protein ([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.

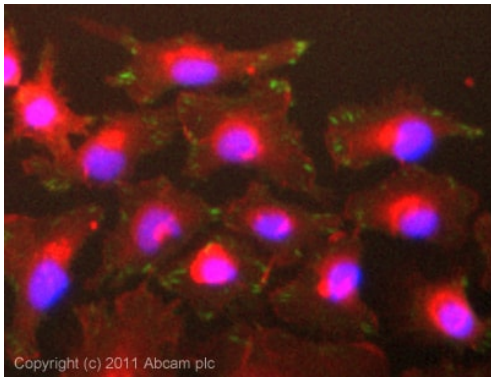
Predicted band size: 99 kDa

Observed band size: 100 kDa

[why is the actual band size different from the predicted?](#)

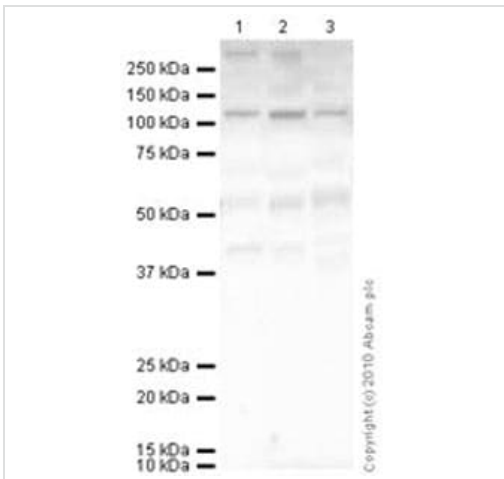
Additional bands at: 180 kDa, 250 kDa, 50 kDa, 65 kDa. We are unsure as to the identity of these extra bands.

Exposure time: 20 minutes



Immunocytochemistry/ Immunofluorescence - Anti-ENPP2/ATX antibody [1F8] (ab77104)

ICC/IF image of ab77104 stained HeLa cells. The cells were 100% methanol fixed (5 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab77104, 10µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-mouse IgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



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

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 [why is the actual band size different from the predicted?](#)

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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