abcam

ab223268

Anti-ENPP1/PC1 antibody [EPR22262-22]

Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268) is a rabbit monoclonal antibody detecting ENPP1/PC1 in **Western Blot, Flow Cytometry (Intra), IP, IHC-P, ICC/IF**. Suitable for **Human**.

- Biophysical QC for unrivalled batch-batch consistency

Recombinant | RabMAb | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR22262-22
Purification technique	Affinity purification Protein A
Concentration	0.437 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	1/30

Notes	-

WB

Tested

Species	Human
Dilution info	1/1000
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/100
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/50
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/500
Notes	Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0). Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Target data

See full target information ENPP1

Function	Nucleotide pyrophosphatase that generates diphosphate (PPi) and functions in bone mineralization and soft tissue calcification by regulating pyrophosphate levels (By similarity). PPi inhibits bone mineralization and soft tissue calcification by binding to nascent hydroxyapatite crystals, thereby preventing further growth of these crystals (PubMed:11004006). Preferentially hydrolyzes ATP, but can also hydrolyze other nucleoside 5' triphosphates such as GTP, CTP and UTP to their corresponding monophosphates with release of pyrophosphate, as well as diadenosine polyphosphates, and also 3',5'-cAMP to AMP (PubMed:25344812, PubMed:27467858, PubMed:28011303, PubMed:35147247, PubMed:8001561). May also be involved in the regulation of the availability of nucleotide sugars in the endoplasmic reticulum and Golgi, and the regulation of purinergic signaling (PubMed:27467858, PubMed:8001561). Inhibits ectopic joint calcification and maintains articular chondrocytes by repressing hedgehog signaling; it is however unclear whether hedgehog inhibition is direct or indirect (By similarity). Appears to modulate insulin sensitivity and function (PubMed:10615944). Also involved in melanogenesis (PubMed:28964717). Also able to hydrolyze 2',3'-cGAMP (cyclic GMP-AMP), a second messenger that activates TMEM173/STING and triggers type-I interferon production (PubMed:25344812). 2',3'-cGAMP degradation takes place in the lumen or extracellular space, and not in the cytosol where it is produced; the role of 2',3'-cGAMP hydrolysis is therefore unclear (PubMed:25344812). Not able to hydrolyze the 2',3'-cGAMP linkage isomer 3'-3'-cGAMP (PubMed:25344812).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

What is this antibody validated in?

Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunoprecipitation (IP), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF) in Human samples.

What is the molecular weight of ENPP1/PC1?

Anti-ENPP1/PC1 [EPR22262-22] (ab223268) specifically detects a band for ENPP1/PC1 (UniProt: P22413) at a molecular weight of 105kDa.

Other related products
We have a range of other formats of antibody clone [EPR22262-22] also available for your convenience: ab223268, Carrier free - ab240832, Alexa Fluor® 488 - ab312900, Alexa Fluor® 647 - ab315846

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	ENPP1 also known as PC-1 is a type II transmembrane glycoprotein with a molecular weight of approximately 130 kDa. It is expressed in various tissues including the liver kidney and bone. ENPP1 exhibits the enzymatic activity of nucleotide pyrophosphatase/phosphodiesterase which hydrolyzes ATP to AMP and diphosphate. This activity is important for the regulation of extracellular nucleotide levels and mineralization processes. Researchers often utilize the ENPP1 assay to study its function and activity.
Biological function summary	ENPP1 significantly influences bone mineralization and soft tissue calcification. It interacts with proteins like ectonucleoside triphosphate diphosphohydrolase-1 (ENTPD1) impacting cellular uptake of calcified materials. The protein is also involved in the regulatory networks responsible for controlling insulin signaling impacting metabolic processes. ENPP1 may form complexes with other proteins which can finely tune its biological activity and effects.
Pathways	ENPP1 plays a role in the insulin signaling pathway and the pyrophosphate pathway. Via its participation in insulin signaling ENPP1 regulates glucose metabolism and influences insulin sensitivity. It affects pathways related to mineral metabolism where it interacts with proteins like ENPP3. By regulating extracellular pyrophosphate levels ENPP1 contributes to inorganic phosphate homeostasis and influences skeletal development and maintenance.

Associated diseases and disorders	ENPP1 has associations with type 2 diabetes and generalized arterial calcification of infancy (GACI). Its role in type 2 diabetes relates to its ability to inhibit insulin receptor function therefore contributing to insulin resistance. ENPP1 also connects to proteins involved in vascular calcification such as matrix Gla protein (MGP) in the context of GACI. These associations highlight the importance of ENPP1 in metabolic and calcification disorders.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

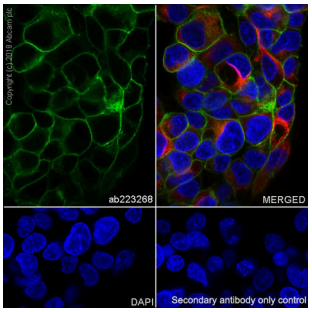
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

8 product images

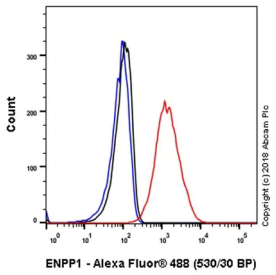


Immunocytochemistry/ Immunofluorescence - Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268)

ENPP1/PC1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-ENPP1/PC1 antibody

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HepG2 (human liver hepatocellular carcinoma cell line) cells labeling ENPP1/PC1 (green) with ab223268 at 1/100 dilution, followed by Alexa Fluor® 488 Goat anti-Rabbit secondary antibody (ab150077) at 1/1000 dilution. Confocal image showing membranous staining in HepG2 cells. Tubulin was detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889) at 1/200 dilution (red). The nuclear countertsain was DAPI (Blue).

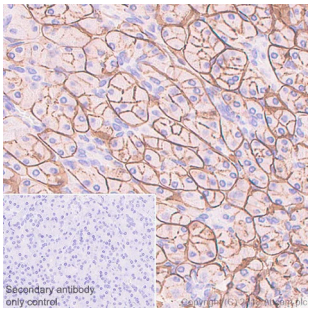
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Alexa Fluor® 488 Goat anti-Rabbit (ab150077) at 1/200 dilution.



Flow Cytometry (Intracellular) - Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268)

ENPP1/PC1 Flow Cytometry (Intracellular) staining using rabbit Anti-ENPP1/PC1 antibody

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized HepG2 (human liver hepatocellular carcinoma cell line) cells labeling ENPP1/PC1 with ab223268 (red) compared with a Rabbit monoclonal IgG isotype control (ab172730) (black)and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.

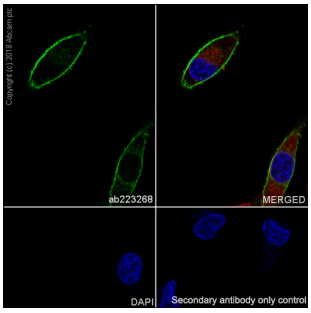


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268)

ENPP1/PC1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-ENPP1/PC1 antibody

Immunohistochemical analysis of paraffin-embedded human pancreas tissue stained for ENPP1/PC1 using ab223268 at 1/500 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Membranous staining on human pancreas (PMID: 9344668; PMID: 23861746) is observed. Counter stained with Hematoxylin.
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) ready to use.

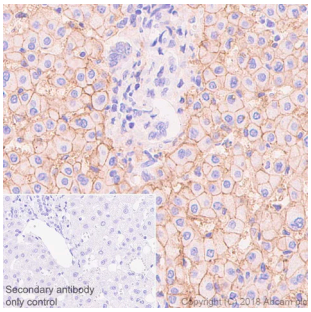
Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).



Immunocytochemistry/ Immunofluorescence - Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268)

ENPP1/PC1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-ENPP1/PC1 antibody

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized MDA-MB-231 (human breast adenocarcinoma cell line) cells labeling ENPP1/PC1 (green) with ab223268 at 1/100 dilution, followed by Alexa Fluor® 488 Goat anti-Rabbit secondary antibody (ab150077) at 1/1000 dilution. Confocal image showing membranous staining in MDA-MB-231 cells. Tubulin was detected using Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889) at 1/200 dilution (Red). The nuclear countertsain was DAPI (Blue).
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Alexa Fluor® 488 Goat anti-Rabbit (ab150077) at 1/200 dilution.

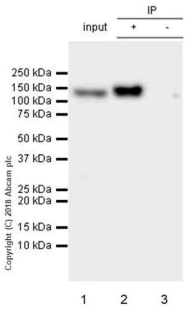


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268)

ENPP1/PC1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-ENPP1/PC1 antibody

Immunohistochemical analysis of paraffin-embedded human liver tissue stained for ENPP1/PC1 using ab223268 at 1/500 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Membranous staining on human liver (PMID: 25539584; PMID: 9344668; PMID: 23861746) is observed. Counter stained with Hematoxylin.
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) ready to use.

Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).



Immunoprecipitation - Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268)

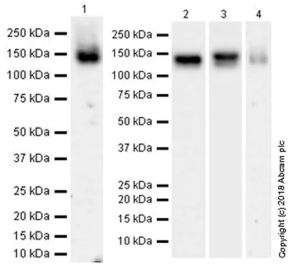
ENPP1/PC1 was immunoprecipitated from 0.35 mg of MDA-MB-231 (human breast adenocarcinoma cell line) whole cell lysate using ab223268 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab223268 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366) was used for detection at 1/5000 dilution.
Lane 1: MDA-MB-231 whole cell lysate 10µg (input)
Lane 2: ab223268 IP in MDA-MB-231 whole cell lysate (+).
Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab223268 in MDA-MB-231 whole cell lysate (-).

Blocking and dilution buffer and concentration: 5% NFDM/TBST.
Exposure time: 30 seconds.

All lanes:
Immunoprecipitation - Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268)

Predicted band size: 105 kDa

Observed band size: 130 kDa



Western blot - Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268)

Blocking and dilution buffer: 5% NFDM/TBST
Exposure time:

Lane 1:3 minutes;
Lane 2:70 seconds;
Lane 3:15 seconds;
Lane 4:3 minutes

The molecular weight observed is consistent with what has been described in the literature (PMID: 19577557).

All lanes:
Western blot - Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268) at 1/1000 dilution

Lane 1:
Human kidney tissue lysate at 20 µg

Lane 2:
MDA-MB-231 (human breast adenocarcinoma cell line) whole cell lysate at 20 µg

Lane 3:
HepG2 (human liver hepatocellular carcinoma cell line) whole cell lysate at 20 µg

Lane 4:
Huh7 (human hepatocellular carcinoma epithelial cell) whole cell lysate at 20 µg

Secondary

Lane 1:
Western blot - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/1000 dilution

Lanes 2 - 4:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 105 kDa

Observed band size: 130 kDa

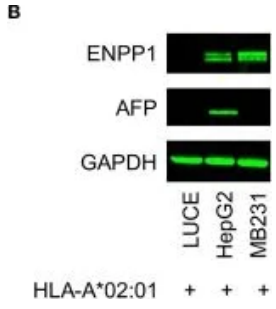


Image collected and cropped by CiteAb under a CC-BY license from the publication

Western blot - Anti-ENPP1/PC1 antibody [EPR22262-22] (ab223268)

ENPP1/PC1 western blot using anti-ENPP1/PC1 antibody [EPR22262-22] ab223268. Publication image and figure legend from Luo, X., Cui, H., et al., 2020, Front Immunol, PubMed 32425926.

ab223268 was used in this publication in western blot. This may not be the same as the application(s) guaranteed by Abcam. For a full list of applications guaranteed by Abcam for ab223268 please see the product overview.

AFP TCR 2 is unlikely to cross-react with other proteins in the human genome. (A) Graphic presentation of X-scan results showing the replaceability of each amino acid of AFP158−166 epitope. Peptides with replacement of each amino acid residue with every other 19 amino acids were used to activate the TCR 2 transduced T cells. The amount of IFN-γ produced by AFP TCR 2 transduced T cells after stimulation with mutated epitope (mut) was compared to that from wild type AFP158−166 peptide (wt) and the ratios are presented. Dashed line indicates the cut-off for tolerance. Data represents the mean + s.d. of duplicate co-cultures and is representative of 3 healthy donors. (B) ENPP1 and AFP expression in the indicated cell lines tested by Western blot. GAPDH was used as loading control. (C) IFN-γ concentrations in the supernatant of overnight, 1:1 ratio co-culture of UT or AFP TCR 2 transduced T cells with the indicated cell lines measured by ELISA. Data represents the mean + s.d. of quadruplicate co-cultures and is representative of 3 healthy donors.

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