abcam

ab134169

Anti-proCathepsin D antibody [EPR3054]

Recombinant | RabMAb | KO Validated

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	EPR3054
Purification technique	Affinity purification Protein A
Dissociation constant	1.16 x 10^{-10} M
Concentration	0.31 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	1/100 - 1/250
Notes	-

WB

Tested

Species	Human
Dilution info	1/1000 - 1/10000
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/100
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

Target data

See full target information CTSD

Function	Acid protease active in intracellular protein breakdown. Plays a role in APP processing following cleavage and activation by ADAM30 which leads to APP degradation (PubMed:27333034). Involved in the pathogenesis of several diseases such as breast cancer and possibly Alzheimer disease.

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage conditions	+4°C

Appropriate long-term storage conditions	-20°C
Storage information	Stable for 12 months at -20°C

Notes

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

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.

Supplementary info

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

Activity summary	Procathepsin D also known as CTSD is a precursor form of the enzyme Cathepsin D. It has a molecular mass of approximately 52 kDa. Procathepsin D is an aspartic protease expressed mainly in lysosomes but also found in many tissues like liver spleen and the endometrium. Its precursor form undergoes post-translational modification to become active involving the removal of its propeptide segment.
Biological function summary	This protease plays significant roles in protein degradation and processing within lysosomes. Procathepsin D remains part of lysosomal enzyme complexes influencing lysosomal balance and turnover of proteins. It contributes to the regulation of cell growth apoptosis and protein activation. Also it impacts antigen presentation and immunity showing importance in cellular maintenance and function.
Pathways	Procathepsin D participates in the lysosomal degradation pathway impacting autophagy and proteostasis. It interfaces with kinases and caspases routing signaling and apoptosis pathways. Through the lysosomal protein pathway it relates to proteins like Cathepsin B and L which also deal with proteolysis. Procathepsin D's degradation function influences metabolic processes interconnected with the immune response and tissue remodeling.

Associated diseases and disorders	Procathepsin D shows connections to breast cancer and Alzheimer's disease. Elevated levels have been observed in breast cancer linking it with tumor progression where it may interact with proteins like cathepsin B. In Alzheimer's disease altered procathepsin D activity affects amyloid processing and neuroinflammation associating closely with proteins like amyloid precursor protein (APP) and other cathepsins involved in neurodegenerative processes.

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

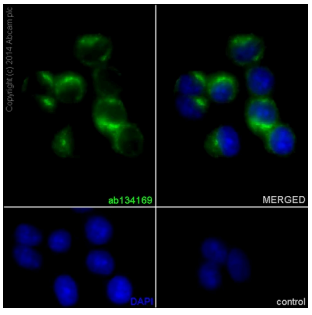
We do not recommend this combination. It is not covered by our product promise.

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.

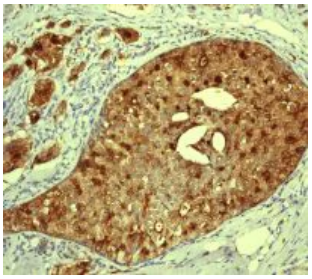
8 product images



Immunocytochemistry/ Immunofluorescence - Anti-proCathepsin D antibody [EPR3054] (ab134169)

ProCathepsin D Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-proCathepsin D antibody

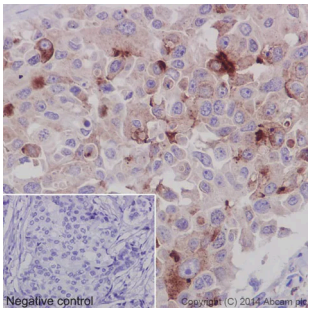
Immunofluorescence staining of MCF7 cells with purified ab134169 at a working dilution of 1 in 50, counter-stained with DAPI. The secondary antibody was Alexa Fluor® 488 goat anti rabbit (ab150077), used at a dilution of 1 in 500. The cells were fixed in 4% PFA and permeabilized using 0.1% Triton X 100. The negative control is shown in bottom right hand panel - for the negative control, purified ab134169 was used at a dilution of 1/200 followed by an Alexa Fluor® 594 goat anti-mouse antibody at a dilution of 1/500.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-proCathepsin D antibody [EPR3054] (ab134169)

ProCathepsin D Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-proCathepsin D antibody

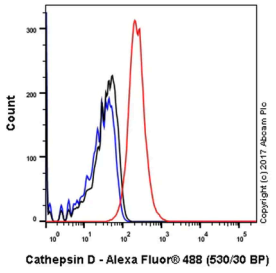
Immunohistochemical analysis of paraffin-embedded Human breast ductal infiltrating carcinoma tissue, staining proCathepsin D using unpurified ab134169 at a 1/250 dilution.
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-proCathepsin D antibody [EPR3054] (ab134169)

ProCathepsin D Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-proCathepsin D antibody

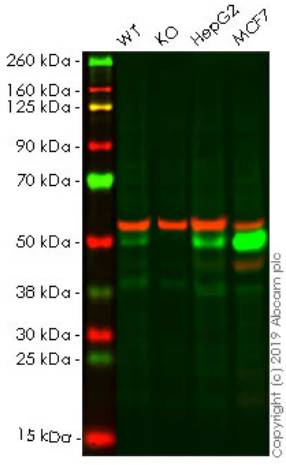
Immunohistochemical staining of paraffin embedded human breast carcinoma with purified ab134169 at a working dilution of 1 in 50. The secondary antibody used is a HRP polymer for rabbit IgG. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



Flow Cytometry (Intracellular) - Anti-proCathepsin D antibody [EPR3054] (ab134169)

ProCathepsin D Flow Cytometry (Intracellular) staining using rabbit Anti-proCathepsin D antibody

Intracellular Flow Cytometry analysis of A431 (human epidermoid carcinoma) cells labeling proCathepsin D with purified ab134169 at 1/100 dilution (10ug/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor® 488) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal IgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) were used as the unlabeled control.



Western blot - Anti-proCathepsin D antibody [EPR3054] (ab134169)

ProCathepsin D Western blot staining using rabbit Anti-proCathepsin D antibody

Lanes 1 - 4: Merged signal (red and green). Green - ab134169 observed at 46 kDa. Red - loading control, ab7291 (mouse anti-tubulin), observed at 50 kDa.
ab134169 was shown to recognize in wild-type A431 cells as signal was lost at the expected MW in CTSD knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and CTSD knockout samples were subjected to SDS-PAGE. The membrane was blocked with 3% NF Milk. ab134169 and ab7291 (Mouse anti-tubulin loading control) were incubated overnight at 4°C at 1/2000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-proCathepsin D antibody [EPR3054] (ab134169) at 1/2000 dilution

Lane 1:
Wild-type A-431 (Human epidermoid carcinoma cell line) whole cell lysate at 40 µg

Lane 2:
Western blot - Human CTSD knockout A-431 cell lysate (ab261700) at 40 µg

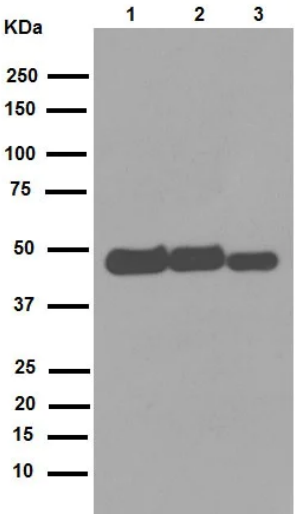
Lane 3:
Hep G2 (Human liver hepatocellular carcinoma cell line) whole cell lysate at 40 µg

Lane 4:
MCF7 (Human breast adenocarcinoma cell line) whole cell lysate at 40 µg

Performed under reducing conditions.

Predicted band size: 44 kDa, 45 kDa

Observed band size: 46 kDa



Western blot - Anti-proCathepsin D antibody [EPR3054] (ab134169)

ProCathepsin D Western blot staining using rabbit Anti-proCathepsin D antibody

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST

All lanes:
Western blot - Anti-proCathepsin D antibody [EPR3054] (ab134169) at 1/2000 dilution

Lane 1:
MCF-7 cell lysate at 20 µg

Lane 2:
A431 cell lysate at 20 µg

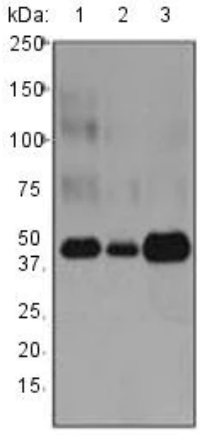
Lane 3:
SKBR-3 cell lysate at 20 µg

Secondary

All lanes:
HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 45 kDa

Observed band size: 44 kDa



Western blot - Anti-proCathepsin D antibody [EPR3054] (ab134169)

All lanes:
Western blot - Anti-proCathepsin D antibody [EPR3054] (ab134169) at 1/2000 dilution

Lane 1:
MCF7 cell lysate at 10 µg

Lane 2:
A431 cell lysate at 10 µg

Lane 3:

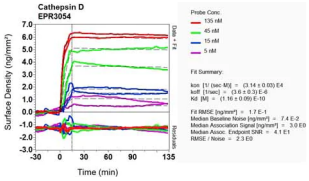
SKBR3 cell lysate at 10 µg

Secondary

All lanes:
Standard HRP labelled goat anti-rabbit at 1/2000 dilution

Developed using the ECL technique.

Predicted band size: 45 kDa



OI-RD Scanning - Anti-proCathepsin D antibody [EPR3054] (ab134169)

We have systematically measured KD (the equilibrium dissociation constant between the antibody and its antigen), of more than 840 recombinant antibodies to assess not only their individual KD values but also to see the average affinity of antibody. Based on the comparison with published literature values for mouse monoclonal antibodies, Recombinant antibodies appear to be on average 1-2 order of magnitude higher affinity.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.