abcam

Product datasheet

Anti-Cathepsin D antibody [EPR3056Y] - BSA and Azide free ab207550

KO VALIDATED Recombinant RabMAb

6 Images

Overview

Product name	Anti-Cathepsin D antibody [EPR3056Y] - BSA and Azide free
Description	Rabbit monoclonal [EPR3056Y] to Cathepsin D - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** IHC-P, ICC/IF, WB, Flow Cyt (Intra)
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: A431, HepG2 and MCF7 whole cell lysate. IHC-P: Human thyroid cancer tissue. ICC/IF: MCF-7 cells. Flow Cyt (intra): MCF-7 cells.
General notes	ab207550 is the carrier-free version of **ab75811**. Our **carrier-free** 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® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® 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® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

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

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR3056Y
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab207550 in the following tested applications.

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

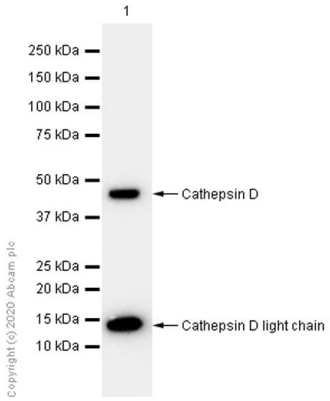
Application	Abreviews	Notes
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**.
ICC/IF		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 46 kDa (predicted molecular weight: 46 kDa).
Flow Cyt (Intra)		Use at an assay dependent concentration.

Target

Function	Acid protease active in intracellular protein breakdown. Involved in the pathogenesis of several diseases such as breast cancer and possibly Alzheimer disease.
Tissue specificity	Expressed in the aorta extrcellular space (at protein level).
Involvement in disease	Ceroid lipofuscinosis, neuronal, 10
Sequence similarities	Belongs to the peptidase A1 family. Contains 1 peptidase A1 domain.
Post-translational modifications	N- and O-glycosylated.

Cellular localization Lysosome. Melanosome. Secreted, extracellular space. Identified by mass spectrometry in melanosome fractions from stage I to stage IV. In aortic samples, detected as an extracellular protein loosely bound to the matrix (PubMed:20551380).

Images



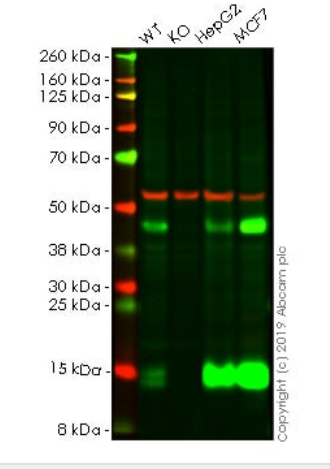
Western blot - Anti-Cathepsin D antibody [EPR3056Y] - BSA and Azide free (ab207550)

Anti-Cathepsin D antibody [EPR3056Y] (**ab75811**) at 1/1000 dilution + MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Secondary
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/20000 dilution

Predicted band size: 46 kDa

This data was developed using **ab75811**, the same antibody clone in a different buffer formulation.



Western blot - Anti-Cathepsin D antibody [EPR3056Y] - BSA and Azide free (ab207550)

All lanes : Anti-Cathepsin D antibody [EPR3056Y] (**ab75811**) at 1/1000 dilution (Unpurified)

Lane 1 : Wild-type A-431 (Human epidermoid carcinoma cell line) whole cell lysate
Lane 2 : CTSD knockout A-431 (Human epidermoid carcinoma cell line) whole cell lysate
Lane 3 : Hep G2 (Human liver hepatocellular carcinoma cell line) whole cell lysate
Lane 4 : MCF7 (Human breast adenocarcinoma cell line) whole cell lysate

Lysates/proteins at 40 µg per lane.

Performed under reducing conditions.

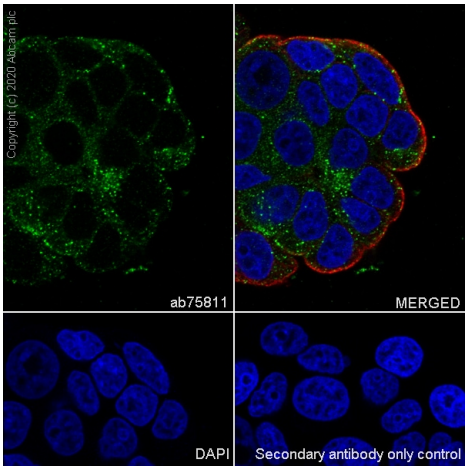
Predicted band size: 46 kDa
Observed band size: 44 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab75811**

observed at 44 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

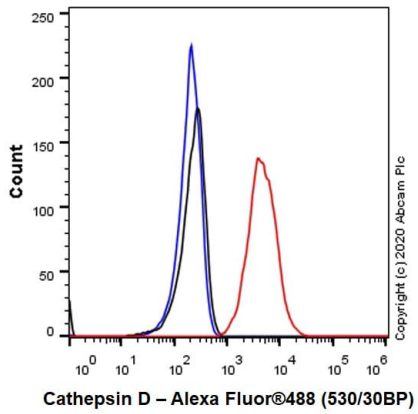
ab75811 was shown to specifically react with in wild-type A431 cells as signal was lost in CTSD knockout cells. Wild-type and CTSD knockout samples were subjected to SDS-PAGE. The membrane was blocked with 3% NF Milk. Ab75811 and **ab8245** (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab75811**).



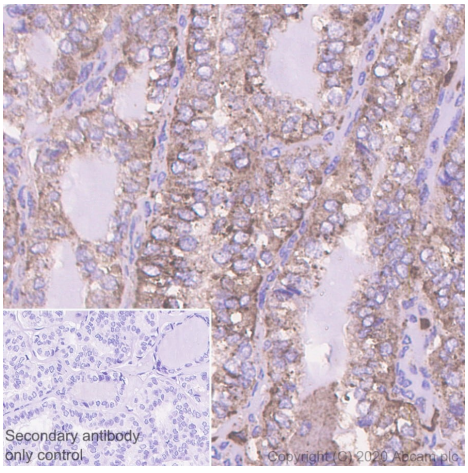
Immunocytochemistry/ Immunofluorescence - Anti-Cathepsin D antibody [EPR3056Y] - BSA and Azide free (ab207550)

This data was developed using **ab75811**, the same antibody clone in a different buffer formulation.
Immunocytochemistry analysis of MCF7 (Human breast adenocarcinoma epithelial cell) cells labeling Cathepsin D with purified **ab75811** at 1/50 dilution (1.88 µg/mL). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5 µg/mL). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 (2 µg/mL) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Flow Cytometry (Intracellular) - Anti-Cathepsin D antibody [EPR3056Y] - BSA and Azide free (ab207550)

This data was developed using **ab75811**, the same antibody clone in a different buffer formulation. Intracellular Flow Cytometry analysis of MCF7 (Human breast adenocarcinoma epithelial cell) cells labeling Cathepsin D with purified **ab75811** at 1/20 dilution (10 µg/mL) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cathepsin D antibody [EPR3056Y] - BSA and Azide free (ab207550)

This data was developed using **ab75811**, the same antibody clone in a different buffer formulation.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human thyroid cancer tissue sections labeling Cathepsin D with purified **ab75811** at 1/100 dilution (0.94 µg/mL). Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 1 (pH 6.0). Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

Anti-Cathepsin D antibody [EPR3056Y] - BSA and Azide free (ab207550)

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