# abcam

# Product datasheet

# Anti-Cathepsin D antibody [EPR3056Y] ab75811



ייבעדיו RabMAb

★★★★★ 1 Abreviews 2 References 画像数 6

#### 製品の概要

製品名 Anti-Cathepsin D antibody [EPR3056Y]

製品の詳細 Rabbit monoclonal [EPR3056Y] to Cathepsin D

由来種 Rabbit

アプリケーション 適用あり: Flow Cyt (Intra), ICC/IF, WB, IHC-P

種交差性 交差種: Human

免疫原 Synthetic peptide within Human Cathepsin D aa 50-150 (N terminal). The exact sequence is

ポジティブ・コントロール WB: A431, HepG2 and MCF7 whole cell lysate. MCF7 (Human breast adenocarcinoma epithelial

cell) whole cell lysate. IHC-P: Human thyroid cancer tissue. ICC/IF: MCF-7 cells. Flow Cyt (intra):

MCF-7 cells.

特記事項 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.

#### 製品の特性

製品の状態 Liquid

保存方法 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.

バッファー pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 0.05% BSA, 40% Glycerol (glycerin, glycerine), 59% PBS

精製度 Protein A purified

**ポリ**Æノ モノクローナル **ウローン名** EPR3056Y

アイソタイプ lgG

# アプリケーション

**The Abpromise guarantee** <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab75811の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		1/20.
ICC/IF		1/50. For unpurified use at 1/100 - 1/250.
WB		1/1000. Detects a band of approximately 44 kDa (predicted molecular weight: 44 kDa).  For unpurified use at 1/1000 - 1/2000.
IHC-P	<b>★★★★</b> (1)	1/100. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.  See IHC antigen retrieval protocols.  For unpurified use at 1/500.

# ターゲット情報

機能 Acid protease active in intracellular protein breakdown. Involved in the pathogenesis of several

diseases such as breast cancer and possibly Alzheimer disease.

組織特異性 Expressed in the aorta extrcellular space (at protein level).

関連疾患
Ceroid lipofuscinosis, neuronal, 10
配列類似性
Belongs to the peptidase A1 family.
Contains 1 peptidase A1 domain.

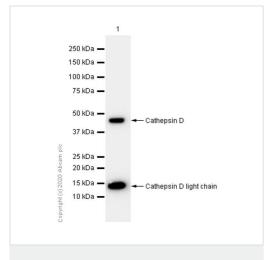
翻訳後修飾 N- and O-glycosylated.

**細胞内局在** 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).

#### 画像



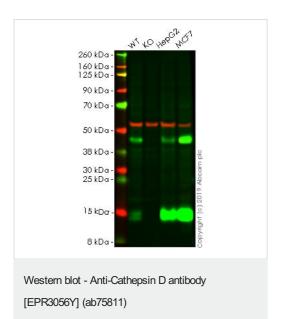
Western blot - Anti-Cathepsin D antibody [EPR3056Y] (ab75811)

Anti-Cathepsin D antibody [EPR3056Y] (ab75811) at 1/1000 dilution + MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysate at 15  $\mu$ g

# Secondary

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

Predicted band size: 44 kDa Observed band size: 14.44 kDa



**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: 44 kDa Observed band size: 44 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab75811 observed at 44 kDa. Red - loading control, **ab7291**, observed at 55 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 <a href="mailto:ab7291">ab7291</a> (Mouse anti Tubulin 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 <a href="mailto:ab216773">ab216773</a> and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed <a href="mailto:ab216776">ab216776</a> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

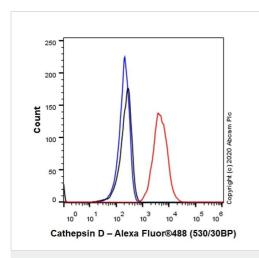
ab75811 MERGED

DAPI

Secondary antibody only control

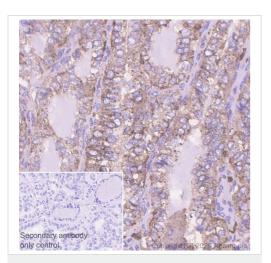
Immunocytochemistry/ Immunofluorescence - Anti-Cathepsin D antibody [EPR3056Y] (ab75811)

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 lgG (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.



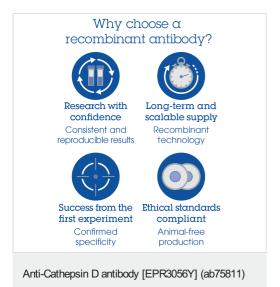
Flow Cytometry (Intracellular) - Anti-Cathepsin D antibody [EPR3056Y] (ab75811)

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 lgG (Alexa Fluor<sup>®</sup> 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal lgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cathepsin D antibody
[EPR3056Y] (ab75811)

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.



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