abcam

Product datasheet

Anti-Cathepsin D antibody [EPR3057Y] ab75852

ועלשעבע RabMAb

★★★★★ 11 Abreviews 94 References 画像数 11

製品の概要

製品名 Anti-Cathepsin D antibody [EPR3057Y]

製品の詳細 Rabbit monoclonal [EPR3057Y] 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, IP, IHC-P, IHC-Fr

種交差性 交差種: Mouse, Human

免疫原 Synthetic peptide within Human Cathepsin D aa 350 to the C-terminus (C terminal). The exact

sequence is proprietary.

ポジティブ・コントロール WB: MCF7, A431, SK-BR-3 and HepG2 whole cell lysate (ab7900) and mouse brain tissue

lysate. IHC-P: Human breast carcinoma and liver tissues. ICC/IF: MCF7 cells. IP: SK-BR-3 cell

lysate. Flow Cyt (intra): HepG2 cells. IHC-Fr: Hu liver tissue sections.

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**.

Rat: We have preliminary internal testing data to indicate this antibody may not react with this

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.

Stable for 12 months at -20°C.

バッファー pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

精製度 Protein A purified

ポリたノ モノクローナル **ウローン名** EPR3057Y

アイソタイプ IgG

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ab75852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)		Use at an assay dependent concentration.
ICC/IF	**** <u>(1)</u>	1/100 - 1/1000.
WB	★★★★★ (8)	1/2000 - 1/10000. Predicted molecular weight: 46 kDa.
IP		1/10 - 1/20.
IHC-P	****(1)	1/100 - 1/1000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols.
IHC-Fr	★★★★☆ (1)	1/1000.

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 [EPR3057Y] (ab75852)

All lanes : Anti-Cathepsin D antibody [EPR3057Y] (ab75852) at 1/5000 dilution (purified)

Lane 1 : MCF7 (Human breast adenocarcinoma cell line) whole cell lysate

Lane 2: SK-BR-3 (Human mammary gland adenocarcinoma cell line) whole cell lysate

Lane 3: HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

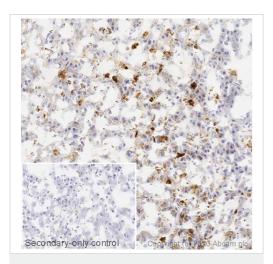
Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/10000 dilution

Predicted band size: 46 kDa

Observed band size: 28,43,46 kDa

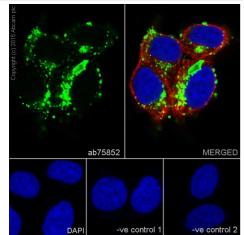
Blocking/Dilution buffer: 5% NFDM /TBST.



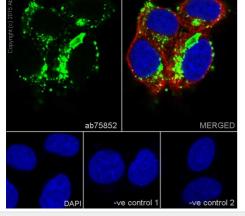
Immunohistochemistry (Frozen sections) - Anti-Cathepsin D antibody [EPR3057Y] (ab75852)

IHC image of Cathepsin D staining in a section of frozen normal human liver performed on a Leica BONDTM system using the standard protocol. The section was fixed in 10% paraformaldehyde (10 min) prior to staining. The section was incubated with ab75852, 1/1000 dilution, 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. The inset secondary-only control image is taken from an identical assay without primary antibody.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunocytochemistry/ Immunofluorescence - Anti-Cathepsin D antibody [EPR3057Y] (ab75852)



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human breast carcinoma tissue labeling Cathepsin D with purified ab75852 at a dilution of 1/100.

Control 2: <u>ab7291</u> (1/1000) and secondary antibody, <u>ab150077</u>, an Alexa Fluor® 488-conjugated goat anti-rabbit lgG (1/1000).

Control 1: primary antibody (1/100) and secondary antibody, ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG

Immunocytochemistry/Immunofluorescence analysis of MCF7 (Human breast adenocarcinoma cell line) cells labeling Cathepsin

Cells were fixed with 100% methanol and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat antirabbit IgG (1/1000) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. ab7291, a mouse antitubulin (1/1000) and ab150120, an Alexa Fluor® 594-conjugated

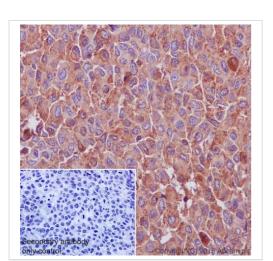
D with purified ab75852 at 1/100.

(1/1000).

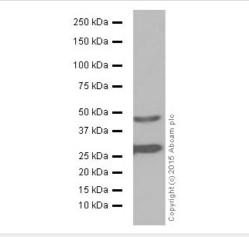
goat anti-mouse IgG (1/1000) were also used.

Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. ab97051, an HRP-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/500).

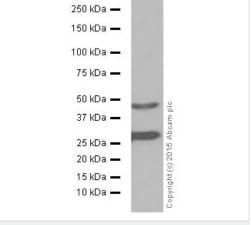
Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cathepsin D antibody [EPR3057Y] (ab75852)



Western blot - Anti-Cathepsin D antibody [EPR3057Y] (ab75852)



Blocking/Dilution buffer: 5% NFDM /TBST.

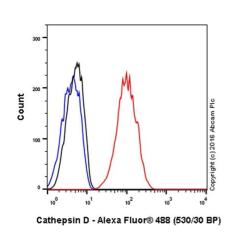
Predicted band size: 46 kDa

Observed band size: 28,43 kDa

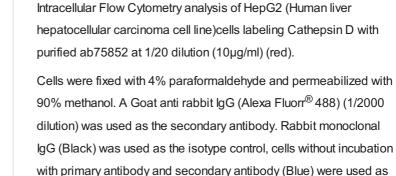
Anti-Cathepsin D antibody [EPR3057Y] (ab75852) at 1/5000

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/10000 dilution

dilution (purified) + Mouse brain tissue lysate at 20 µg



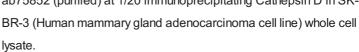
Flow Cytometry (Intracellular) - Anti-Cathepsin D antibody [EPR3057Y] (ab75852)



the unlabeled control.

Secondary

ab75852 (purified) at 1/20 immunoprecipitating Cathepsin D in SK-



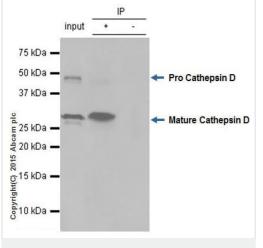


Lane 2 (+): ab75852 + SK-BR-3 whole cell lysate (10µg).

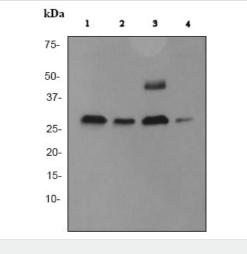
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab75852 in SK-BR-3 whole cell lysate.

For western blotting, VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10,000 dilution.





Immunoprecipitation - Anti-Cathepsin D antibody [EPR3057Y] (ab75852)



Western blot - Anti-Cathepsin D antibody [EPR3057Y] (ab75852)

All lanes : Anti-Cathepsin D antibody [EPR3057Y] (ab75852) at 1/10000 dilution (unpurified)

Lane 1 : MCF7 (Human breast adenocarcinoma cell line) cell lysate

Lane 2 : A431 (Human epidermoid carcinoma cell line) cell lysate

Lane 3 : SK-BR-3 (Human mammary gland adenocarcinoma cell

line) cell lysate

Lane 4 : HepG2 (Human liver hepatocellular carcinoma cell line) cell lysate

Lysates/proteins at 10 µg per lane.

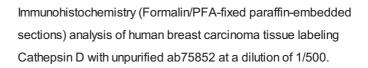
Secondary

All lanes: HRP-conjugated goat anti-rabbit lgG at 1/2000 dilution

Predicted band size: 46 kDa **Observed band size:** 46 kDa

Additional bands at: 28 kDa (possible cleavage fragment)

Bands at 28kDa are the Cathepsin D heavy chain.



Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cathepsin D antibody
[EPR3057Y] (ab75852)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cathepsin D antibody
[EPR3057Y] (ab75852)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human liver tissue labeling Cathepsin D with unpurified ab75852 at a dilution of 1/500.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



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