# abcam

# Product datasheet

# Anti-VCP antibody [EPR3307(2)] ab109240

יולצעבע RabMAb

★★★★★ 1 Abreviews 24 References 画像数 10

#### 製品の概要

製品名 Anti-VCP antibody [EPR3307(2)]

製品の詳細 Rabbit monoclonal [EPR3307(2)] to VCP

由来種 Rabbit

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

種交差性 交差種: Mouse, Rat, Human

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール MCF-7 cell lysate, HeLa whole cell lysate (ab150035), A549 cell lysate; SH-SY5Y cell lysate;

human breast carcinoma tissue; HeLa 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**.

#### 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.

Avoid freeze / thaw cycle. Stable for 12 months at -20°C.

バッファー pH: 7.20

Preservative: 0.01% Sodium azide

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

精製度 Protein A purified

ポリモノ モノクローナル クローン名 EPR3307(2)

アイソタイプ ΙgG

**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ab109240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
WB	<b>★★★★</b> (1)	1/10000. Predicted molecular weight: 89 kDa.  For unpurifed use at 1/10000-1/50000.
IP		1/10 - 1/100.
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol. See <b>IHC antigen retrieval protocols</b> .
ICC/IF		1/500. For unpurified use at 1/100 - 1/250.
Flow Cyt (Intra)		1/300. For unpurified use at 1/10 - 1/100. <u>ab172730</u> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.

#### ターゲット情報

#### 機能

Necessary for the fragmentation of Golgi stacks during mitosis and for their reassembly after mitosis. Involved in the formation of the transitional endoplasmic reticulum (tER). The transfer of membranes from the endoplasmic reticulum to the Golgi apparatus occurs via 50-70 nm transition vesicles which derive from part-rough, part-smooth transitional elements of the endoplasmic reticulum (tER). Vesicle budding from the tER is an ATP-dependent process. The ternary complex containing UFD1L, VCP and NPLOC4 binds ubiquitinated proteins and is necessary for the export of misfolded proteins from the ER to the cytoplasm, where they are degraded by the proteasome. The NPLOC4-UFD1L-VCP complex regulates spindle disassembly at the end of mitosis and is necessary for the formation of a closed nuclear envelope (By similarity). Regulates E3 ubiquitin-protein ligase activity of RNF19A.

## 関連疾患

Defects in VCP are the cause of inclusion body myopathy with early-onset Paget disease and frontotemporal dementia (IBMPFD) [MIM:167320]; also known as muscular dystrophy, limb-girdle, with Paget disease of bone or pagetoid amyotrophic lateral sclerosis or pagetoid neuroskeletal syndrome or lower motor neuron degeneration with Paget-like bone disease. IBMPFD features adult-onset proximal and distal muscle weakness (clinically resembling limb girdle muscular dystrophy), early-onset Paget disease of bone in most cases and premature frontotemporal dementia.

#### 配列類似性

Belongs to the AAA ATPase family.

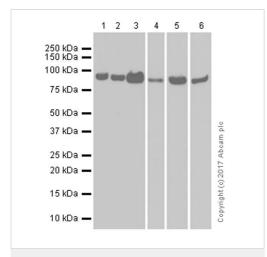
# 翻訳後修飾

Phosphorylated by tyrosine kinases in response to T-cell antigen receptor activation (By similarity). Phosphorylated upon DNA damage, probably by ATM or ATR. ISGylated.

#### 細胞内局在

Cytoplasm > cytosol. Nucleus. Present in the neuronal hyaline inclusion bodies specifically found in motor neurons from amyotrophic lateral sclerosis patients. Present in the Lewy bodies

#### 画像



Western blot - Anti-VCP antibody [EPR3307(2)] (ab109240)

**All lanes :** Anti-VCP antibody [EPR3307(2)] (ab109240) at 1/10000 dilution (purified)

**Lane 1 :** HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates

**Lane 2**: A549 (Human lung carcinoma epithelial cell) whole cell lysates

Lane 3: C6 (Rat glial tumor glial cell) whole cell lysates)

Lane 4: NIH/3T3 (Mouse embryonic fibroblast) whole cell lysates

Lane 5: RAW264.7 (Mouse Abelson murine leukemia virus-

induced tumor macrophage) whole cell lysates

Lane 6 : SH-SY5Y (Human neuroblastoma epithelial cell) whole cell lysates

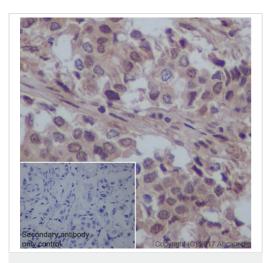
Lysates/proteins at 20 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

**Predicted band size:** 89 kDa **Observed band size:** 89 kDa

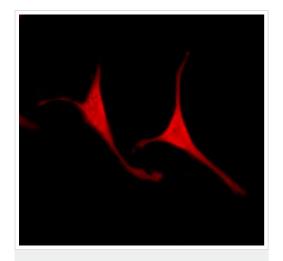
Blocking and diluting buffer: 5% NFDM/TBST



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-VCP antibody

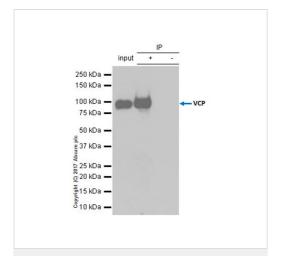
[EPR3307(2)] (ab109240)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human breast carcinoma tissue sections labeling VCP with purified ab109240 at 1:250 dilution (1.4 µg/ml). Heat mediated antigen retrieval was performed using EDTA Buffer, pH9.0. Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.



Immunocytochemistry/ Immunofluorescence - Anti-VCP antibody [EPR3307(2)] (ab109240)

Immunofluorescent staining of HeLa cells using unpurified ab109240 at 1/100 dilution.



Immunoprecipitation - Anti-VCP antibody [EPR3307(2)] (ab109240)

ab109240 (purified) at 1:20 dilution (2ug) immunoprecipitating VCP in HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate.

Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10ug

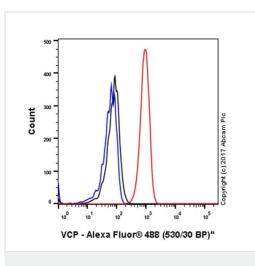
Lane 2 (+): ab109240 & HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 3 (-): Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab109240 in HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

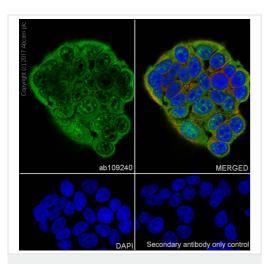
For western blotting, VeriBlot for IP Detection Reagent (HRP) (ab131366) was used for detection at 1:1000 dilution.

Blocking and diluting buffer: 5% NFDM/TBST.

Intracellular Flow Cytometry analysis of HL-60 (Human acute promyelocytic leukemia promyeloblast) cells labeling VCP with purified ab109240 at 1/300 dilution (1 ug/ml) (red). Cells were fixed with 4% Paraformaldehyde. A Goat anti rabbit lgG (Alexa Fluorr® 488) secondary antibody was used at 1/2000 dilution. Isotype control - 90% methanol. Unlabeled control - Rabbit monoclonal lgG (Black).Cell without incubation with primary antibody and secondary antibody (Blue).



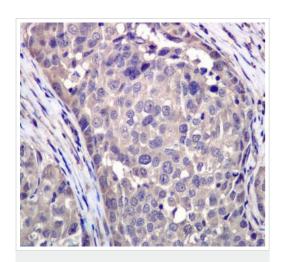
Flow Cytometry (Intracellular) - Anti-VCP antibody [EPR3307(2)] (ab109240)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-VCP antibody
[EPR3307(2)] (ab109240)

Immunocytochemistry/ Immunofluorescence analysis of MCF-7 (Human breast adenocarcinoma epithelial cell) cells labeling VCP with Purified ab109240 at 1:500 dilution (0.7μg/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<sup>®</sup> 594) 1:200 (2.5 μg/ml). ab150077 Goat anti rabbit lgG(Alexa Fluor<sup>®</sup> 488) was used as the secondary antibody at 1:1000 dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

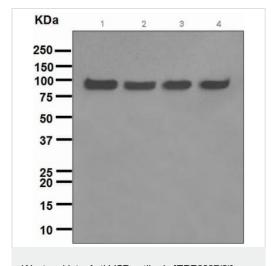
Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-VCP antibody
[EPR3307(2)] (ab109240)

Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue using unpurified ab109240 at 1/100 dilution.

Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.



Western blot - Anti-VCP antibody [EPR3307(2)] (ab109240)

**All lanes :** Anti-VCP antibody [EPR3307(2)] (ab109240) at 1/10000 dilution (unpurified)

Lane 1: MCF7 cell lysate

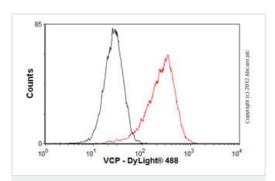
Lane 2 : HeLa cell lysate

Lane 3: A549 cell lysate

Lane 4: SH-SY5Y cell lysate

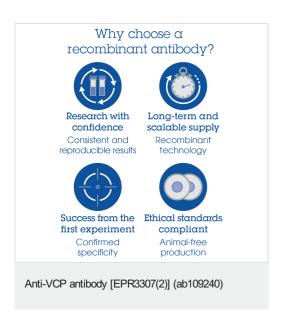
Lysates/proteins at 10 µg/ml per lane.

Predicted band size: 89 kDa



Flow Cytometry (Intracellular) - Anti-VCP antibody [EPR3307(2)] (ab109240)

Overlay histogram showing HL60 cells stained with unpurified ab109240 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab109240, 1/100) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit lgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) (1µg/1x10<sup>6</sup> cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HL60 cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



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