abcam

Product datasheet

Anti-ATP6V1A antibody [EPR19270] ab199326

ועלשעבע RabMAb

★★★★★ 2 Abreviews 12 References 画像数 13

製品の概要

製品名 Anti-ATP6V1A antibody [EPR19270]

製品の詳細 Rabbit monoclonal [EPR19270] to ATP6V1A

由来種 Rabbit

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

種交差性 交差種: Mouse, Rat, Human

免疫原 Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: Human fetal heart, fetal liver and fetal kidney lysates; HeLa, K562, HEK-293, C6, PC-12 and

> NIH/3T3 whole cell lysates; Mouse brain and kidney lysates; Rat brain and kidney lysates. IHC-P: Human kidney, Human thyroid cancer, mouse kidney and rat stomach tissues. ICC/IF: HeLa and

NIH/3T3 cells. Flow Cyt (intra): HeLa cells. IP: HeLa and NIH/3T3 whole cell lysates.

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 long

term. Avoid freeze / thaw cycle.

バッファー pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol, 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 EPR19270

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ab199326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)		1/120.
IHC-P		1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB	★★★★	1/2000. Detects a band of approximately 68 kDa (predicted molecular weight: 68 kDa).
ICC/IF		1/250.
IP		1/40.

ターゲット情報

機能 Catalytic subunit of the peripheral V1 complex of vacuolar ATPase. V-ATPase vacuolar ATPase

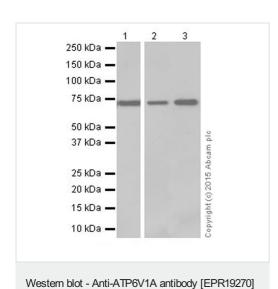
is responsible for acidifying a variety of intracellular compartments in eukaryotic cells.

組織特異性 Present in all tissues analyzed.

配列類似性 Belongs to the ATPase alpha/beta chains family.

画像

(ab199326)



All lanes: Anti-ATP6V1A antibody [EPR19270] (ab199326) at

1/2000 dilution

Lane 1 : Human fetal heart lysate

Lane 2 : Human fetal liver lysate

Lane 3 : Human fetal kidney lysate

Lane 9. Human letal Runey lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 68 kDa **Observed band size:** 68 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1: 3 minutes; Lane 2 and 3: 8 seconds.

1 2 3
250 kDa —
150 kDa —
100 kDa —
75 kDa —
37 kDa —
25 kDa —
20 kDa —
15 kDa —
10 kDa —
10 kDa —

Western blot - Anti-ATP6V1A antibody [EPR19270] (ab199326)

All lanes : Anti-ATP6V1A antibody [EPR19270] (ab199326) at 1/2000 dilution

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 2: K562 (Human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate

Lane 3: HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

Lysates/proteins at 20 µg per lane.

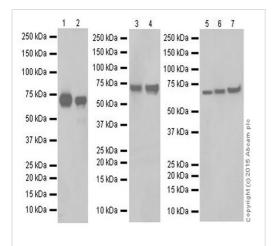
Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 68 kDa **Observed band size:** 68 kDa

Exposure time: 8 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-ATP6V1A antibody [EPR19270] (ab199326)

All lanes : Anti-ATP6V1A antibody [EPR19270] (ab199326) at 1/2000 dilution

Lane 1: Mouse brain lysate

Lane 2: Mouse kidney lysate

Lane 3: Rat brain lysate

Lane 4: Rat kidney lysate

Lane 5: C6 (Rat glial tumor cell line) whole cell lysate

Lane 6: PC-12 (Rat adrenal gland pheochromocytoma cell line)

whole cell lysate

Lane 7: NIH/3T3 (Mouse embryo fibroblast cell line) whole cell

lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at

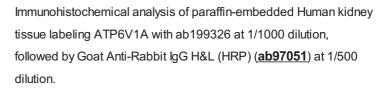
1/100000 dilution

Predicted band size: 68 kDa **Observed band size:** 68 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1 and 2: 4 seconds; Lane 3 and 4: 1

second; Lane 5,6 and 7:4 seconds.

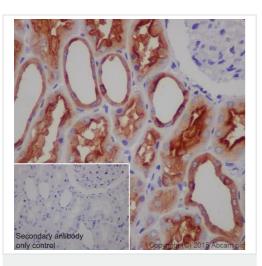


Cytoplasm staining on kidney tubules of the normal Human kidney is observed.

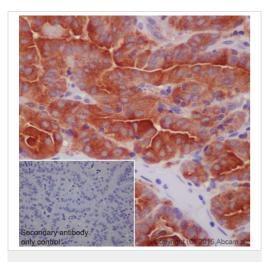
Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATP6V1A antibody
[EPR19270] (ab199326)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATP6V1A antibody
[EPR19270] (ab199326)

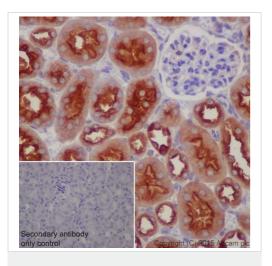
Immunohistochemical analysis of paraffin-embedded Human thyroid cancer tissue labeling ATP6V1A with ab199326 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Cytoplasm staining on tumor cells of the Human thyroid cancer is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATP6V1A antibody
[EPR19270] (ab199326)

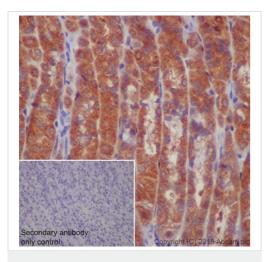
Immunohistochemical analysis of paraffin-embedded mouse kidney tissue labeling ATP6V1A with ab199326 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Cytoplasm staining on kidney tubules of the mouse kidney is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATP6V1A antibody
[EPR19270] (ab199326)

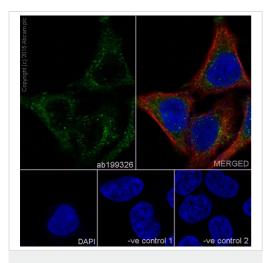
Immunohistochemical analysis of paraffin-embedded rat stomach tissue labeling ATP6V1A with ab199326 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Cytoplasm staining on rat stomach tissue is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-ATP6V1A antibody [EPR19270] (ab199326)

Immunofluorescent analysis of 100% methanol-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling ATP6V1A with ab199326 at 1/250 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing cytoplasmic staining on HeLa cell line.

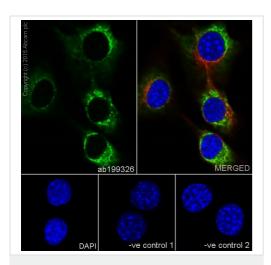
The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin antibody [EPR19270]-Loading Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse lgG H&L (Alexa Fluor® 594) preadsorbed (ab150120) at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab199326 at 1/250 dilution followed by $\underline{ab150120}$ at 1/1000 dilution.

-ve control 2: $\underline{ab7291}$ at 1/1000 dilution followed by $\underline{ab150077}$ at 1/1000 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-ATP6V1A antibody [EPR19270] (ab199326)

Immunofluorescent analysis of 100% methanol-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling ATP6V1A with ab199326 at 1/250 dilution, followed by Goat Anti-Rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing cytoplasmic staining on NIH/3T3 cell line.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin antibody [EPR19270] - Loading Control (**ab7291**) at 1/1000 dilution and Goat Anti-Mouse lgG H&L (Alexa Fluor® 594) preadsorbed (**ab150120**) at 1/1000 dilution (red).

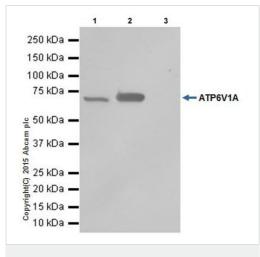
The negative controls are as follows:-

-ve control 1: ab199326 at 1/250 dilution followed by <u>ab150120</u> at 1/1000 dilution.

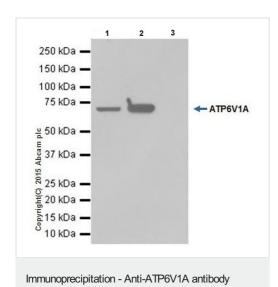
-ve control 2: $\underline{ab7291}$ at 1/1000 dilution followed by $\underline{ab150077}$ at 1/1000 dilution.

Flow Cytometry (Intracellular) - Anti-ATP6V1A antibody [EPR19270] (ab199326)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling ATP6V1A with ab199326 at 1/120 dilution (red) compared with a Rabbit lgG,monoclonal -lsotype Control (ab172730; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit lgG (FITC) at 1/500 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-ATP6V1A antibody [EPR19270] (ab199326)



[EPR19270] (ab199326)

ATP6V1A was immunoprecipitated from 1mg of HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate with ab199326 at 1/40 dilution.

Western blot was performed from the immunoprecipitate using ab199326 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>), was used for detection at 1/10000 dilution.

Lane 1: HeLa whole cell lysate 10µg (Input).

Lane 2: ab199326 IP in HeLa whole cell lysate.

Lane 3: Rabbit IgG,monoclonal [EPR19270] - Isotype
Control (ab172730) instead of ab199326 in HeLa whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 5 seconds.

ATP6V1A was immunoprecipitated from 1mg of NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell lysate with ab199326 at 1/40 dilution.

Western blot was performed from the immunoprecipitate using ab199326 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>), was used for detection at 1/10000 dilution.

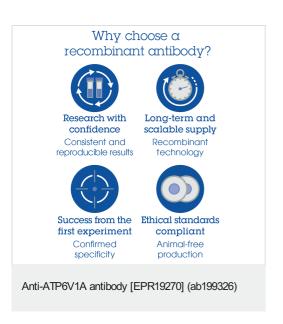
Lane 1: NIH/3T3 whole cell lysate 10µg (Input).

Lane 2: ab199326 IP in NIH/3T3 whole cell lysate.

Lane 3: Rabbit IgG,monoclonal [EPR19270] - Isotype
Control (<u>ab172730</u>) instead of ab199326 in NIH/3T3 whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 5 seconds.



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