abcam

Product datasheet

Anti-LSD2 / AOF1 antibody [EPR18508] ab193080



ייבער RabMAb

4 References 画像数 10

製品の概要

製品名 Anti-LSD2 / AOF1 antibody [EPR18508]

製品の詳細 Rabbit monoclonal [EPR18508] to LSD2 / AOF1

由来種 Rabbit

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

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 WB: HeLa, HAP1, HCT116, NIH/3T3, C6, RAW 264.7 and PC-12 cell lysates; Human fetal brain

and fetal heart lysates; Mouse thymus lysate. ICC/IF: HeLa and A431 cells. IP: K562 whole cell

lysate.

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: 7.20

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol (glycerin, glycerine), 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 **EPR18508**

アプリケーション

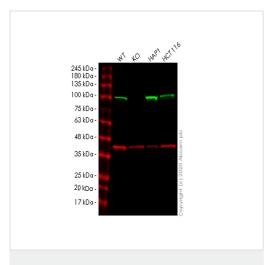
The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab193080の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		Use at an assay dependent concentration. Purified format.
ICC/IF		1/500.
IP		1/80.
WB		1/2000. Detects a band of approximately 92 kDa (predicted molecular weight: 92 kDa).

ターゲット情報

機能	Histone demethylase that demethylates 'Lys-4' of histone H3, a specific tag for epigenetic transcriptional activation, thereby acting as a corepressor. Required for de novo DNA methylation of a subset of imprinted genes during oogenesis. Acts by oxidizing the substrate by FAD to generate the corresponding imine that is subsequently hydrolyzed. Demethylates both mono- and di-methylated 'Lys-4' of histone H3. Has no effect on tri-methylated 'Lys-4', mono-, di- or tri-methylated 'Lys-9', mono-, di- or tri-methylated 'Lys-27', mono-, di- or tri-methylated 'Lys-36' of histone H3, or on mono-, di- or tri-methylated 'Lys-20' of histone H4.
配列類似性	Belongs to the flavin monoamine oxidase family. Contains 1 CW-type zinc finger. Contains 1 SWIRM domain.
ドメイン	The SWIRM domain may act as an anchor site for a histone tail.
細胞内局在	Nucleus.
製品の状態	There are 3 isoforms produced by alternative splicing.

画像



Western blot - Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080)

All lanes : Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: KDM1B knockout HeLa cell lysate

Lane 3 : HAP1 cell lysate

Lane 4 : HCT116 cell lysate

Lysates/proteins at 20 µg per lane.

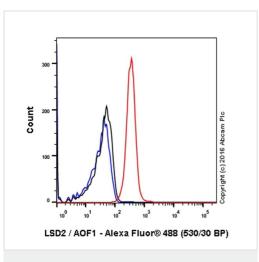
Secondary

All lanes : Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<u>ab216773</u>) at 1/10000 dilution

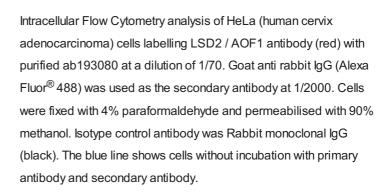
Predicted band size: 92 kDa
Observed band size: 95 kDa

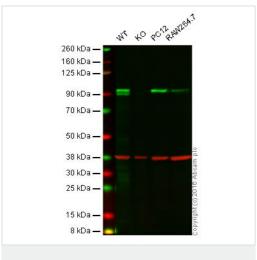
Lanes 1-4: Merged signal (red and green). Green - ab193080 observed at 95 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

ab193080 Anti-LSD2 / AOF1 antibody [EPR18508] was shown to specifically react with LSD2 / AOF1 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab265969 (knockout cell lysate ab258016) was used. Wild-type and LSD2 / AOF1 knockout samples were subjected to SDS-PAGE. ab193080 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated at room temperature for 2. 5 hours at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Flow Cytometry (Intracellular) - Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080)





Western blot - Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080)

Lane 1: Wild-type HAP1 cell lysate (20 µg)

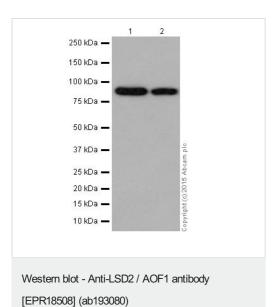
Lane 2: LSD2 / AOF1 knockout HAP1 cell lysate (20 µg)

Lane 3: PC12 cell lysate (20 µg)

Lane 4: Raw264.7 cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab193080 observed at 95 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab193080 was shown to specifically react with LSD2 / AOF1 when LSD2 / AOF1 knockout samples were used. Wild-type and LSD2 / AOF1 knockout samples were subjected to SDS-PAGE.
ab193080 and ab8245 (loading control to GAPDH) were diluted at 1/2000 and 1/10 000 respectively and incubated overnight at 4°C.
Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1/10 000 dilution for 1 h at room temperature before imaging.



All lanes : Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080) at 1/2000 dilution

Lane 1 : Human fetal brain lysate
Lane 2 : Human fetal heart lysate

Lysates/proteins at 10 µg per lane.

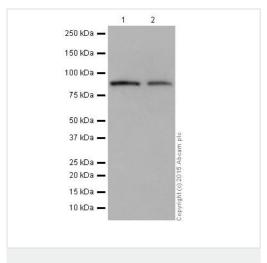
Secondary

All lanes : Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 92 kDa
Observed band size: 92 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080) **All lanes :** Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080) at 1/2000 dilution

Lane 1: NIH/3T3 (Mouse embyro fibroblast cells) cell lysate

Lane 2: Mouse thymus lysate

Lysates/proteins at 20 µg per lane.

Secondary

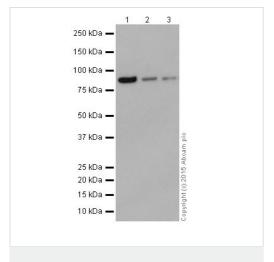
All lanes : Goat Anti-Rabbit lgG H&L (HRP) ($\underline{ab97051}$) at 1/50000

dilution

Predicted band size: 92 kDa Observed band size: 92 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080)

All lanes : Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080) at 1/2000 dilution

Lane 1: C6 (Rat glial tumor cells) cell lysate

Lane 2 : RAW 264.7 (Mouse macrophage cells transformed with Abelson murine leukemia virus) cell lysate

Abolison maine leakernia viras joeli lysate

Lane 3: PC-12 (Rat adrenal gland pheochromocytoma) cell lysate

Lysates/proteins at 10 µg per lane.

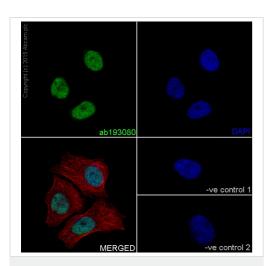
Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/1000 dilution

Predicted band size: 92 kDa Observed band size: 92 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Immunocytochemistry/ Immunofluorescence - Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling LSD2 / AOF1 with ab193080 at 1/500 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

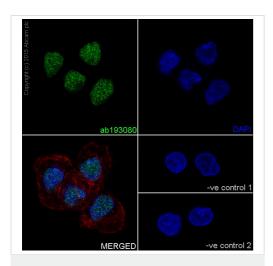
Confocal image showing nuclear staining on HeLa cell line.

The nuclear counterstain is DAPI (blue).

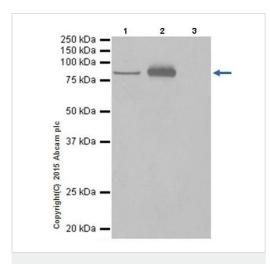
Tubulin is detected with <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution and <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 dilution (red).

The negative controls are as follows:

-ve control 1: ab193080 at 1/500 dilution followed by <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 dilution. -ve control 2: <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit lgG H&L) at 1/1000 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080)



Immunoprecipitation - Anti-LSD2 / AOF1 antibody [EPR18508] (ab193080)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized A431 (Human epidermoid carcinoma) cells labeling LSD2 / AOF1 with ab193080 at 1/500 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing nuclear staining on A431 cell line.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution and <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab193080 at 1/500 dilution followed by <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 dilution. -ve control 2: <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit lgG H&L) at 1/1000 dilution.

LSD2 / AOF1 was immunoprecipitated from 1mg of K562 (Human chronic myelogenous leukemia cells from bone marrow) whole cell lysate with ab193080 at 1/80 dilution.

Lane 1: K562 whole cell lysate 10ug (Input).

Lane 2: ab193080 IP in K562 whole cell lysate.

Lane 3: Rabbit monoclonal lgG ($\underline{ab172730}$) instead of ab193080 in K562 whole cell lysate.

Western blot was performed from the immunoprecipitate using ab193080 at 1/1000 dilution. Anti-Rabbit lgG (HRP), specific to the non-reduced form of lgG, was used as secondary antibody at 1/1500.

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

Exposure time: 8 seconds.



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