abcam

Product datasheet

Anti-LSD2 / AOF1 antibody [EPR18508] ab193080

KO VALIDATED Recombinant RabMAb

4 References 10 Images

Overview

Product name	Anti-LSD2 / AOF1 antibody [EPR18508]
Description	Rabbit monoclonal [EPR18508] to LSD2 / AOF1
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), ICC/IF, IP, WB
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	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.
General notes	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**.

Properties

Form	Liquid
Storage instructions	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.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR18508

Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab193080 in the following tested applications.

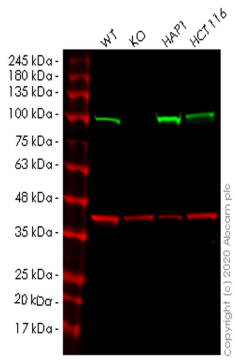
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
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).

Target

Function	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.
Sequence similarities	Belongs to the flavin monoamine oxidase family. Contains 1 CW-type zinc finger. Contains 1 SWIRM domain.
Domain	The SWIRM domain may act as an anchor site for a histone tail.
Cellular localization	Nucleus.
Form	There are 3 isoforms produced by alternative splicing.

Images



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

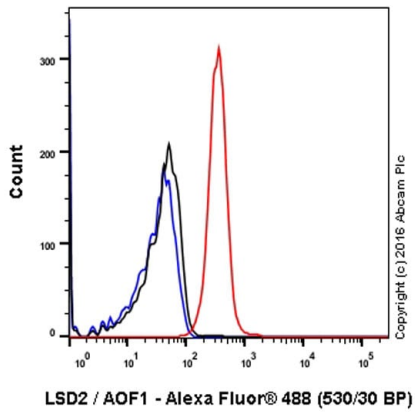
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/10000 dilution

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

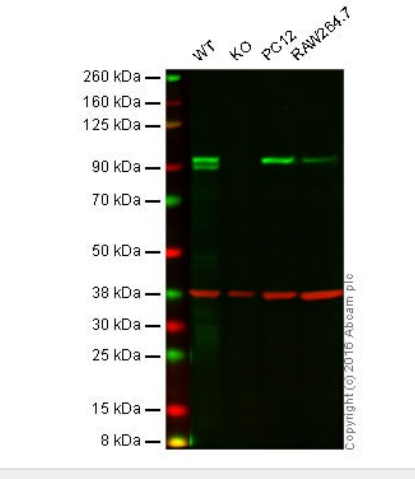
Lanes 1-4: Merged signal (red and green). Green - ab193080 observed at 95 kDa. Red - loading control **ab8245** observed at 36 kDa.

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)

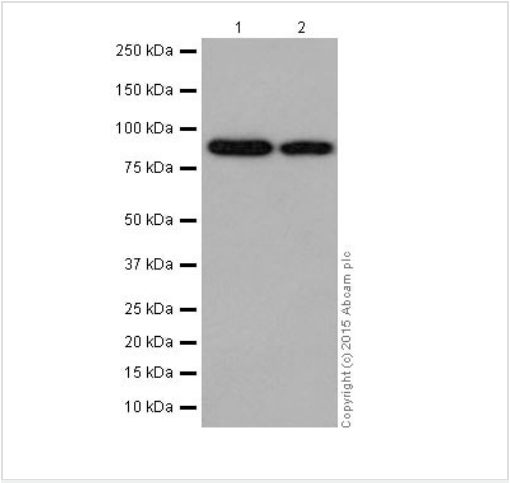
Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labelling LSD2 / AOF1 antibody (red) with purified ab193080 at a dilution of 1/70. Goat anti rabbit IgG (Alexa Fluor® 488) was used as the secondary antibody at 1/2000. Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. Isotype control antibody was Rabbit monoclonal IgG (black). The blue line shows cells without incubation with primary antibody and secondary antibody.



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.



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

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

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

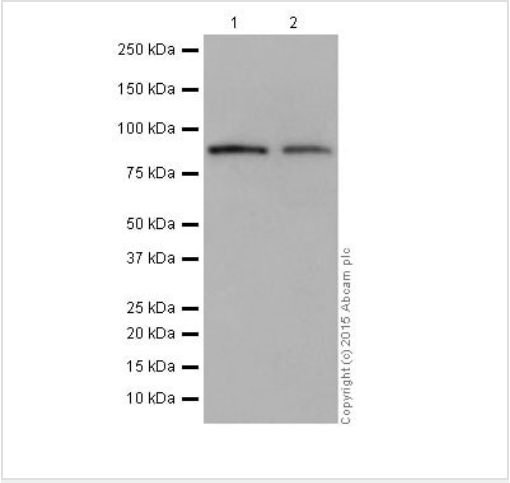
Lysates/proteins at 10 µg per lane.

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

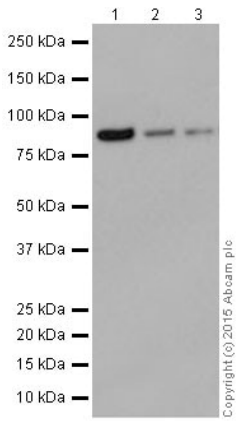
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**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
Lane 3 : PC-12 (Rat adrenal gland pheochromocytoma) cell lysate

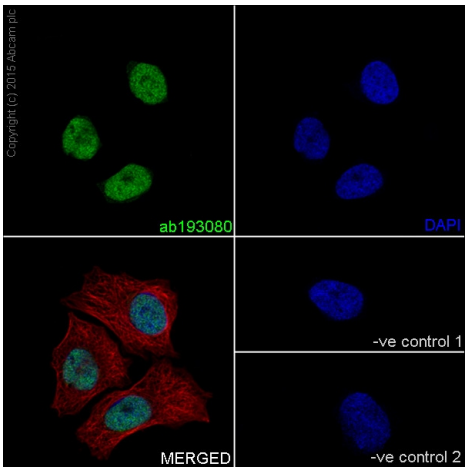
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG 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)

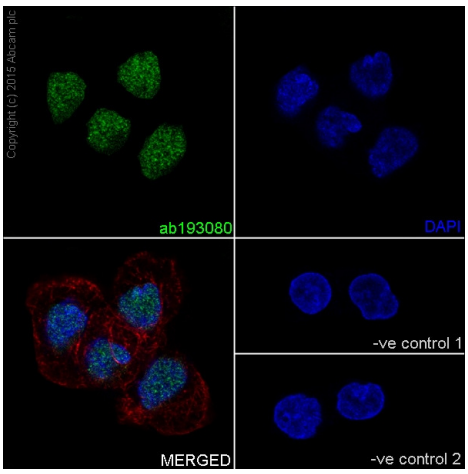
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling LSD2 / AOF1 with ab193080 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000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

Confocal image showing nuclear staining on HeLa cell line.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution and **ab150120** (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 **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 dilution.
-ve control 2: **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/1000 dilution.



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

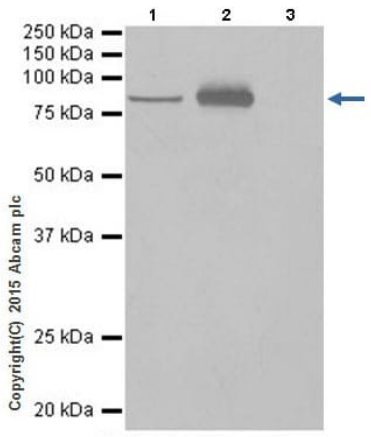
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized A431 (Human epidermoid carcinoma) cells labeling LSD2 / AOF1 with ab193080 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green).

Confocal image showing nuclear staining on A431 cell line.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution and **ab150120** (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 **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 dilution.
-ve control 2: **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/1000 dilution.



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

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 IgG (**ab172730**) instead of ab193080 in K562 whole cell lysate.

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

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

Exposure time: 8 seconds.

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

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