# abcam

### Product datasheet

# Anti-SMYD3 antibody [EPR11107(2)] ab187149





## 13 References 9 Images

#### Overview

**Product name** Anti-SMYD3 antibody [EPR11107(2)]

Rabbit monoclonal [EPR11107(2)] to SMYD3 **Description** 

**Host species** Rabbit

**Tested applications** Suitable for: Flow Cyt (Intra), WB, IHC-P, ICC/IF, IP

Species reactivity Reacts with: Mouse, Rat, Human

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Wild-type HAP1, MCF7, HeLa, HEK-293, T47-D and NIH/3T3 cell lysates; Mouse brain and

Rat spleen tissue lysates. IHC-P: Human colonic carcinoma and mouse skeletal muscle tissues.

ICC/IF: MCF7 and HeLa cells.

**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  $\mathsf{RabMAb}^{\mathsf{®}}$  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: 7.2

Preservative: 0.01% Sodium azide

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

**Purity** Protein A purified

Clonality Monoclonal Clone number EPR11107(2)

**Isotype** IgG

#### **Applications**

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab187149 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)		1/30. <b>ab172730</b> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB		1/1000 - 1/20000. Detects a band of approximately 45 kDa (predicted molecular weight: 49 kDa).
IHC-P		1/100. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
ICC/IF		1/250.
IP		1/40 - 1/60.

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Function Histone methyltransferase. Specifically methylates 'Lys-4' of histone H3, inducing di- and tri-

methylation, but not monomethylation. Plays an important role in transcriptional activation as a member of an RNA polymerase complex. Binds DNA containing 5'-CCCTCC-3' or 5'-GAGGGG-

3' sequences.

**Tissue specificity** Expressed in skeletal muscles and testis. Overexpressed in a majority of colorectal and

hepatocellular carcinomas.

**Sequence similarities**Belongs to the histone-lysine methyltransferase family.

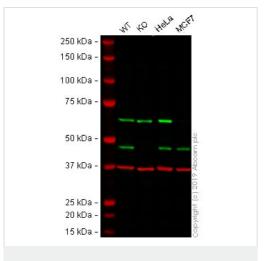
Contains 1 MYND-type zinc finger.

Contains 1 SET domain.

Cytoplasm. Nucleus. Mainly cytoplasmic when cells are arrested at G0/G1. Accumulates in the

nucleus at S phase and G2/M.

#### **Images**



Western blot - Anti-SMYD3 antibody [EPR11107(2)] (ab187149)

**All lanes :** Anti-SMYD3 antibody [EPR11107(2)] (ab187149) at 1/1000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: SMYD3 knockout HAP1 whole cell lysate

Lane 3 : HeLa whole cell lysate

Lane 4 : MCF7 whole cell lysate

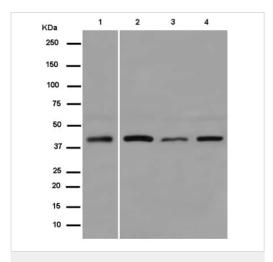
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 49 kDa Observed band size: 49 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab187149 observed at 49 kDa. Red - loading control, <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab187149 was shown to react with SMYD3 in HAP1 wild-type cells in Western blot. Loss of signal was observed when SMYD3 knockout sample was used. HAP1 wild-type and SMYD3 knockout whole cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% Milk in TBS-T (0.1% Tween®) before incubation with ab187149 and <a href="mailto:ab8245">ab8245</a> (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-SMYD3 antibody [EPR11107(2)] (ab187149)

**All lanes :** Anti-SMYD3 antibody [EPR11107(2)] (ab187149) at 1/20000 dilution

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

Lane 2: HeLa (human epithelial cell line from cervix

adenocarcinoma) cell lysate

**Lane 3**: HEK-293 (human epithelial cell line from embryonic kidney) cell lysate

**Lane 4 :** T47-D (human ductal breast epithelial tumor cell line) cell lysate

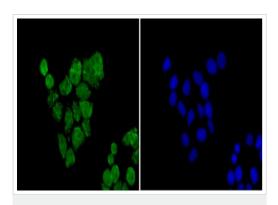
Lysates/proteins at 20 µg per lane.

#### **Secondary**

**All lanes :** Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/1000 dilution

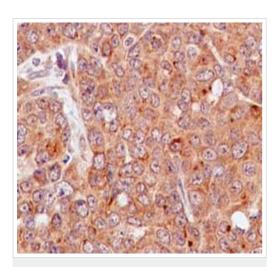
Predicted band size: 49 kDa

Blocking and dilution buffer: 5% NFDM/TBST



Immunocytochemistry/ Immunofluorescence - Anti-SMYD3 antibody [EPR11107(2)] (ab187149)

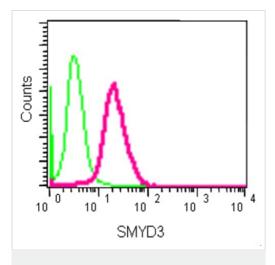
Immunofluorescent analysis of 4% paraformaldehyde-fixed MCF7 (human breast adenocarcinoma cell line) cells, labeling SMYD3 with ab187149 at 1/250 dilution (left image; green); secondary antibody was Goat anti rabbit lgG Alexa Fluor®488, and counterstained with Dapi (right image; blue).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-SMYD3 antibody
[EPR11107(2)] (ab187149)

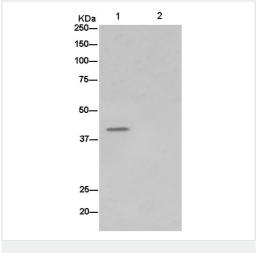
Immunohistochemical analysis of paraffin-embedded human colonic carcinoma tissue, labeling SMYD3 with ab187149 at 1/100 dilution. Counterstained with Hematoxylin.

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



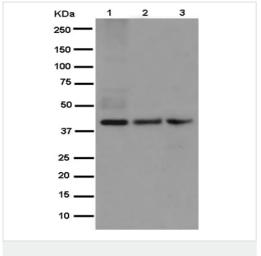
Flow Cytometry (Intracellular) - Anti-SMYD3 antibody [EPR11107(2)] (ab187149)

Intracellular flow cytometric analysis of 2%paraformaldehyde-fixed HeLa (human epithelial cell line from cervix adenocarcinoma) cells, labeling SMYD3 with ab187149 at 1/30 dilution. Secondary antibody wasGoat anti rabbit lgG (FITC) at 1/150 dilution. Isotype control was a Rabbit monoclonal lgG.



Immunoprecipitation analysis of HeLa (human epithelial cell line from cervix adenocarcinoma) cell lysate labeling SMYD3 using ab187149 at 1/50 dilution (Lane 1). A Anti-Rabbit lgG (HRP), specific to the non-reduced form of lgG at 1/1500 was used as secondary antibody. Lane 2: PBS instead of HeLa lysate.





Western blot - Anti-SMYD3 antibody [EPR11107(2)] (ab187149)

**All lanes :** Anti-SMYD3 antibody [EPR11107(2)] (ab187149) at 1/5000 dilution

Lane 1: Mouse brain tissue lysate

Lane 2: Rat spleen tissue lysate

Lane 3: NIH/3T3 cell lysate

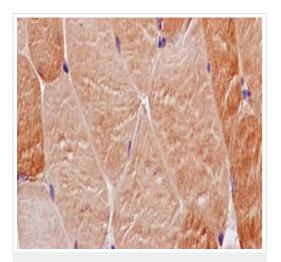
Lysates/proteins at 10 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 49 kDa

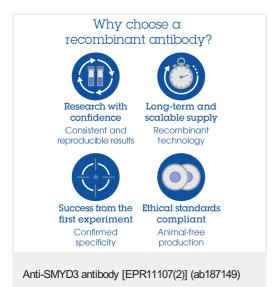
Blocking and dilution buffer: 5% NFDM/TBST



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-SMYD3 antibody
[EPR11107(2)] (ab187149)

Immunohistochemical analysis of paraffin-embedded mouse skeletal muscle tissue, labeling SMYD3 with ab187149 at 1/100 dilution. Counterstained with Hematoxylin.

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



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