# abcam

# Product datasheet

# Anti-Menin antibody [EPR3986] ab92443





7 References 9 Images

Overview

**Product name** Anti-Menin antibody [EPR3986]

**Description** Rabbit monoclonal [EPR3986] to Menin

**Host species** Rabbit

**Tested applications** Suitable for: WB, IP, IHC-P, ICC/IF

Unsuitable for: Flow Cyt

Species reactivity Reacts with: Mouse, Rat, Human

Synthetic peptide within Human Menin aa 600-700 (C terminal). The exact sequence is **Immunogen** 

proprietary.

Positive control WB: Wild-type HAP1, Jurkat cell lysates. IP: Jurkat whole and MEF cell lysates. ICC/IF: Jurkat

**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** 

Storage instructions Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

pH: 7.20 Storage buffer

Preservative: 0.01% Sodium azide

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

**Purity** Protein A purified

Clonality Monoclonal Clone number **EPR3986** 

**Isotype** IgG

#### **Applications**

#### The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab92443 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
WB		1/1000. Predicted molecular weight: 68 kDa. For unpurified use between 1/10,000-1/50,000.
IP		1/10 - 1/100.
IHC-P		1/100 - 1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. For unpurified use between 1/100-1/250.
ICC/IF		1/50. For unpurified use between 1/250-1/500.

#### **Application notes**

Is unsuitable for Flow Cyt.

#### **Target**

#### **Function**

Essential component of a MLL/SET1 histone methyltransferase (HMT) complex, a complex that specifically methylates 'Lys-4' of histone H3 (H3K4). Functions as a transcriptional regulator. Binds to the TERT promoter and represses telomerase expression. Plays a role in TGFB1-mediated inhibition of cell-proliferation, possibly regulating SMAD3 transcriptional activity. Represses JUND-mediated transcriptional activation on AP1 sites, as well as that mediated by NFKB subunit RELA. Positively regulates HOXC8 and HOXC6 gene expression. May be involved in normal hematopoiesis through the activation of HOXA9 expression (By similarity). May be involved in DNA repair.

## Tissue specificity

## Involvement in disease

Ubiquitous.

Defects in MEN1 are the cause of familial multiple endocrine neoplasia type I (MEN1) [MIM:131100]. Autosomal dominant disorder characterized by tumors of the parathyroid glands, gastro-intestinal endocrine tissue, the anterior pituitary and other tissues. Cutaneous lesions and nervous-tissue tumors can exist. Prognosis in MEN1 patients is related to hormonal hypersecretion by tumors, such as hypergastrinemia causing severe peptic ulcer disease (Zollinger-Ellison syndrome, ZES), primary hyperparathyroidism, and acute forms of hyperinsulinemia.

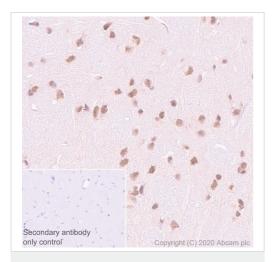
Defects in MEN1 are the cause of familial isolated hyperparathyroidism (FIHP) [MIM:145000]; also known as hyperparathyroidism type 1 (HRPT1). FIHP is an autosomal dominant disorder characterized by hypercalcemia, elevated parathyroid hormone (PTH) levels, and uniglandular or multiglandular parathyroid tumors.

# Post-translational modifications

Phosphorylated upon DNA damage, probably by ATM or ATR.

# **Cellular localization**

Nucleus. Concentrated in nuclear body-like structures. Relocates to the nuclear matrix upon gamma irradiation.



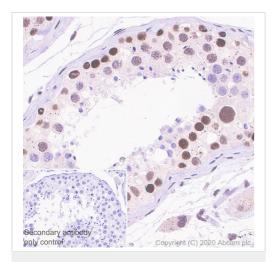
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Menin antibody
[EPR3986] (ab92443)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat cerebrum tissue sections labelling Menin with purified ab92443 at 1/100 dilution (6.08 µg/ml). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using <a href="mailto:ab93684">ab93684</a> (Tris/EDTA buffer, pH 9.0). 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.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Menin antibody
[EPR3986] (ab92443)

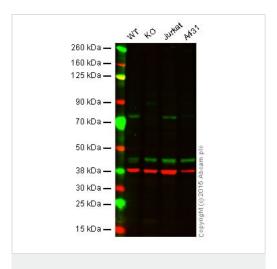
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse liver tissue sections labelling Menin with purified ab92443 at 1/100 dilution (6.08 µg/ml). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using <a href="mailto:ab93684">ab93684</a> (Tris/EDTA buffer, pH 9.0). 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.



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

[EPR3986] (ab92443)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human testis tissue sections labelling Menin with purified ab92443 at 1/500 dilution (1.22 µg/ml). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using <a href="mailto:ab93684">ab93684</a> (Tris/EDTA buffer, pH 9.0). 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.



Western blot - Anti-Menin antibody [EPR3986] (ab92443)

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

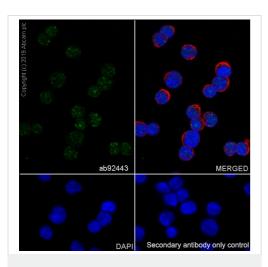
Lane 2: Menin knockout HAP1 cell lysate (20 µg)

Lane 3: Jurkat cell lysate (20 µg)

Lane 4: A431 cell lysate (20 µg)

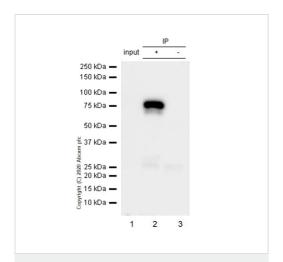
**Lanes 1 - 4**: Merged signal (red and green). Green - ab92443 observed at 74 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab92443 was shown to recognize Menin when Menin knockout samples were used, along with additional cross-reactive bands. Wild-type and Menin knockout samples were subjected to SDS-PAGE. ab92443 and <u>ab8245</u> (loading control to GAPDH) were diluted 1/10,000 and 1/2000 respectively and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (<u>ab216773</u>) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1/10 000 dilution for 1 h at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-Menin antibody [EPR3986] (ab92443)

Immunocytochemistry analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labeling Menin with purified ab92443 at 1/50 dilution (12.1  $\mu$ g/mL). Cells were fixed in 100% Methanol and permeabilized with 0.1% tritonX-100. Cells were counterstained with <u>ab195889</u> Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor<sup>®</sup> 594) 1/200 (2.6  $\mu$ g/mL). Goat anti rabbit lgG (Alexa Fluor<sup>®</sup> 488, <u>ab150078</u>) was used as the secondary antibody at 1/1000 (2  $\mu$ g/mL) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Immunoprecipitation - Anti-Menin antibody [EPR3986] (ab92443)

Purified ab92443 at 1/50 dilution ( $2\mu g$ ) immunoprecipitating Menin in Jurkat whole cell lysate.

Lane 1 (input): Jurkat (Human T cell leukemia T lymphocyte) whole cell lysate  $10\mu g$ 

Lane 2 (+): ab92443 + Jurkat whole cell lysate.

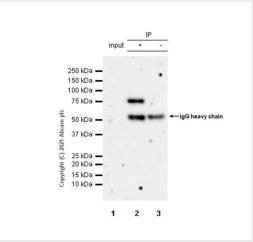
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab92443 in Jurkat whole cell lysate.

VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>) (1/1000 dilution) was used for Western blotting.

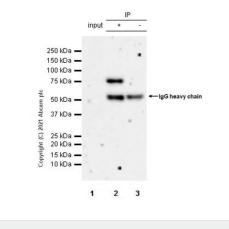
Blocking Buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM/TBST.

Observed band size: 75 kDa



Immunoprecipitation - Anti-Menin antibody [EPR3986] (ab92443)



All lanes: Anti-Menin antibody [EPR3986] (ab92443) at 1/1000

Lane 1: M⊞ (mouse embryonic fibroblast (immortalized)) w hole cell lysate 10 µg

**Lane 3**: Rabbit monoclonal  $lgG(\underline{ab172730})$  instead of ab92443 in MHF whole cell lysate



Western blot - Anti-Menin antibody [EPR3986] (ab92443)

dilution (Purified)

Menin was immunoprecipitated from 0.35 mg M⊞ (mouse embryonic fibroblast (immortalized)) whole cell lysate 10 ug with ab92443 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab92443 at 1/1000 dilution. VeriBlot for IP Detection

Reagent (HRP)(ab131366) was used at 1/5000 dilution.

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

Lane 2: ab92443 IP in M⊞ whole cell lysate

Exposure time: 3 minutes

Lane 1: Jurkat (Human T cell leukemia T lymphocyte) whole cell lysate

Lane 2: MEF (Mouse embryonic fibroblast (immortalized)) whole cell lysate

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

Lysates/proteins at 15 µg per lane.

## Secondary

All lanes: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 68 kDa Observed band size: 75 kDa



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