abcam

Product datasheet

Anti-BRAF antibody [EP152Y] ab33899

敲除验证 重组 RabMAb

12 Abreviews 30 References 11 图像

概述

产品名称	Anti-BRAF抗体[EP152Y]
描述	兔单克隆抗体[EP152Y] to BRAF
宿主	Rabbit
特异性	The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat.
经测试应用	**适用于:** WB, IHC-P, IP **不适用于:** ICC
种属反应性	**与反应:** Mouse, Rat, Human
免疫原	Synthetic peptide within Human BRAF aa 50-150. The exact sequence is proprietary. Database link: **P15056**
阳性对照	WB: HAP1, PC-12 and HeLa whole cell lysate (**ab150035**); Mouse brain tissue lysate. IHC-P: Human prostate and breast cancer tissue; Rat brain tissue. IP: HeLa cells.
常规说明	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**.

性能

形式	Liquid
存放说明	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.
存储溶液	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

纯度	Protein A purified
克隆	单克隆
克隆编号	EP152Y
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab33899于以下的经测试应用**

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★★★ (4)	1/1000 - 1/5000. Detects a band of approximately 87 kDa (predicted molecular weight: 85 kDa).
IHC-P	★★★☆☆ (4)	1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**. The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat.
IP		1/20.

应用说明 Is unsuitable for ICC.

靶标

功能 Involved in the transduction of mitogenic signals from the cell membrane to the nucleus. May play a role in the postsynaptic responses of hippocampal neuron.

组织特异性 Brain and testis.

疾病相关 Note=Defects in BRAF are found in a wide range of cancers.
Defects in BRAF may be a cause of colorectal cancer (CRC) [MIM:114500].
Defects in BRAF are involved in lung cancer (LNCR) [MIM:211980].
Defects in BRAF are involved in non-Hodgkin lymphoma (NHL) [MIM:605027]. NHL is a cancer that starts in cells of the lymph system, which is part of the body's immune system. NHLs can occur at any age and are often marked by enlarged lymph nodes, fever and weight loss.
Defects in BRAF are a cause of cardiofaciocutaneous syndrome (CFC syndrome) [MIM:115150]; also known as cardio-facio-cutaneous syndrome. CFC syndrome is characterized by a distinctive facial appearance, heart defects and mental retardation. Heart defects include pulmonic stenosis, atrial septal defects and hypertrophic cardiomyopathy. Some affected individuals present with ectodermal abnormalities such as sparse, friable hair, hyperkeratotic skin lesions and a generalized ichthyosis-like condition. Typical facial features are similar to Noonan syndrome. They include high forehead with bitemporal constriction, hypoplastic supraorbital ridges, downslanting palpebral fissures, a depressed nasal bridge, and posteriorly angulated ears with prominent helices. The inheritance of CFC syndrome is autosomal dominant.
Defects in BRAF are the cause of Noonan syndrome type 7 (NS7) [MIM:613706]. Noonan syndrome is a disorder characterized by facial dysmorphic features such as hypertelorism, a downward eyeslant and low-set posteriorly rotated ears. Other features can include short stature,

a short neck with webbing or redundancy of skin, cardiac anomalies, deafness, motor delay and variable intellectual deficits.
Defects in BRAF are the cause of LEOPARD syndrome type 3 (LEOPARD3) [MIM:613707]. LEOPARD3 is a disorder characterized by lentigines, electrocardiographic conduction abnormalities, ocular hypertelorism, pulmonic stenosis, abnormalities of genitalia, retardation of growth, and sensorineural deafness.
Note=A chromosomal aberration involving BRAF is found in pilocytic astrocytomas. A tandem duplication of 2 Mb at 7q34 leads to the expression of a KIAA1549-BRAF fusion protein with a constitutive kinase activity and inducing cell transformation.

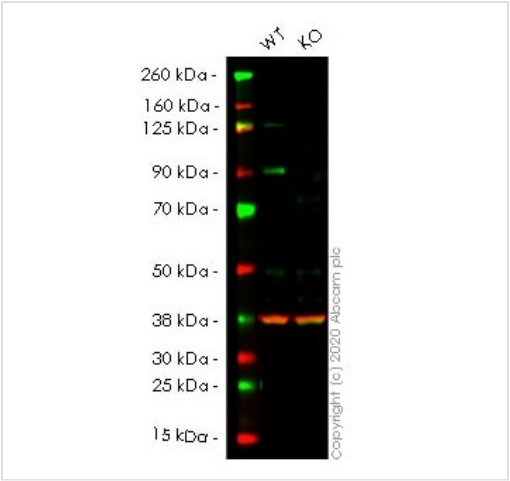
序列相似性

Belongs to the protein kinase superfamily. TKL Ser/Thr protein kinase family. RAF subfamily.
Contains 1 phorbol-ester/DAG-type zinc finger.
Contains 1 protein kinase domain.
Contains 1 RBD (Ras-binding) domain.

细胞定位

Nucleus. Cytoplasm. Cell membrane. Colocalizes with RGS14 and RAF1 in both the cytoplasm and membranes.

图片



Western blot - Anti-BRAF antibody [EP152Y] (ab33899)

All lanes : Anti-BRAF antibody [EP152Y] (ab33899) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : BRAF knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

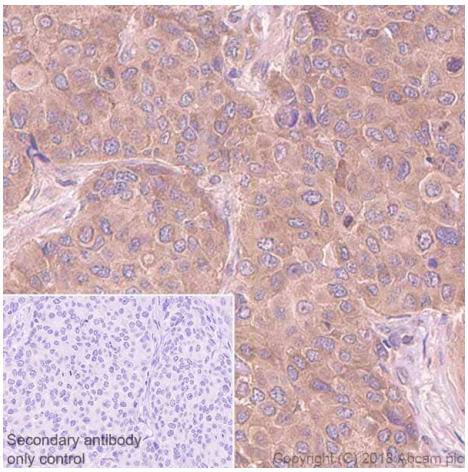
Performed under reducing conditions.

Predicted band size: 85 kDa
Observed band size: 90 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab33899 observed at 90 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

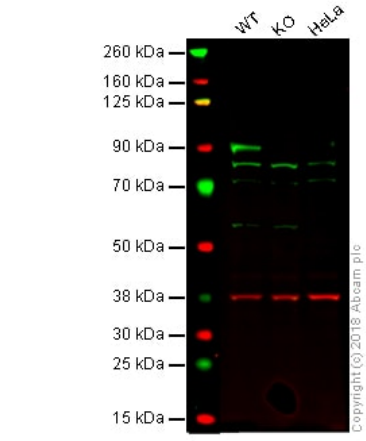
ab33899 was shown to react with BRAF in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab265373** (knockout cell lysate **ab257078**) was used. Wild-type HeLa and BRAF knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab33899 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) 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) 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.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRAF antibody [EP152Y] (ab33899)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human breast cancer tissue sections labeling BRAF with purified ab33899 at 1/100 dilution (4.77 µg/ml). Heat mediated antigen retrieval was performed using heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Western blot - Anti-BRAF antibody [EP152Y] (ab33899)

All lanes : Anti-BRAF antibody [EP152Y] (ab33899) at 1/1000 dilution

Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : BRAF knockout HAP1 whole cell lysate
Lane 3 : HeLa whole cell lysate

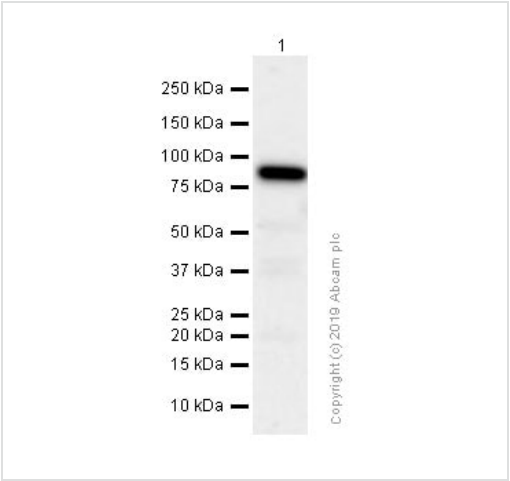
Lysates/proteins at 40 µg per lane.

Predicted band size: 85 kDa

Lanes 1 - 3: Merged signal (red and green). Green - ab33899 (unpurified) observed at 90 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab33899 was shown to recognize B Raf in wild-type HAP1 cells as signal was lost at the expected MW in B Raf knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and B Raf knockout samples were subjected to SDS-PAGE. ab33899 and **ab8245** (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 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 1/20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-BRAF antibody [EP152Y] (ab33899)

Anti-BRAF antibody [EP152Y] (ab33899) at 1/5000 dilution (Purified) + PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysates at 15 µg

Secondary
Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 85 kDa
Observed band size: 87 kDa

Western blot - Anti-BRAF antibody [EP152Y] (ab33899)

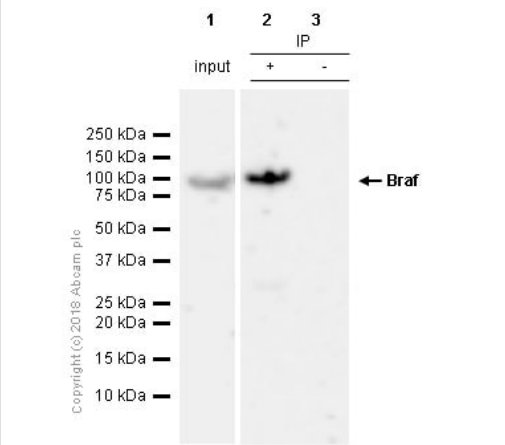
All lanes : Anti-BRAF antibody [EP152Y] (ab33899) at 1/2000 dilution (Purified)

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates
Lane 2 : Mouse brain lysates

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: 85 kDa
Observed band size: 87 kDa



Immunoprecipitation - Anti-BRAF antibody [EP152Y] (ab33899)

ab33899 (Purified) at 1/20 dilution (20 µg/ml) immunoprecipitating BRAF in HeLa whole cell lysate.

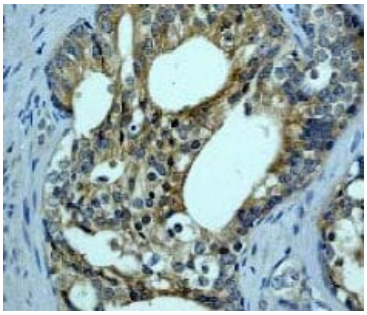
Lane 1 (input): HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate 10 µg

Lane 2 (+): ab33899 & HeLa whole cell lysate

Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of ab33899 in HeLa whole cell lysate

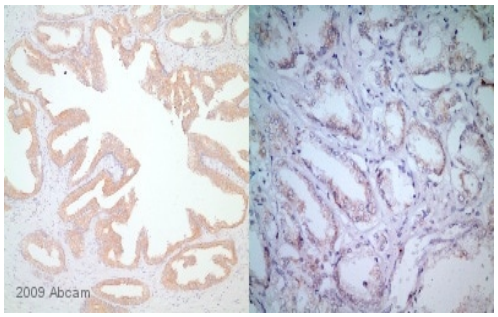
For western blotting, ab33899 at 1/500 and VeriBlot for IP secondary antibody (HRP) (**ab131366**) was used as the secondary antibody at 1/1000 dilution.

Blocking and diluting buffer: 5% NFDM /TBST .



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRAF antibody [EP152Y] (ab33899)

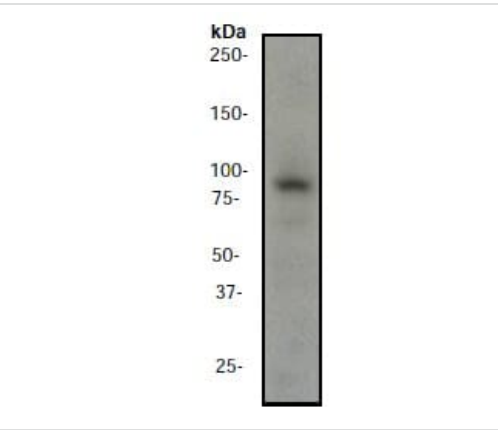
This image shows paraffin embedded human prostate cancer tissue sample stained with ab33899 (unpurified) at 1/250 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRAF antibody [EP152Y] (ab33899)

This image is courtesy of an Abreview submitted by Sedar Balci

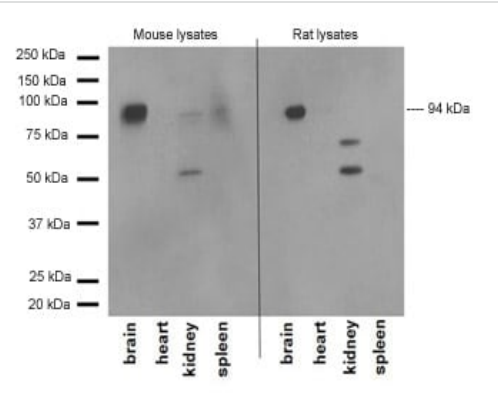
ab33899 (unpurified) staining B Raf cells from human prostate tissue by immunohistochemistry (formalin/PFA-fixed paraffin-embedded sections). Cells were formaldehyde fixed and permeabilized in PBS-Tween 20 prior to blocking in 70% serum for 10 minutes at 25°C. The primary antibody was diluted 1/250 and incubated with the sample for 1 hour at 25°C. A biotin conjugated goat polyclonal to mouse Ig was used as the secondary.



Western blot - Anti-BRAF antibody [EP152Y] (ab33899)

Anti-BRAF antibody [EP152Y] (ab33899) at 1/5000 dilution (unpurified) + HeLa cell lysate

Predicted band size: 85 kDa
Observed band size: 87 kDa



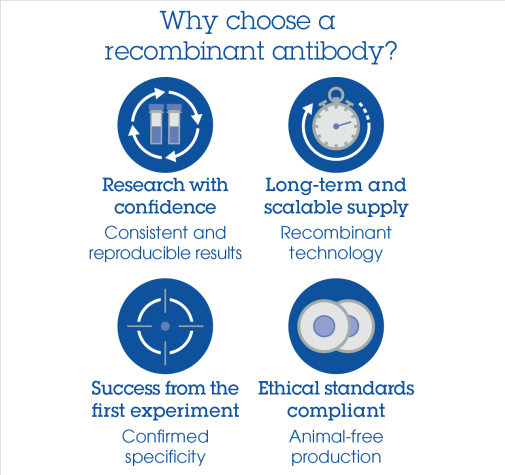
Western blot - Anti-BRAF antibody [EP152Y] (ab33899)

All lanes : Anti-BRAF antibody [EP152Y] (ab33899) at 1/1000 dilution (unpurified)

Lane 1 : Lysate prepared from mouse brain
Lane 2 : Lysate prepared from mouse heart
Lane 3 : Lysate prepared from mouse kidney
Lane 4 : Lysate prepared from mouse spleen
Lane 5 : Lysate prepared from rat brain
Lane 6 : Lysate prepared from rat heart
Lane 7 : Lysate prepared from rat kidney
Lane 8 : Lysate prepared from rat spleen

Predicted band size: 85 kDa

Exposure time: 3 minutes



Anti-BRAF antibody [EP152Y] (ab33899)

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