# abcam

# Product datasheet

# Anti-Ras antibody [EP1125Y] ab52939

Recombinant RabMAb

★★★★★ 12 Abreviews 68 References 9 Images

#### Overview

**Product name** Anti-Ras antibody [EP1125Y]

**Description** Rabbit monoclonal [EP1125Y] to Ras

**Host species** Rabbit

Specificity This antibody is predicted to react with H-Ras, N-Ras and K-Ras.

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

Species reactivity Reacts with: Mouse, Rat, Human

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

Positive control Flow Cyt (intra): PC-12 cells. WB: Jurkat, 293T, RAW 246.7, Neuro-2a, PC-12 and C6 lysates;

Human Ras full length protein. ICC/IF: MCF7 cells. IP: Jurkat 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<sup>®</sup> 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: 59% PBS, 40% Glycerol, 0.05% BSA

**Purity** Protein A purified

Clonality Monoclonal Clone number EP1125Y

**Isotype** IgG

#### **Applications**

#### The Abpromise guarantee

Our Abpromise guarantee covers the use of ab52939 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.  ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody. For unpurified use at 1/100.
ICC/IF		1/500.
WB	<b>★★★★★</b> (9)	1/5000. Detects a band of approximately 21 kDa (predicted molecular weight: 21 kDa).  For unpurified use at 1/10000-1/50000.
IP	*** <u>*</u>	1/20. For unpurified use at 1/30.

#### **Target**

#### **Function**

#### Involvement in disease

Ras proteins bind GDP/GTP and possess intrinsic GTPase activity.

Defects in HRAS are the cause of faciocutaneoskeletal syndrome (FCSS) [MIM:218040]. A rare condition characterized by prenatally increased growth, postnatal growth deficiency, mental retardation, distinctive facial appearance, cardiovascular abnormalities (typically pulmonic stenosis, hypertrophic cardiomyopathy and/or atrial tachycardia), tumor predisposition, skin and musculoskeletal abnormalities.

Defects in HRAS are the cause of congenital myopathy with excess of muscle spindles (CMEMS) [MIM:218040]. CMEMS is a variant of Costello syndrome.

Defects in HRAS may be a cause of susceptibility to Hurthle cell thyroid carcinoma (HCTC) [MIM:607464]. Hurthle cell thyroid carcinoma accounts for approximately 3% of all thyroid cancers. Although they are classified as variants of follicular neoplasms, they are more often multifocal and somewhat more aggressive and are less likely to take up iodine than are other follicular neoplasms.

Note=Mutations which change positions 12, 13 or 61 activate the potential of HRAS to transform cultured cells and are implicated in a variety of human tumors.

Defects in HRAS are a cause of susceptibility to bladder cancer (BLC) [MIM:109800]. A malignancy originating in tissues of the urinary bladder. It often presents with multiple tumors appearing at different times and at different sites in the bladder. Most bladder cancers are transitional cell carcinomas. They begin in cells that normally make up the inner lining of the bladder. Other types of bladder cancer include squamous cell carcinoma (cancer that begins in thin, flat cells) and adenocarcinoma (cancer that begins in cells that make and release mucus and other fluids). Bladder cancer is a complex disorder with both genetic and environmental influences.

Note=Defects in HRAS are the cause of oral squamous cell carcinoma (OSCC).

### Sequence similarities

Belongs to the small GTPase superfamily. Ras family.

# Post-translational modifications

Palmitoylated by the ZDHHC9-GOLGA7 complex. A continuous cycle of de- and re-palmitoylation

regulates rapid exchange between plasma membrane and Golgi.

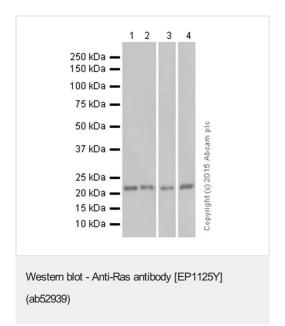
 $S-nitrosylated; critical for redox \, regulation. \, Important \, for \, stimulating \, guanine \, nucleotide \, exchange.$ 

No structural perturbation on nitrosylation.

#### **Cellular localization**

Cell membrane. Golgi apparatus membrane. The active GTP-bound form is localized most strongly to membranes than the inactive GDP-bound form (By similarity). Shuttles between the plasma membrane and the Golgi apparatus.

#### **Images**



**All lanes :** Anti-Ras antibody [EP1125Y] (ab52939) at 1/5000 dilution (purified)

**Lane 1 :** Jurkat (human T cell leukemia cell line from peripheral blood) whole cell lysates

**Lane 2 :** HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysates

Lane 3: RAW 264.7 (mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysates

**Lane 4 :** Neuro-2a (mouse neuroblastoma cell line) whole cell lysates

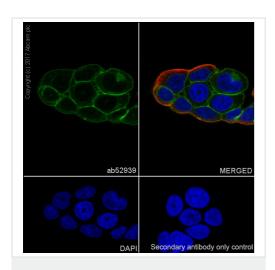
Lysates/proteins at 20 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

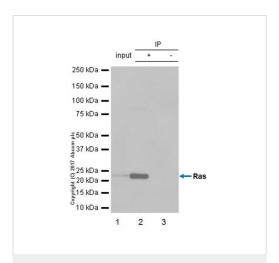
**Predicted band size:** 21 kDa **Observed band size:** 21 kDa

Blocking and diluting buffer: 5% NFDM/TBST



Immunocytochemistry/ Immunofluorescence - Anti-Ras antibody [EP1125Y] (ab52939)

Immunocytochemistry analysis of MCF7 (human breast adenocarcinoma cell line) cells labeling Ras with Purified ab52939 at 1:500 dilution. Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor ® 594) 1:200 (2.5 µg/ml). ab150077 Goat anti rabbit lgG(Alexa Fluor ® 488) was used as the secondary antibody at 1:1000 dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Immunoprecipitation - Anti-Ras antibody [EP1125Y] (ab52939)

ab52939 (purified) at 1:20 dilution (2 $\mu$ g) immunoprecipitating Ras in Jurkat whole cell lysate.

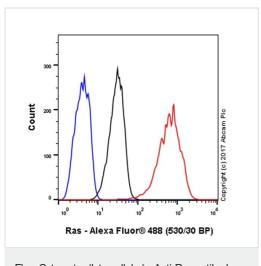
Lane 1 (input): Jurkat (human T cell leukemia cell line from peripheral blood) whole cell lysate 10µg

Lane 2 (+): ab52939 & Jurkat whole cell lysate

**Lane 3 (-):** Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab52939 in Jurkat whole cell lysate

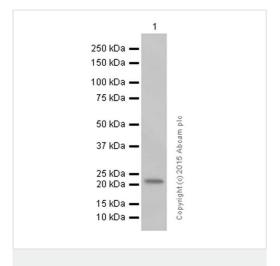
For western blotting, VeriBlot for IP Detection Reagent (HRP) (ab131366) was used for detection at 1:1000 dilution.

Blocking and diluting buffer: 5% NFDM/TBST.



Intracellular Flow Cytometry analysis of PC-12 (rat adrenal gland pheochromocytoma cell line) cells labeling Ras with purified ab52939 at 1/30 dilution (red). Cells were fixed with 4% Paraformaldehyde. A Goat anti rabbit lgG (Alexa Fluor<sup>®</sup> 488) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal lgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).





Western blot - Anti-Ras antibody [EP1125Y] (ab52939)

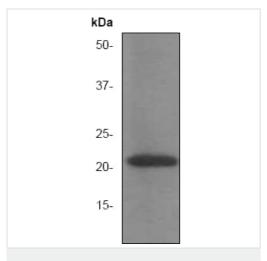
Anti-Ras antibody [EP1125Y] (ab52939) at 1/5000 dilution (purified) + PC-12 (rat adrenal gland pheochromocytoma cell line) whole cell lysates at 15  $\mu g$ 

# **Secondary**

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

Predicted band size: 21 kDa

Blocking and diluting buffer: 5% NFDM/TBST.



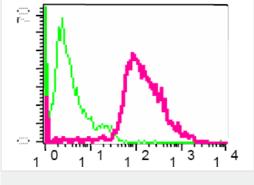
Western blot - Anti-Ras antibody [EP1125Y] (ab52939)

Anti-Ras antibody [EP1125Y] (ab52939) at 1/500000 dilution (unpurified) + C6 (rat glial tumor cell line) cell lysate at 10 µg/ml

# **Secondary**

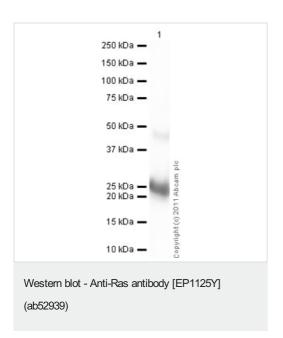
goat anti-rabbit HRP at 1/2000 dilution

**Predicted band size:** 21 kDa **Observed band size:** 18 kDa



Flow Cytometry (Intracellular) - Anti-Ras antibody [EP1125Y] (ab52939)

Unpurified ab52939 at 1/100 dilution staining Ras in permeabilized PC-12 (rat adrenal gland pheochromocytoma cell line) cells by intracellular flow cytometry (red). Rabbit IgG negative control (green).



Anti-Ras antibody [EP1125Y] (ab52939) at 1/500 dilution (unpurified) + Human Ras full length protein (ab56526) at 0.01 µg

#### Secondary

Goat Anti-Rabbit  $\lg G$  H&L (HRP) preadsorbed ( $\frac{ab97080}{1}$ ) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 21 kDa

Exposure time: 1 minute



Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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