abcam

Product datasheet

Anti-Ras antibody [EPR3255] ab108602



★★★★★ 9 Abreviews 24 References 10 Images

Overview

Product name Anti-Ras antibody [EPR3255]

Description Rabbit monoclonal [EPR3255] to Ras

Host species Rabbit

Specificity ab108602 also detects H-Ras and N-Ras.

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

Species reactivity Reacts with: Mouse, Rat, Human

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

Positive control WB: 293T and SH-SY5Y cell lysates; Rat brain lysates; Mouse heart lysates IP: SH-SY5Y cell

lysates Flow Cyt (intra): HEK-293 cells ICC/IF: HEK-293T cells IHC-P: Human breast carcinoma,

mouse cerebrum, rat cerebrum and rat cardiac muscle tissues.

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. 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 EPR3255

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise quarantee covers the use of ab108602 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
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
Flow Cyt (Intra)		1/30.
WB	★★★★★ (6)	1/1000 - 1/10000. Predicted molecular weight: 22 kDa.
IP	★★★★★ (2)	1/10 - 1/100.
ICC/IF		1/50 - 1/100.

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



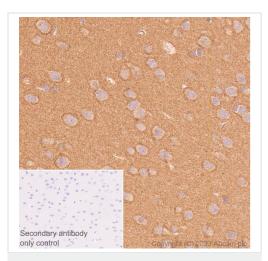
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Ras antibody [EPR3255] (ab108602)

Immunohistochemical analysis of paraffin-embedded rat cardiac muscle tissue labelling Ras with <u>ab209974</u> at 1/10000 (0.104 µg/ml) dilution followed by a LeicaDS9800 (Bond™ Polymer Refine Detection) at ready to use dilution. Positive staining on rat cardiac muscle. The section was incubated with <u>ab209974</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is LeicaDS9800 (Bond™ Polymer Refine Detection) at ready to use dilution.

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.

This data was developed using the same antibody clone in a different buffer formulation (<u>ab209974</u>).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Ras antibody [EPR3255] (ab108602)

Immunohistochemical analysis of paraffin-embedded rat cerebrum tissue labelling Ras with <u>ab209974</u> at 1/10000 (0.104 µg/ml) dilution followed by a LeicaDS9800 (Bond™ Polymer Refine Detection) at ready to use dilution. Positive staining on rat cerebrum. The section was incubated with <u>ab209974</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with hematoxylin.

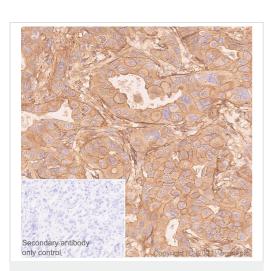
Secondary antibody only control: Secondary antibody is LeicaDS9800 (Bond™ Polymer Refine Detection) at Ready to use dilution.

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.

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Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Ras antibody [EPR3255] (ab108602)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Ras antibody [EPR3255] (ab108602)

Immunohistochemical analysis of paraffin-embedded mouse cerebrum tissue labelling Ras with <u>ab209974</u> at 1/10000 (0.104 µg/ml) dilution followed by a LeicaDS9800 (Bond™ Polymer Refine Detection) at ready to use dilution. Positive staining on mouse cerebrum. The section was incubated with <u>ab209974</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is LeicaDS9800 (Bond™ Polymer Refine Detection) at ready to use dilution.

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.

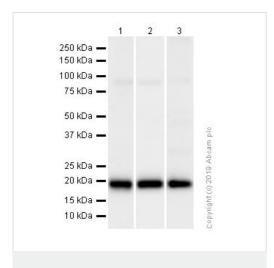
This data was developed using the same antibody clone in a different buffer formulation (ab209974).

Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue labelling Ras with <u>ab209974</u> at 1/10000 (0.104 µg/ml) dilution followed by a LeicaDS9800 (Bond™ Polymer Refine Detection) at ready to use dilution. Positive staining on human breast carcinoma. The section was incubated with <u>ab209974</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is LeicaDS9800 (Bond™ Polymer Refine Detection) at ready to use dilution.

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.

This data was developed using the same antibody clone in a different buffer formulation ($\underline{ab209974}$).



Western blot - Anti-Ras antibody [EPR3255] (ab108602)



Lane 1 : HEK-293 (Human embryonic kidney epithelial cell) whole cell lysates

Lane 2: Rat brain lysates

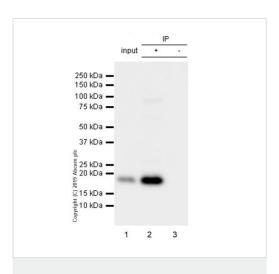
Lane 3: Mouse heart lysates

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: 22 kDa **Observed band size:** 21 kDa



Immunoprecipitation - Anti-Ras antibody [EPR3255] (ab108602)

ab108602 (purified) at 1/20 dilution (2 $\mu g)$ immunoprecipitating Ras in SH-SY5Y whole cell lysate.

Lane 1 (input): SH-SY5Y (Human neuroblastoma epithelial cell) whole cell lysate 10 μg

Lane 2 (+): ab108602 & SH-SY5Y whole cell lysate

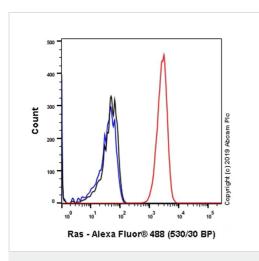
Lane 3 (-): Rabbit monoclonal $\lg G (\underline{ab172730})$ instead of

ab108602 in SH-SY5Y whole cell lysate

For western blotting, VeriBlot for IP Detection Reagent (HRP)

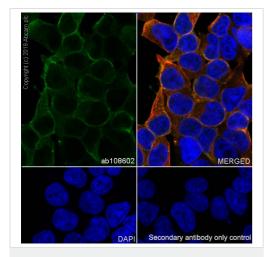
(ab131366) was used at 1/1000 dilution.

Blocking and diluting buffer: 5% NFDM/TBST.



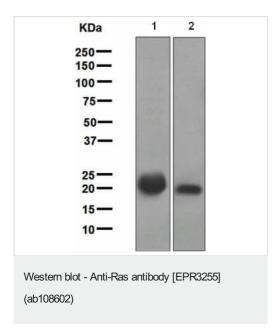
Flow Cytometry (Intracellular) - Anti-Ras antibody [EPR3255] (ab108602)

Intracellular Flow Cytometry analysis of HEK-293 (Human embryonic kidney epithelial cell) cells labeling Ras with purified ab108602 at 1/30 dilution (10 µg/ml) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor[®] 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal lgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunocytochemistry/ Immunofluorescence - Anti-Ras antibody [EPR3255] (ab108602)

Immunocytochemistry/ Immunofluorescence analysis of 293T (Human embryonic kidney epithelial cell) cells labeling Ras with purified ab108602 at 1:100 dilution (10 μ g/ml). 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 μ g/ml). Goat anti rabbit lgG (Alexa Fluor® 488, <u>ab150077</u>) was used as the secondary antibody at 1:1000 (2 μ g/ml) dilution. DAPI (blue) was used as the secondary antibody only control.



All lanes : Anti-Ras antibody [EPR3255] (ab108602) at 1/1000 dilution (unpurified)

Lane 1: HEK-293 whole cell lysate

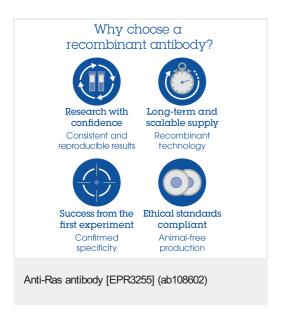
Lane 2: SH-SY5Y cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: HRP labelled goat anti-rabbit at 1/2000 dilution

Predicted band size: 22 kDa



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

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