

Anti-Raf1 Rabbit Monoclonal Antibody

Catalog # ABO13334

Specification

Anti-Raf1 Rabbit Monoclonal Antibody - Product Information

Application Primary Accession Host Isotype Reactivity Clonality Format Description WB, IHC, IF, ICC <u>P04049</u> Rabbit Rabbit IgG Rat, Human, Mouse Monoclonal Liquid

Anti-Raf1 Rabbit Monoclonal Antibody . Tested in WB, IHC, ICC/IF applications. This antibody reacts with Human, Mouse, Rat.

Anti-Raf1 Rabbit Monoclonal Antibody - Additional Information

Gene ID 5894

Other Names RAF proto-oncogene serine/threonine-protein kinase, 2.7.11.1, Proto-oncogene c-RAF, cRaf, Raf-1, RAF1 (HGNC:9829), RAF

Calculated MW 73052 MW KDa

Application Details WB 1:1000-1:2000
IHC 1:50-1:200
ICC/IF 1:50-1:200

Subcellular Localization

Cytoplasm. Cell membrane. Mitochondrion. Nucleus. Colocalizes with RGS14 and BRAF in both the cytoplasm and membranes. Phosphorylation at Ser-259 impairs its membrane accumulation. Recruited to the cell membrane by the active Ras protein. Phosphorylation at Ser-338 and Ser-339 by PAK1 is required for its mitochondrial localization. Retinoic acid- induced Ser-621 phosphorylated form of RAF1 is predominantly localized at the nucleus.

Tissue Specificity

In skeletal muscle, isoform 1 is more abundant than isoform 2..

Contents

Rabbit IgG in phosphate buffered saline, pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol, 0.4-0.5mg/ml BSA.

Immunogen A synthesized peptide derived from human Raf1

Purification



Affinity-chromatography

Storage

Store at -20°C for one year. For short term storage and frequent use, store at 4°C for up to one month. Avoid repeated freeze-thaw cycles.

Anti-Raf1 Rabbit Monoclonal Antibody - Protein Information

Name RAF1 (HGNC:9829)

Synonyms RAF

Function

Serine/threonine-protein kinase that acts as a regulatory link between the membrane-associated Ras GTPases and the MAPK/ERK cascade, and this critical regulatory link functions as a switch determining cell fate decisions including proliferation, differentiation, apoptosis, survival and oncogenic transformation. RAF1 activation initiates a mitogen-activated protein kinase (MAPK) cascade that comprises a sequential phosphorylation of the dual-specific MAPK kinases (MAP2K1/MEK1 and MAP2K2/MEK2) and the extracellular signal- regulated kinases (MAPK3/ERK1 and MAPK1/ERK2). The phosphorylated form of RAF1 (on residues Ser-338 and Ser-339, by PAK1) phosphorylates BAD/Bcl2-antagonist of cell death at 'Ser-75'. Phosphorylates adenylyl cyclases: ADCY2, ADCY5 and ADCY6, resulting in their activation. Phosphorylates PPP1R12A resulting in inhibition of the phosphatase activity. Phosphorylates TNNT2/cardiac muscle troponin T. Can promote NF-kB activation and inhibit signal transducers involved in motility (ROCK2), apoptosis (MAP3K5/ASK1 and STK3/MST2), proliferation and angiogenesis (RB1). Can protect cells from apoptosis also by translocating to the mitochondria where it binds BCL2 and displaces BAD/Bcl2-antagonist of cell death. Regulates Rho signaling and migration, and is required for normal wound healing. Plays a role in the oncogenic transformation of epithelial cells via repression of the TJ protein, occludin (OCLN) by inducing the up-regulation of a transcriptional repressor SNAI2/SLUG, which induces down-regulation of OCLN. Restricts caspase activation in response to selected stimuli, notably Fas stimulation, pathogen-mediated macrophage apoptosis, and erythroid differentiation.

Cellular Location

Cytoplasm. Cell membrane. Mitochondrion. Nucleus. Note=Colocalizes with RGS14 and BRAF in both the cytoplasm and membranes. Phosphorylation at Ser-259 impairs its membrane accumulation. Recruited to the cell membrane by the active Ras protein Phosphorylation at Ser-338 and Ser-339 by PAK1 is required for its mitochondrial localization. Retinoic acid-induced Ser-621 phosphorylated form of RAF1 is predominantly localized at the nucleus

Tissue Location

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Anti-Raf1 Rabbit Monoclonal Antibody - Protocols

Provided below are standard protocols that you may find useful for product applications.

- <u>Western Blot</u>
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation



Flow Cytomety

<u>Cell Culture</u>

Anti-Raf1 Rabbit Monoclonal Antibody - Images

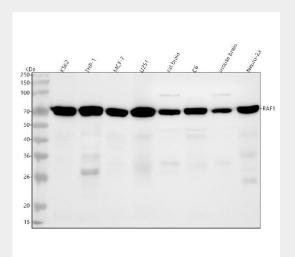


Figure 1. Western blot analysis of Raf1 using anti-Raf1 antibody (M00446-1).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human K562 whole cell lysates,

Lane 2: human THP-1 whole cell lysates,

Lane 3: human MCF-7 whole cell lysates,

Lane 4: human U251 whole cell lysates,

Lane 5: rat brain tissue lysates,

Lane 6: rat C6 whole cell lysates,

Lane 7: mouse brain tissue lysates,

Lane 8: mouse Neuro-2a whole cell lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-Raf1 antigen affinity purified monoclonal antibody (Catalog # M00446-1) at 1:1000 overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:500 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for Raf1 at approximately 73 kDa.