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[Marker Primary Antibody]

Rabbit Anti-phospho-c-Raf (Thr269)/HRP Conjugated antibody

Catalog Number: SL5651R-HRP

FOR RESEARCH USE ONLY

[DATASHEET]

Host:	Rabbit
Isotype:	IgG
Clonality:	Polyclonal
Target:	Anti-phospho-c-Raf(Thr269)/HRP
Immunogen:	KLH conjugated Synthesised phosphopeptide derived from human c-Raf around the phosphorylation site of Thr269
Purification:	affinity purified by Protein A
Concentration:	1mg/ml
Storage:	Store at -20 °C for one year. Avoid repeated freeze/thaw cycles. The lyophilized antibody is stable at room temperature for at least one month and for greater than a year when kept at -20°C. When reconstituted in sterile pH 7.4 1M PBS or diluent of antibody the antibody is stable for at least two weeks at 2-4 °C.
Applications:	IHC-P=1:100-500 IHC-F=1:100-500 not yet tested in other applications. optimal dilutions/concentrations should be determined by the end user.
Reactivity:	Mouse(predicted:Human,Rat,Dog,Pig,Cow,Horse,Rabbit,Sheep)
Background:	background: The Raf family of serine/threonine specific kinases is comprised of three members (aRaf, bRaf, and cRaf) that play a critical role in regulating cell growth and differentiation, and couple growth factor receptor stimulation to nuclear transcription factors via the Ras/mitogen activated protein kinase (MAPK) pathway. cRaf kinase (also known as Raf1) is a small GTPase like kinase of 73 kDa, and is a signal transducer of multiple extracellular stimuli that is regulated by several pathways, and that once activated, phosphorylates MEK which in turn phosphorylates ERK. Raf1 is involved in the transduction of mitogenic signals from the cell membrane to the nucleus. It is part of the Ras dependent signaling pathway from receptors to the nucleus. **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

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	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. [CATALYTIC ACTIVITY] ATP + a protein = ADP + a phosphoprotein. **Subunit:** Monomer. Homodimer. Heterodimerizes with BRAF and this heterodimer possesses a highly increased kinase activity compared to the respective homodimers or monomers. Heterodimerization is mitogen-regulated and enhanced by 14-3-3 proteins. MAPK1/ERK2 activation can induce a negative feedback that promotes the dissociation of the heterodimer. Forms a multiprotein complex with Ras (M-Ras/MRAS), SHOC2 and protein phosphatase 1 (PPP1CA, PPP1CB and PPP1CC). Interacts with Ras proteins; the interaction is antagonized by RIN1. Weakly interacts with RIT1. Interacts (via N-terminus) with RGS14 (via RBD domains); the interaction mediates the formation of a ternary complex with BRAF, a ternary complex inhibited by GNAI1 (By similarity). Interacts with STK3/MST2; the interaction inhibits its pro-apoptotic activity. Interacts (when phosphorylated at Ser-259) with YWHAZ (unphosphorylated at 'Thr-232'). Interacts with MAP2K1/MEK1 and MAP2K2/MEK2 (By similarity).

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	Interacts with MAP3K5/ASF1 (via N-terminus) and this interaction inhibits the proapoptotic function of MAP3K5/ASK1. Interacts with PAK1 (via kinase domain). The phosphorylated form interacts with PIN1. The Ser-338 and Ser-339 phosphorylated form (by PAK1) interacts with BCL2. Interacts with PEBP1/RKIP and this interaction is enhanced if RAF1 is phosphorylated on residues Ser-338, Ser-339, Tyr-340 and Tyr-341. Interacts with ADCY2, ADCY5, ADCY6, DGKH, RCAN1/DSCR1, ROCK2, PPP1R12A, PKB/AKT1, PPP2CA, PPP2R1B, SPRY2, SPRY4, CNKSR1/CNK1, KSR2 and PHB/prohibitin. **Subcellular 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 Specificity:** In skeletal muscle, isoform 1 is more abundant than isoform 2. **Post-translational modifications:** Phosphorylated upon DNA damage, probably by ATM or ATR. Phosphorylation at Thr-269, Ser-338, Tyr-341, Thr-491 and Ser-494 results in its activation. Phosphorylation at Ser-29, Ser-43, Ser-289, Ser-296, Ser-301 and Ser-642 by MAPK1/ERK2 results in its inactivation. Phosphorylation at Ser-259 induces the interaction with YWHAZ and inactivates kinase activity. Dephosphorylation of Ser-259 by the complex containing protein phosphatase 1, SHOC2 and M-Ras/MRAS relieves inactivation, leading to stimulate RAF1 activity. Phosphorylation at Ser-338 by PAK1 and PAK7/PAK5 and Ser-339 by PAK1 is required for its mitochondrial localization. DISEASE: Defects in RAF1 are the cause of Noonan syndrome type 5 (NS5) [MIM:611553]. Noonan syndrome (NS) is a disorder characterized by dysmorphic facial features, short stature, hypertelorism, cardiac anomalies, deafness, motor delay, and a bleeding diathesis. It is a genetically heterogeneous and relatively common syndrome, with an estimated incidence of 1 in 1000-2500 live births. Defects in RAF1 are the cause of LEOPARD syndrome type 2 (LEOPARD2) [MIM:611554]. LEOPARD syndrome is an autosomal dominant disorder allelic with Noonan syndrome. The acronym LEOPARD stands for lentigines, electrocardiographic conduction abnormalities, ocular hypertelorism, pulmonic stenosis, abnormalities of genitalia, retardation of growth, and deafness. **Similarity:** Belongs to the protein kinase superfamily. TKL Ser/Thr protein

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	kinase family. RAF subfamily. Contains 1 phorbol-ester/DAG-type zinc finger. Contains 1 protein kinase domain. Contains 1 RBD (Ras-binding) domain. **Database links:** **Entrez Gene:** 5894 Human **Entrez Gene:** 110157 Mouse **Entrez Gene:** 24703 Rat Omim: 164760 Human **SwissProt:** P04049 Human **SwissProt:** Q99N57 Mouse **SwissProt:** P11345 Rat **Unigene:** 159130 Human **Unigene:** 184163 Mouse **Unigene:** 33262 Rat Important **Note:** This product as supplied is intended for research use only, not for use in human, therapeutic or diagnostic applications.