

## Anti-Phospho-RAF1 (Ser43) Rabbit mAb

Purified Recombinant Rabbit Monoclonal Antibody

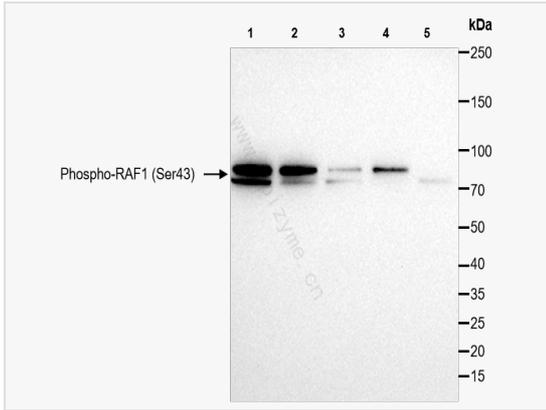
Catalog # R013916

### Product Information

Application	WB, IHC-P/IF (Tissue-P), IF (Cell)/ICC, ELISA
Reactivity	Mouse, Rat, Human
Dilution	WB 1:1,000~1:2,000; IHC-P 1:100~1:200; IF 1:100~1:200
Host	Rabbit
Clonality	Monoclonal
Clone No.	37K08D45
Isotype	IgG
Label	Unconjugated
Immunogen	A synthetic phosphopeptide corresponding to residues surrounding Ser43 of human RAF1
Format	Affinity purified monoclonal antibody supplied in PBS with 0.01% sodium azide and 50% glycerol, pH 7.3.
Storage	Shipped on wet ice. Store at -20°C. Stable for 24 months from date of receipt. Aliquoting is unnecessary for -20°C storage.
Precautions	Anti-Phospho-RAF1 (Ser43) Rabbit mAb [37K08D45] is for research use only and not for use in diagnostic or therapeutic procedures.

### Protein Information

Synonyms	RAF1(phospho Ser43), RAF1(phospho S43), murine leukemia viral (v-raf-1) oncogene homolog 1 (3611-MSV), v-raf-1 murine leukemia viral oncogene homolog 1, cRaf, C-RAF, proto-oncogene c-RAF, 6430402F14Rik, AA990557, BB129353, c-Raf, Craf1, D830050J10Rik, MGC102375, Raf-1, Raf 1, v-Raf, c Raf, Craf 1 transforming gene, Craf1 transforming gene, EC 2.7.11.1, Murine sarcoma 3611 oncogene 1, RAF, Raf proto oncogene serine/threonine protein kinase, vraf1 murine leukemia viral oncogene homolog 1, c-Raf.
Calculated MW	Calculated MW: 73 kDa; Observed MW: 73-80 kDa
Uniprot ID	P04049, Q99N57, P11345
Gene ID	5894, 110157, 24703
Background	This gene is the cellular homolog of viral raf gene (v-raf). The encoded protein is a MAP kinase kinase kinase (MAP3K), which functions downstream of the Ras family of membrane associated GTPases to which it binds directly. Once activated, the cellular RAF1 protein can phosphorylate to activate the dual specificity protein kinases MEK1 and MEK2, which in turn phosphorylate to activate the serine/threonine specific protein kinases, ERK1 and ERK2. Activated ERKs are pleiotropic effectors of cell physiology and play an important role in the control of gene expression involved in the cell division cycle, apoptosis, cell differentiation and cell migration. Mutations in this gene are associated with Noonan syndrome 5 and LEOPARD syndrome 2. [provided by RefSeq, Jul 2008]
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



Western Blot - Anti-Phospho-RAF1 (Ser43) Rabbit mAb [37K08D45]

All lanes: R013916 at 1:1,000 dilution

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

Lane 2: HePG2 (Human hepatocellular carcinoma epithelial cell) whole cell lysates

Lane 3: HCT116 (Human colorectal carcinoma epithelial cell) whole cell lysates

Lane 4: A431 (Human epidermoid teratoma cell line) whole cell lysates

Lane 5: T24 (Human bladder cancer epithelial cell) whole cell lysates

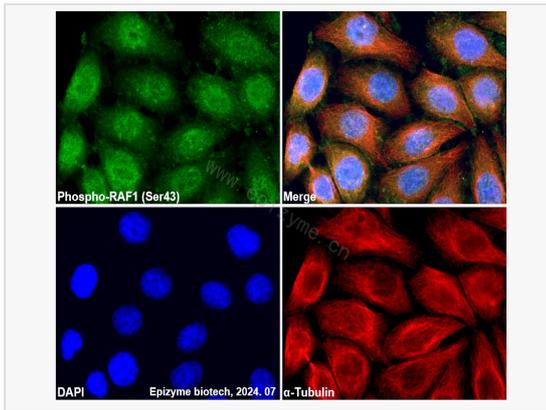
Lysates/proteins at 10 µg per lane.

Secondary antibody: Goat Anti-Rabbit IgG(H+L), HRP Conjugated (Cat. No. LF102) at 1:5,000 dilution

Predicted band size: 73 kDa

Observed band size: 73-80 kDa

Developed using the ECL technique (Cat. No. SQ201).



Immunofluorescence - Anti-Phospho-RAF1 (Ser43) Rabbit mAb [37K08D45]

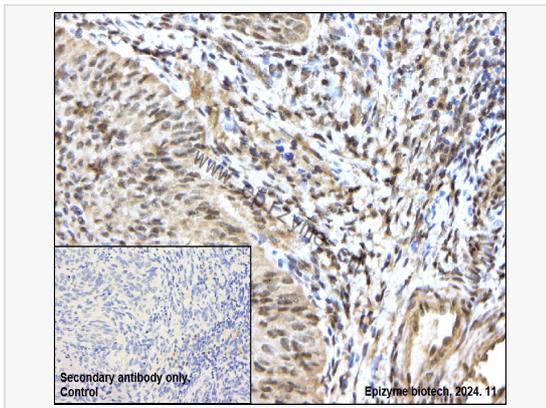
Sample: HeLa cells

The cells were fixed with 4% paraformaldehyde (10 min), permeabilized with 0.5% Triton X-100 for 10 minutes and then blocked with 5% BSA in 0.1% PBS-Tween for 0.5 hours.

Primary antibodies: R013916 at 1:100 dilution and  $\alpha$ -tubulin Mouse Monoclonal Antibody (Cat. No. LF209) at 1:100 dilution

Secondary antibodies: Goat anti-Rabbit (488) at 1:1,000 dilution (shown in green) and Goat anti-Mouse (555) at 1:1,000 dilution (shown in red)

Nuclei were stained with DAPI (shown in blue).



Immunohistochemistry - Anti-Phospho-RAF1 (Ser43) Rabbit mAb [37K08D45]

Sample: Paraformaldehyde-fixed, paraffin embedded human endometrial carcinoma tissue

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins.

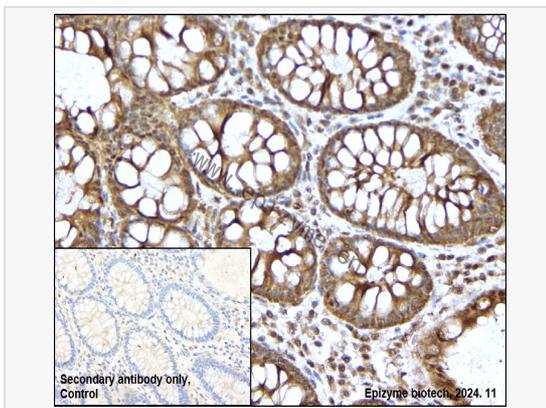
Primary antibody: R013916 at 1:200 dilution

Secondary antibody: Goat Anti-Rabbit IgG (H+L), HRP conjugated at 1:1,000 dilution DAB was used as the chromogen.

Counter stained with hematoxylin.

Positive/negative staining were presented.

Only the secondary antibody was used as the negative control.



Immunohistochemistry - Anti-Phospho-RAF1 (Ser43) Rabbit mAb [37K08D45]

Sample: Paraformaldehyde-fixed, paraffin embedded human colonic cancer tissue

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins.

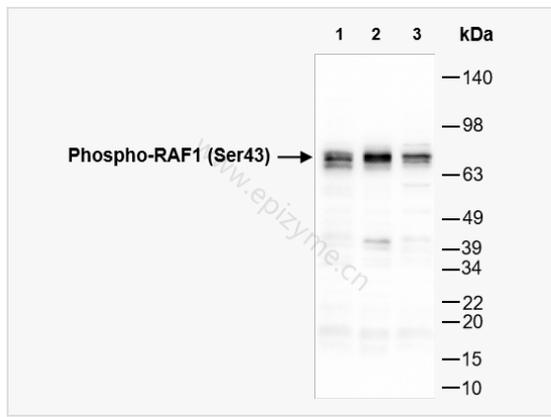
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Counter stained with hematoxylin.

Positive/negative staining were presented.

Only the secondary antibody was used as the negative control.



Western Blot -Anti-Phospho-RAF1 (Ser43) Rabbit mAb [37K08D45]

All lanes: R013916 at 1:1,000 dilution

Lane 1: C2C12 (Mouse myoblasts epithelial cell) whole cell lysates

Lane 2: Raw264.7 (Mouse mononuclear macrophage leukemia cell) whole cell lysates

Lane 3: PC-12 (Rat adrenal pheochromocytoma epithelial cell) whole cell lysates

Lysates/proteins at 10  $\mu$ g per lane.

Secondary antibody: Goat Anti-Rabbit IgG(H+L), HRP Conjugated (Cat. No. LF102) at 1:5,000 dilution

Predicted band size: 73 kDa

Observed band size: 73-80 kDa

Developed using the ECL technique (Cat. No. SQ201).