

Store at
-20°C
#2478

Phospho-VEGF Receptor 2 (Tyr1175) (19A10) Rabbit mAb

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Entrez-Gene ID #3791
UniProt ID #P35968

rev. 09/03/19

For Research Use Only. Not For Use In Diagnostic Procedures.

Applications W, IHC-P, IF-IC Endogenous	Species Cross-Reactivity* H, M	Molecular Wt. 230 kDa	Isotype Rabbit IgG**
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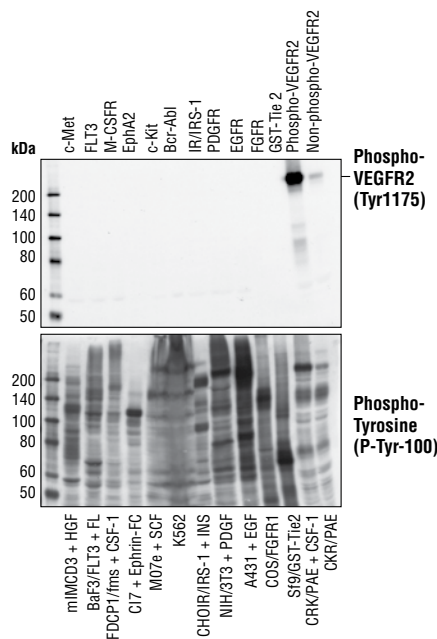
Background: Vascular endothelial growth factor receptor 2 (VEGFR2, KDR, Flk-1) is a major receptor for VEGF-induced signaling in endothelial cells. Upon ligand binding, VEGFR2 undergoes autophosphorylation and becomes activated (1). Major autophosphorylation sites of VEGFR2 are located in the kinase insert domain (Tyr951/996) and in the tyrosine kinase catalytic domain (Tyr1054/1059) (2). Activation of the receptor leads to rapid recruitment of adaptor proteins, including Shc, GRB2, PI3 kinase, NCK, and the protein tyrosine phosphatases SHP-1 and SHP-2 (3). Phosphorylation at Tyr1212 provides a docking site for GRB2 binding and phospho-Tyr1175 binds the p85 subunit of PI3 kinase and PLC γ , as well as Shb (1,4,5). Signaling from VEGFR2 is necessary for the execution of VEGF-stimulated proliferation, chemotaxis and sprouting, as well as survival of cultured endothelial cells *in vitro* and angiogenesis *in vivo* (6-8).

Specificity/Sensitivity: Phospho-VEGF Receptor-2 (Tyr1175) (19A10) Rabbit mAb detects endogenous levels of VEGFR-2 proteins only when phosphorylated at tyrosine 1175. This antibody may cross-react with VEGFR1.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Tyr1175 of human VEGF receptor-2.

Background References:

- (1) Meyer, M. et al. (1999) *EMBO J* 18, 363-74.
- (2) Dougher-Vermazen, M. et al. (1994) *Biochem Biophys Res Commun* 205, 728-38.
- (3) Kroll, J. and Waltenberger, J. (1997) *J Biol Chem* 272, 32521-7.
- (4) Takahashi, T. et al. (2001) *EMBO J* 20, 2768-78.
- (5) Holmqvist, K. et al. (2004) *J Biol Chem* 279, 22267-75.
- (6) Karkkainen, M.J. and Petrova, T.V. (2000) *Oncogene* 19, 5598-605.
- (7) Rahimi, N. et al. (2000) *J Biol Chem* 275, 16986-92.
- (8) Claesson-Welsh, L. (2003) *Biochem Soc Trans* 31, 20-4.



Phospho-VEGF Receptor 2 (Tyr1175) (19A10) Rabbit mAb specifically binds to phosphorylated VEGFR2, but not other phosphorylated tyrosine kinases. Western blot analysis of extracts from cells expressing different activated tyrosine kinase proteins, using Phospho-VEGF Receptor-2 (Tyr1175) (19A10) Rabbit mAb (upper) and Phospho-Tyrosine mAb (P-Tyr-100) #9411 (lower). CRK/PAE cells (lanes 12 and 13) express chimeric receptors containing human CSF-1 extracellular binding domain/mouse VEGF receptor-2 intracellular domain (7). CSF-1 stimulates phosphorylation of Tyr1175 of intracellular VEGF receptor-2 domain (lane 12), which was specifically detected by Phospho-VEGF Receptor-2 (Tyr1175) (19A10) Rabbit mAb.

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 μ g/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

*Species cross-reactivity is determined by western blot.

**Anti-rabbit secondary antibodies must be used to detect this antibody.

Recommended Antibody Dilutions:

Western blotting	1:1000
Immunohistochemistry (Paraffin)	1:50-1:600
<i>Optimal IHC dilutions determined using SignalStain® Boost IHC Detection Reagent.</i>	
Unmasking buffer: SignalStain® Citrate Unmasking Solution (10X) #14746	
Antibody diluent: SignalStain® Antibody Diluent #8112	
Detection reagent: SignalStain® Boost (HRP, Rabbit) #8114	
Immunofluorescence (IF-IC)	1:100-1:400

For product specific protocols and a complete listing of recommended companion products please see the product web page at www.cellsignal.com.

U. S. Patent No. 5,675,063

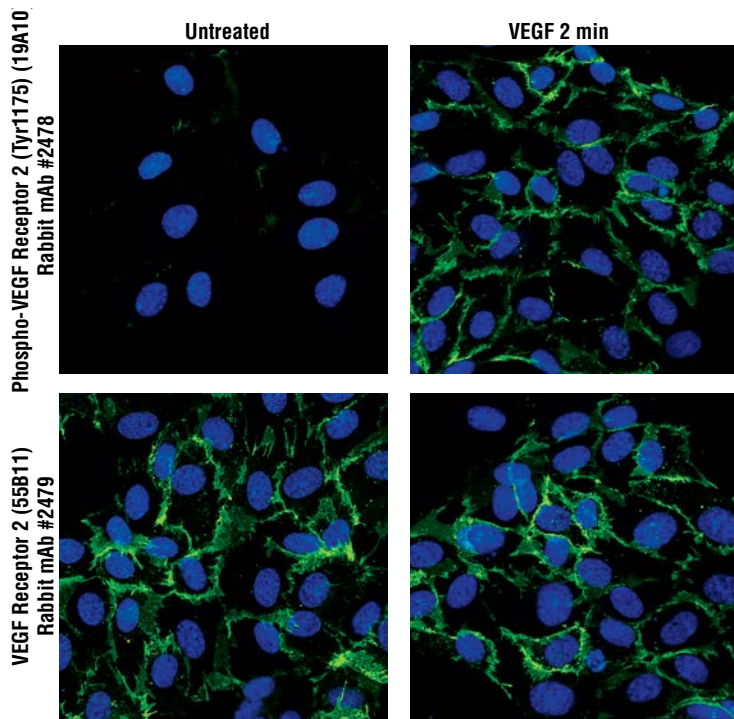
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IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween®20 at 4°C with gentle shaking, overnight.

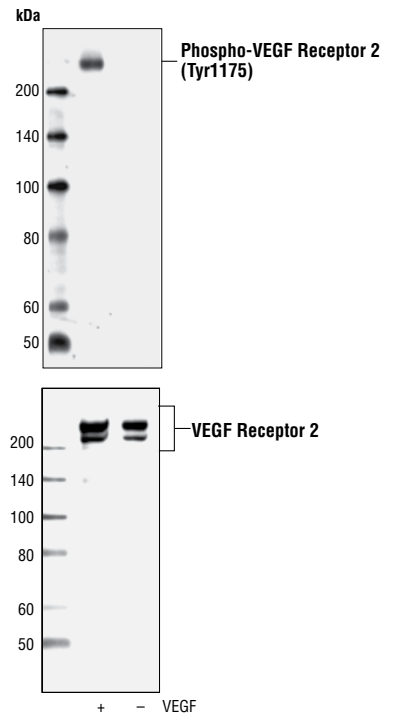
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Applications: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide Species Cross-Reactivity: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—Horse All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.

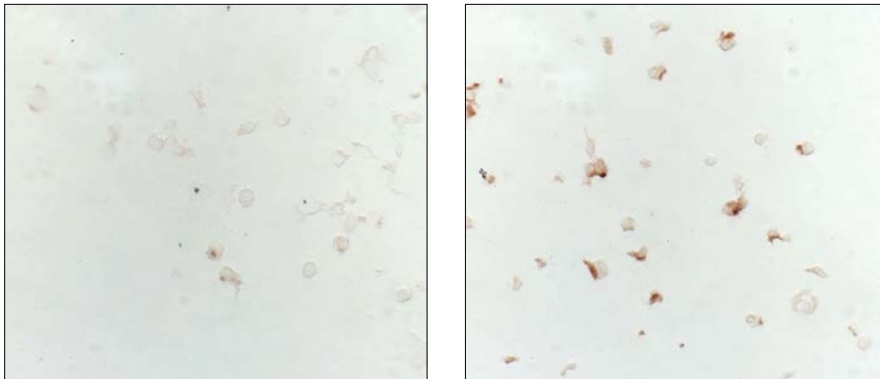




Confocal immunofluorescent images of HUVE cells untreated (left) or stimulated with Vascular Endothelial Growth Factor (VEGF) #9943 (right) and labeled with Phospho-VEGF Receptor 2 (Tyr1175) (19A10) Rabbit mAb (top, green) and VEGF Receptor 2 (55B11) Rabbit mAb #2479 (bottom, green). Blue pseudocolor = DRAQ5[®] #4084 (fluorescent DNA dye).



Western blot analysis of extracts from HUVE cells, untreated or stimulated with VEGF (50 ng/ml for 2 minutes), using Phospho-VEGF Receptor 2 (Tyr1175) (19A10) Rabbit mAb.



Immunohistochemical analysis of paraffin-embedded HUVE cells, untreated (left) or VEGF treated (right).