Store at -20°C

#52928

Phospho-FGF Receptor 1 (Tyr653/654) (D4X3D) Rabbit mAb

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Entrez-Gene ID #2260
UniProt ID #P11362

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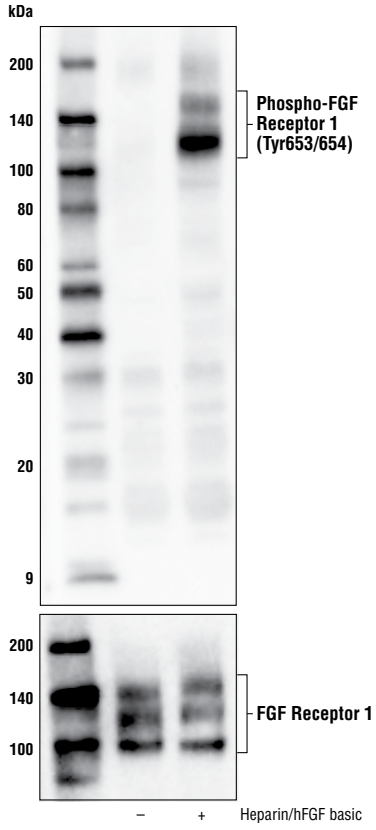
For Research Use Only. Not For Use In Diagnostic Procedures.

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP, IF-IC	H, (M, R)	120, 145 kDa	Rabbit IgG**
Endogenous			

Background: Fibroblast growth factors (FGFs) produce mitogenic and angiogenic effects in target cells by signaling through cell surface receptor tyrosine kinases. There are four members of the FGF receptor family: FGFR1 (flg), FGFR2 (bek, KGFR), FGFR3, and FGFR4. Each receptor contains an extracellular ligand binding domain, a transmembrane domain, and a cytoplasmic kinase domain (1). Following ligand binding and dimerization, the receptors are phosphorylated at specific tyrosine residues (2). Seven tyrosine residues in the cytoplasmic tail of FGFR1 can be phosphorylated: Tyr463, 583, 585, 653, 654, 730, and 766. Tyr653 and Tyr654 are important for catalytic activity of activated FGFR and are essential for signaling (3). The other phosphorylated tyrosine residues may provide docking sites for downstream signaling components such as Crk and PLCγ (4,5).

Specificity/Sensitivity: Phospho-FGF Receptor 1 (Tyr653/654) (D4X3D) Rabbit mAb recognizes endogenous levels of FGFR1 protein when phosphorylated at Tyr653 and/or Tyr654. This antibody exhibits some cross-reactivity with FGF Receptors 2 and 4 when over-expressed exogenously. Based on sequence comparisons, cross-reactivity with FGF Receptor 3 is possible but has not been experimentally confirmed.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic phospho-peptide corresponding to residues surrounding Tyr653 of human FGFR1 protein.



Western blot analysis of extracts from A-204 cells, untreated (-) or treated with heparin (1 µg/mL, 5 min) followed by the Human Basic Fibroblast Growth Factor (hFGF basic/FGF2) #61977 (100 ng/mL, 5 min, +), using Phospho-FGF Receptor 1 (Tyr653/654) (D4X3D) Rabbit mAb (upper) and FGF Receptor 1 (D8E4) XP® Rabbit mAb #9740 (lower).

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/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
Immunoprecipitation	1:100
Immunofluorescence (IF-IC)	1:400-1:1600
Fixative:	4% Formaldehyde
Permeabilization:	0.3% Triton X-100

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

Background References:

(1) Powers, C.J. et al. (2000) *Endocr Relat Cancer* 7, 165-97.
(2) Reilly, J.F. et al. (2000) *J Biol Chem* 275, 7771-8.
(3) Mohammadi, M. et al. (1996) *Mol Cell Biol* 16, 977-89.
(4) Mohammadi, M. et al. (1991) *Mol Cell Biol* 11, 5068-78.
(5) Larsson, H. et al. (1999) *J Biol Chem* 274, 25726-34.

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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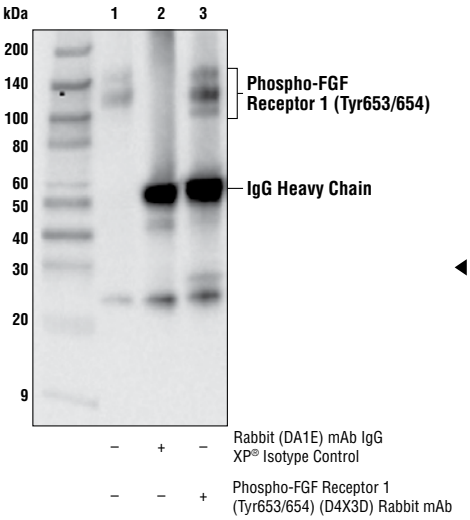
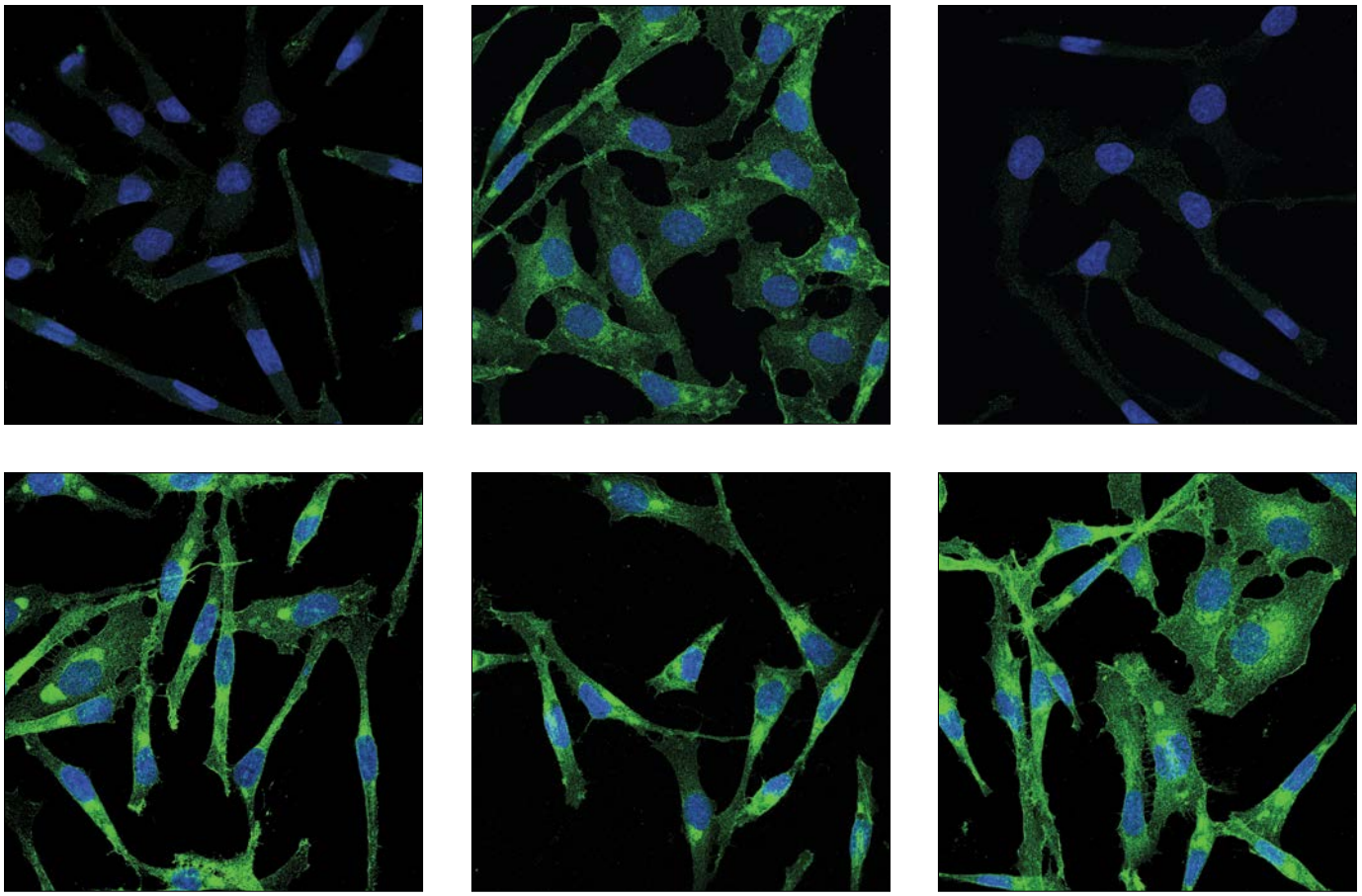
Applications: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide **Species Cross-Reactivity:** **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **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.



◀ *Immunoprecipitation of Phospho-FGF receptor 1 (Tyr653/654) protein from A-204 cell treated with heparin (1 µg/mL, 5 min) followed by the Human Basic Fibroblast Growth Factor (hFGF basic/FGF2) #61977 (100 ng/mL, 5 min). Lane 1 is 10% input, lane 2 is Rabbit (DA1E) mAb IgG XP® Isotype Control #3900, and lane 3 is Phospho-FGF Receptor 1(Tyr653/654) (D4X3D) Rabbit mAb. Western blot analysis was performed using Phospho-FGF Receptor 1(Tyr653/654) (D4X3D) Rabbit mAb as primary antibody. Anti-rabbit IgG, HRP-linked Antibody #7074 was used as secondary antibody.*



Confocal immunofluorescent analysis of serum-starved A-204 cells, untreated (left), treated with heparin (1 µg/mL, 5 min) followed by the addition of Human Basic Fibroblast Growth Factor (hFGF basic/FGF2) #61977 (100 ng/mL, 5 min; center), or treated with heparin/FGF2 and post-processed with λ-phosphatase (2 hr; right), using Phospho-FGF Receptor 1 (Tyr653/654) (D4X3D) Rabbit mAb (upper, green) and FGF Receptor 1 (D8E4) XP® Rabbit mAb #9740 (lower, green). Samples were mounted in ProLong® Gold Antifade Reagent with DAPI #8961 (blue).