Phospho-EphA2 (Tyr594) Antibody

Background: The Eph receptors are the largest known family of receptor tyrosine kinases (RTKs). They can be divided into two groups based on sequence similarity and on their preference for a subset of ligands: EphA receptors bind to a glycosylphosphatidylinositol-anchored ephrin A ligand, and EphB receptors bind to ephrin B proteins that have a transmembrane and cytoplasmic domain (1,2). Eph receptors and ligands may be involved in many diseases including cancer (3). Both ephrin A and ephrin B ligands have dual functions. As RTK ligands, the ephrins stimulate the kinase activity of the Eph receptors and activate signaling pathways in receptor-expressing cells. The ephrin extracellular domain is sufficient for this function as long as it is clustered (4). The second function of ephrins has been described as “reverse signaling,” whereby the cytoplasmic domain becomes tyrosine phosphorylated, allowing interactions with other proteins that may activate signaling pathways in the ligand-expressing cells (5). Various stimuli can induce tyrosine phosphorylation of ephrin B, including binding to EphB receptors, activation of Src kinase and stimulation by PDGF and FGF (6). Tyrosines 324/327 have been identified as major phosphorylation sites of ephrin B1 in vivo (7).

Phosphorylation of Tyr594 was identified in several tumor cell lines (8,9). It was demonstrated that phosphorylated Tyr588 and Tyr594 of EphA2 provide binding sites for EphB receptors, activation of Src kinase and stimulation by PDGF and FGF (6). Tyrosines 324/327 have been identified as major phosphorylation sites of ephrin B, including binding to EphB receptors and activate signaling pathways in receptor-expressing cells. The ephrin extracellular domain is sufficient for this function as long as it is clustered (4). The second function of ephrins has been described as “reverse signaling,” whereby the cytoplasmic domain becomes tyrosine phosphorylated, allowing interactions with other proteins that may activate signaling pathways in the ligand-expressing cells (5). Various stimuli can induce tyrosine phosphorylation of ephrin B, including binding to EphB receptors, activation of Src kinase and stimulation by PDGF and FGF (6). Tyrosines 324/327 have been identified as major phosphorylation sites of ephrin B1 in vivo (7).

Specificity/Sensitivity: Phospho-EphA2 (Tyr594) Antibody detects endogenous levels of EphA2 protein only when phosphorylated on Tyr594. This antibody does not cross-react with other activated protein tyrosine kinases.

Source/Purification: Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Tyr594 of human EphA2. Antibodies are purified by protein A and peptide corresponding to residues surrounding Tyr594 of human EphA2. This antibody does not cross-react with other activated protein tyrosine kinases.

Background References:

Applications Key:
W—Western
M—Immunoprecipitation
IHC—Immunohistochemistry
ChIP—Chromatin Immunoprecipitation
IF—Immunofluorescence
FC—Flow cytometry
ELISA-Peptide

Species Cross-Reactivity Key:
H—human
M—mouse
R—rat
N—rabbit
Mm—monkey
Mi—mink
Ch—chicken
Dm—D. melanogaster
X—Xenopus
Z—zebrafish
B—bovine
D—dog
P—pig
Sc—S. cerevisiae
Ce—C. elegans
Hr—horse

Entrez-Gene ID #1969
Swiss-Prot Acc. #P29317

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA and 50% glycerol. 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

For application specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

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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