

Product Information

Anti-phospho-MAP Kinase Kinase 1/2 (MEK 1/2) (phosphoserine 217/221)

Produced in Rabbit, Affinity Isolated Antibody

Product Number **M 7683**

Product Description

Anti-phospho-MAP Kinase Kinase 1/2 (MEK 1/2) (phosphoserine 217/221) is developed in rabbit using as immunogen a synthetic phosphopeptide SER217/221 corresponding to residues around SER217/221 of human MEK1/2, conjugated to KLH. The antibody is affinity-purified using protein A and peptide affinity chromatography.

Anti-phospho-MAP Kinase Kinase 1/2 (MEK 1/2) (phosphoserine 217/221) detects MEK1/2 only when activated by phosphorylation at SER217 and SER221. This antibody reacts with human, rat, and mouse. Anti-phospho-MAP Kinase Kinase 1/2 does not cross-react with other related family members including SEK (MKK4), MKK3 or MKK6. This antibody reacts equally well with MEK1/2 when phosphorylated only at Ser217 and less well when phosphorylated only at Ser221. The antibody may be used for immunoblotting (45 kDa), immunoprecipitation, immunocytochemistry, immunohistochemistry (paraffin-embedded), and flow cytometry.

MAP Kinase Kinase is also known as MAPKK, mitogen-activated protein kinase kinase, or MEK. MEK1 and MEK2 are dual specificity protein kinases that function in a mitogen activated protein kinase cascade controlling cell growth and differentiation.¹⁻³ MAP kinases are considered to play a crucial role in various signal transduction pathways, leading signals of growth factor tyrosine kinase receptors. MAP Kinase Kinase regulates several cellular processes including proliferation, differentiation, cellular morphology, and oncogenesis.^{4,5} Activation of MEK-1 and MEK-2 in mitogen-stimulated cells is directly mediated by MAP

kinase kinase such as Raf-1 kinase, which phosphorylates two serine residues in the regulatory sites of MEK.^{4,5} Active forms of MEK1/2 are sufficient for the transformation of NIH3T3 cells of the differentiation of PC-12 cells. Following activation, MEK phosphorylates MAP kinase (ERK-1 and ERK-2) in the MAP kinase cascade. MEK isoforms appear to be widely expressed in the central nervous system, thymus, spleen, heart, lung, and kidney.

Reagent

The antibody is supplied in 10 mM sodium HEPES, pH 7.5, containing 150 mM sodium chloride, 100 µg/mL bovine serum albumin, and 50% glycerol.

Storage/Stability

Store at -20 °C. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discarded if not used within 12 hours.

Product Profile

Recommended working dilution is 1:1,000 for immunoblotting (chemiluminescent) using serum treated NIH-3T3 cells. For immunoblotting, incubate membrane with diluted antibody in 5% bovine serum albumin (BSA), 1X Tris buffered saline and 0.1% Tween-20 at 2-8 °C with gentle shaking, overnight.

Immunoblotting note: To reduce basal levels of MEK phosphorylation culture the NIH-3T3 cells in medium containing 0.5% calf serum for 48 hours. Then culture the cells in 0% calf serum for 3 hours prior to treatment.

For immunoprecipitation, a working antibody dilution of 1:50 is recommended.

For immunohistochemistry, a working antibody dilution of 1:100 is recommended using paraffin-embedded tissue sections from human breast carcinoma.

For immunocytochemistry (immunofluorescence), a working antibody dilution of 1:1,000 is recommended using HeLa cells.

For flow cytometry, a working antibody dilution of 1:50 is recommended using PMA treated human peripheral blood lymphocytes and Jurkat cells.

Note: In order to obtain the best results in various techniques and preparations, we recommend determining optimal working dilution by titration.

References

1. Crews, C.M., et al., Science, **258**, 478 (1992).
2. Alessi, D.R., et al., EMBO J., **13**, 1610 (1994).
3. Rosen, L.B., et al., Neuron, **12**, 1207 (1994).
4. Crowley, S., et al., Cell, **77**, 841 (1994).

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