

Product Information

Anti-phospho-ERK1 (pThr²⁰²/pTyr²⁰⁴) & ERK2 (pThr¹⁸⁵/pTyr¹⁸⁷) (MAPK)

Developed in Rabbit, Affinity Isolated Antibody

Product Number **E7028**

Product Description

Anti-phospho-ERK1 (pThr²⁰²/pTyr²⁰⁴) & ERK2 (pThr¹⁸⁵/pTyr¹⁸⁷) (MAPK) is developed in rabbit using a synthetic phosphopeptide derived from the region of human ERK1 & 2 containing threonine 202/185 and tyrosine 204/187, respectively, as immunogen. The serum is affinity purified using epitope-specific affinity chromatography. The antibody is preadsorbed to remove any reactivity towards non-phosphorylated ERK1 & ERK2 enzymes.

This antibody recognizes endogenous active forms of ERK1 & 2 (44 kDa and 42 kDa, respectively) in a variety of cell types, including human, mouse, rat and chick embryo. It has been used in immunoblotting and immunostaining applications.

Extracellular Signal-Regulated Kinases (ERKs) are members of mitogen-activated protein kinase superfamily (MAPK). MAPK cascade is an evolutionary conserved module that mediates the signaling from various extracellular stimuli to the nucleus. The core elements of a MAPK pathway are three sequentially activated protein kinases, terminating in MAP kinase family member. In mammals a MAP/ERK kinase kinase (MEKK) activates a MAP/ERK kinase (MEK), which activates an ERK or MAP kinase.

There are three well defined mammalian MAP kinase modules: the ERK1/2 module, the c-Jun N-terminal protein kinase/stress-activated protein kinase module, and the p38 module. ERK3, ERK4, and ERK5 and other p38 isoforms have also been identified, but the cascades leading to activation of these kinases are not well characterized. Each member of MAPKs is activated by both tyrosine and threonine phosphorylation, catalyzed by distinct upstream kinase, a member of the MAPK kinase family.¹⁻⁴

The well-characterized ERK module is activated in response to stimuli such as cytokines, growth factors, osmotic shock, or UV irradiation. ERKs regulate transcription, cell cycle, differentiation, learning, and memory through signal transduction in the cytoplasm and the nucleus.

ERK1 and 2 phosphorylate microtubule-associated protein-2 (MAP2), myelin basic protein (MBP), and ELK-1. They may promote entry in the cell cycle. The ERK cascade connects to G proteins through a multitude of distinct signal transduction pathways. Both receptor and non-receptor tyrosine kinases play roles in these signaling pathways.⁵

ERK1 (p44) and ERK2 (p42) require the dual phosphorylation in the catalytic kinase domain by MEKs for their full activity. ERK1 is phosphorylated on Tyr²⁰⁴ and Thr²⁰², and ERK2 on Tyr¹⁸⁷ and Thr¹⁸⁵. ERK1 and 2 may also undergo autophosphorylation on these residues. Recent studies have found activated ERK1 and ERK2 in agonist-stimulated platelets, in core proteins of chronic hepatitis C (HCV) virus, and in angiogenin-activated umbilical vein endothelial cells.⁶⁻⁹ G proteins, NO, and ROS play an essential role in ERK1/2 activation.

Reagent

Anti-phospho-ERK1 & 2 is supplied at ~0.5 mg/ml, as a solution in Dulbecco's phosphate buffered saline (without Mg²⁺ and Ca²⁺), pH 7.3, containing 50% glycerol, 1.0 mg/ml BSA (IgG, protease free), and 0.05% sodium azide.

Precautions and Disclaimer

Due to the sodium azide content, a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazards and safe handling practices.

Storage/Stability

Store at -20 °C. Due to the presence of 50% glycerol the antibody will remain in solution. For extended storage, centrifuge the vial briefly before opening and prepare working aliquots. To ensure accurate dilutions mix gently, remove excess solution from pipette tip with clean absorbent paper, and pipette slowly. The antibody is stable for at least six months when stored appropriately. Working dilutions should be discarded if not used within 12 hours.

Product Profile

A recommended working dilution of 1:1000 is determined by immunoblotting using PC12 cells +/- Sorbitol. Data show the specificity of Anti-phospho-ERK1 and ERK2 antibody for the ERK1 and ERK2 phosphorylated on threonine 202/185 and tyrosine 204/187, respectively.

Note: In order to obtain best results in different techniques and preparations we recommend determining optimal working concentration by titration test.

References

1. Elion, E.A., Routing MAP kinase cascades. *Science*, **281**, 1625-1631 (1998).
2. Kamakura, S., et al., Activation of the protein kinase ERK5/BMK1 by receptor tyrosine kinases. *J. Biol. Chem.*, **274**, 26563-26571 (1999).
3. Cobb, M.H., MAP kinase pathways. *Prog. Biophys. Mol. Biol.*, **71**, 479-500 (1999).
4. Khokhlatchev, A., et al., Reconstitution of mitogen-activated protein kinase phosphorylation cascades in bacteria. *J. Biol. Chem.*, **272**, 11057-11062 (1997).
5. Wilsbacher, J.L., et al., Phosphorylation of MAP kinases by MAP/ERK involves multiple regions of MAP kinases. *J. Biol. Chem.*, **274**, 16988-16994 (1999).
6. English, J.M., et al., Identification of substrates and regulators of the mitogen-activated protein kinase ERK5 using chimeric protein kinases. *J. Biol. Chem.*, **273**, 3854-3860 (1998).
7. Gudermann, T., Multiple pathways of ERK activation by G protein-coupled receptors. *Novartis Found. Symp.*, **239**, 68-79 (2001).
8. McNicol, A., et al., Incorporation of map kinases into the platelet cytoskeleton. *Thromb. Res.*, **103**, 25-34 (2001).
9. Liu, S., et al., Angiogenin activates Erk1/2 in human umbilical vein endothelial cells. *Biochem. Biophys. Res. Comm.*, **287**, 305-310 (2001).

AH,JK,MAM 06/05-1

Sigma brand products are sold through Sigma-Aldrich, Inc.

Sigma-Aldrich, Inc. warrants that its products conform to the information contained in this and other Sigma-Aldrich publications. Purchaser must determine the suitability of the product(s) for their particular use. Additional terms and conditions may apply. Please see reverse side of the invoice or packing slip.