

Product Information

ANTI-ERK5 (BIG-MAPK, BMK1) Developed in Rabbit, IgG Fraction of Antiserum

Product Number **E 1523**

Product Description

Anti-ERK5 (big-MAPK, BMK1) is developed in rabbit using a synthetic peptide K-ADIESLQREIQMDS corresponding to the C-terminus of ERK5 of human origin (amino acids 789-802 with N-terminally added lysine) as immunogen. This sequence corresponds to amino acids 790-803 of human BMK1. The peptide is coupled to KLH with glutaraldehyde. Whole antiserum is fractionated and then further purified by ion-exchange chromatography to provide the IgG fraction of antiserum that is essentially free of other rabbit serum proteins.

Anti-ERK5 may be used for the detection and localization of ERK5 by immunoblotting using a whole cell extract of NIH 3T3 mouse fibroblasts.

Mitogen-activated protein kinases (MAP kinases, MAPKs) consist of a family of protein kinases which are considered to play a crucial role in the signal transduction pathways of mammalian cells by leading mitogenic signals to their intracellular targets.^{1,2} MAP kinases regulate several cellular processes including proliferation, differentiation, response to stress and oncogenesis. Several MAP kinase subgroups have been identified in mammalian cells based on their dual phosphorylation motifs Thr-Glu-Tyr (TEY), Thr-Pro-Tyr (TPY) and Thr-Gly-Tyr (TGY). The TEY MAP kinase subgroup includes the extracellular signal-regulated kinases (ERKs) isoforms ERK1 and ERK2 (p44 and p42 MAPK)^{1,3} and ERK5 (also known as big-MAP kinase 1 or BMK1).^{4,5} The TPY group includes the c-Jun N-terminal kinases isoforms (JNK1, JNK2 also termed SAPKs).⁶ The TGY group includes the p38 MAP kinase (also termed RK, Hog1 and CSBP)^{1,7-10} and ERK6 (also termed as SAPK3).¹¹ Each of these subgroups seem to operate in separate MAP kinase signaling cascades. The ERK5/BMK1 gene has been identified in human cell libraries.^{4,5} It has at least three different forms of mRNA and it encodes a protein of 815 to 816 amino acid residues. *In vitro* expressed ERK5 protein interacts specifically with MEK5 but not MEK1, suggesting that the MEK5/ERK5 protein kinases represent a new mammalian signal transduction

pathway. ERK5 contains a distinct 400-amino acid C-terminal and loop-12 domain, which includes two proline-rich regions. The proline-rich region (amino acids 577-699) contains several small Pro-Ala repeats (PA)₃, followed by Pro-Thr repeats (PT)₃. Also, the (PA)_n repeat is present in myosin light chain kinase, and it has been shown to directly interact with actin, thus targeting the kinase to a specific location in the cell.¹² The ERK5 C-terminal sequence, unique among ERKs, may serve as a localization domain and/or regulatory domain. This suggests that ERK5 may regulate signaling events distinct from those regulated by the other ERKs. In addition, ERK5 appears to be activated by stimuli different than those required for ERK1/2 activation. It is activated to a greater extent by H₂O₂ than by growth factors, suggesting that it mediates oxidative stress signals. PDGF, TNF α , angiotensin and phorbol esters, which are potent activators of ERK1/2, only weakly activate ERK5 in cultured rat vascular smooth muscle cells. Moreover, ERK5, but not ERK1/2, is potentially activated by H₂O₂ in a calcium-dependent manner in several human cell types. The oxidative stress-mediated activation of ERK5 seems to be mediated by the tyrosine kinase, c-src.¹⁴ ERK5 is expressed in several human tissues. The highest levels are found in heart, skeletal muscle, placenta and kidney. It is not found in liver. Antibodies that react specifically with ERK5 are useful for the study of the differential tissue expression, intracellular localization of this MAP Kinase isoform in normal and neoplastic tissue.

Reagents

Anti-ERK5 (big-MAPK, BMK1) reacts specifically with ERK5 (97 kD) derived from mouse fibroblast cell extract. The antibody may be used in immunoblotting of NIH 3T3 cultured cells whole extracts. Staining of the ERK5 band (97 kD) is specifically inhibited with ERK5 peptide (human, amino acids 789-802 with N-terminally added lysine). The product is provided as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

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 hazardous and safe handling practices.

Storage/Stability

For continuous use, store at 2-8EC for up to one month. For extended storage freeze in working aliquots. Repeated freezing and thawing is not recommended. Storage in "frost-free" freezers is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

Product Profile

A minimum working dilution of 1: 8,000 is determined by immunoblotting using a whole cell extract of cultured NIH 3T3 mouse fibroblasts.

Note: In order to obtain best results and assay sensitivity in different techniques and preparations we

recommend determining optimal working dilutions by titration test.

References

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