

Protein Kinase Substrate

Inactive p42 MAP Kinase (Erk2)

Concentration: 2 mg/ml

Recombinant

Small 0.1 mg

Large 0.5 mg

rev. 08/07/07



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Background: Both p44 and p42 MAP kinases (Erk1 and Erk2) function in a protein kinase cascade that plays a critical role in the regulation of cell growth and differentiation (1-4). MAP kinases are activated by a wide variety of extracellular signals including growth and neurotrophic factors, cytokines, hormones and neurotransmitters. Activation of MAP kinases occurs through phosphorylation of threonine and tyrosine (202 and 204 of human MAP kinase or 183 and 185 of rat MAP kinase) at the sequence T*EY* by a single upstream MAP kinase kinase (MEK) (5,6). Both kinases are known to weakly autophosphorylate on tyrosine (5).

Description: Inactive p42 MAP Kinase (Erk2) serves as a useful substrate for MEK (MAPK/ERK kinase), which will phosphorylate it at Thr183 and Tyr185 (1). It is expressed as a recombinant full-length untagged protein, and carries a Lys52Arg mutation in the ATP-binding region, rendering it kinase-inactive regardless of its phosphorylation state (2).

Source/Purification: Cloned from a mouse NIH/3T3 cell cDNA library (3) and overexpressed in *E. coli*.

Quality Assurance: The purified protein was run on two identical SDS-polyacrylamide gels. One was stained with Coomassie brilliant blue and the other was blotted to nitrocellulose membrane and the protein band detected with

p44/42 MAP Kinase Antibody (#9102). Greater than 95% of the observable banding was identified as the Inactive p42 MAP Kinase (Erk2) by apparent molecular weight (42 kDa) and 90% of the antibody signal was identified by immunological staining.

Apparent Molecular Weight: 42 kDa

Specific Activity: Inactive p42 MAP Kinase at a concentration of 2 µg/20 µl reaction can be phosphorylated by an up-stream kinase in an *in vitro* kinase assay with 1X Kinase Buffer #9802 and 200 µM ATP #9804. After a 30-minute assay at 30°C, phosphorylation can be detected by Western blot with Phospho-p44/42 MAP Kinase (Thr202/Tyr204) Antibody #9101.

Background References:

- (1) Marshall, C.J. (1995) *Cell* 80, 179-185.
- (2) Hunter, T. (1995) *Cell* 80, 225-236.
- (3) Hill, C.S. and Treisman, R. (1995) *Cell* 80, 199-211.
- (4) Cowley, S. et al. (1994) *Cell* 77, 841-852.
- (5) Sturgill, T.W. et al. (1988) *Nature* 334, 715-718.
- (6) Payne, D.M. et al. (1991) *EMBO J.* 10, 885-892.

Storage: Supplied in 20 mM Tris-HCl (pH 7.5 at 25°C), 50 mM NaCl, 2 mM Na2EDTA, 1 mM dithiothreitol (DTT) and 50% glycerol. Store at -20°C.

Companion Products:

- Phospho-p44/42 MAP Kinase (Thr202/Tyr204) Antibody #9101
- p44/42 MAP Kinase Antibody #9102
- Phospho-p44/42 MAPK (Thr202/Tyr204) (E10) Mouse mAb #9106
- p44/42 MAP Kinase Assay Kit (nonradioactive) #9800
- Kinase Buffer (10X) #9802
- ATP (10 mM) #9804