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Active Human ERK1

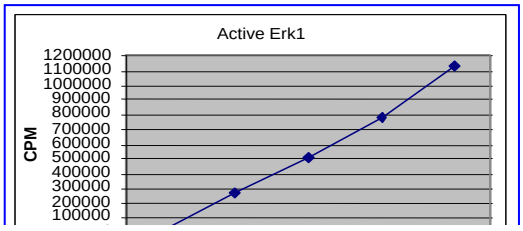
ERK1, Active
(Full-length (tag-free) recombinant human ERK1 expressed in Sf9 cells)

Catalog #: 7741-5
Lot #: _____
Concentration: 0.1 µg/µl
Specific activity: 383 nmol/min/mg

Quality Control Analysis
Activity assessment
ERK1 protein 100 ng/µl concentration) was diluted to 20ng/µl with assay dilution buffer (4 mM MOPS, pH 7.2, 2.5 mM β-glycerophosphate, 1 mM EGTA, 0.4 mM EDTA, 4 mM $MgCl_2$, 0.05 mM DTT), followed by 2-fold serial dilutions, and then the 10µl diluted proteins were used to phosphorylate the MBP using the following assay condition:

10 µl diluted ERK1 protein
5 µl MBP (1 mg/ml stock)
5 µl water
5 µl [^{32}P] ATP mixture (250 µM ATP, 0.16 µCi/µl in 4x assay dilution buffer)

The various reaction components, except [^{32}P] ATP, were incubated at 30° C and the reaction started by the addition of [^{32}P] ATP. After 15 minutes, the reaction was terminated by spotting 20 µl of the reaction mixture onto a phosphocellulose P81 paper. The P81 paper was dried and washed several times in 1% phosphoric acid prior to counting in the presence of scintillation fluid in a scintillation counter. The actual counts, using various dilutions of the enzyme in the assay, are shown in Fig. 1.



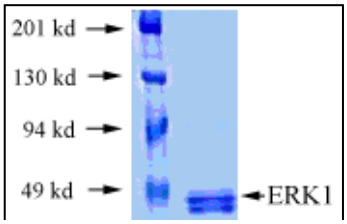


Fig. 1 ERK1 activity assay

Fig. 2 ERK1 protein gel

Purity assessment
1 µg of ERK1 protein was subjected to SDS-PAGE and Coomassie blue staining. The scan of the gel showed >90% purity of the ERK1 product, and the band was at ~45 kDa (Fig. 2).

Product Description
Recombinant full-length human ERK1 was expressed in Sf-9. The ERK1 was activated *in vitro* in the presence of ATP and active MEK1.
Erk1 gene accession number is NM_002746.
This material is sold for research purposes only.

Specific Activity
383 nmol phosphate incorporated into MBP per minute per mg protein at 30° C for 15 minutes using a final concentration of 50 µM ATP (0.83 µCi/assay).

Formulation
Recombinant proteins in storage buffer (50 mM Tris-HCl, pH 7.5, 150 mM NaCl, 0.25 mM DTT, 0.1 mM EGTA, 0.1 mM EDTA, 0.1 mM PMSF, 25% glycerol).

Storage and Stability
Store product frozen at or below -70° C. Stable for 1 year at -70° C as undiluted stock. Aliquot to avoid repeated thawing and freezing.

Scientific Background
ERK1 is a protein serine/threonine kinase that is a member of the extracellular signal-regulated kinases (ERKs), which are activated in response to numerous growth factors and cytokines. Activation of ERK1 requires both tyrosine and threonine phosphorylation that is mediated by MEK. ERK1 is ubiquitously distributed in tissues with the highest expression in heart, brain and spinal cord. In vitro studies indicate that ERK1 phosphorylate both nuclear and cytoplasmic proteins. Activated ERK1 translocates into the nucleus where it phosphorylates various transcription factors (e.g., Elk-1, c-Myc, c-Jun, c-Fos, and C/EBP beta). The consensus primary sequence for substrate phosphorylation by ERK1 has been identified as -Pro-Leu-Ser/Thr-Pro-. ERK1 has been implicated in the control of a broad spectrum of cellular events in many types of cells. In somatic cells, ERK1 activation seems to be triggered after exit from a quiescent state (in G0 or G2) only and then inactivated by entry into a proliferative state.

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