

MEK1 K97M

mitogen-activated protein kinase kinase 1

Recombinant Protein Kinase Substrate

HGNC Symbol: MAP2K1

Synonyms: PRKMK1, MAPKK1, MKK1

Product No.: 0785-0000-1

Lot: 049

Description: Human MEK1, full length, amino acids M_1 - V_{393} (as in <u>NCBI/Protein</u> entry NP_002746.1), mutationally inactivated by mutation $K_{97}M$, untagged, expressed in insect cells

Theoretical MW_{Fusion Protein}: 43,784 Da

Expression host: Sf9 insect cells

Purification: GST-Affinity and size exclusion chromatography

ATPase activity: In an ADP-GloTM assay (Promega) with 10 μ M ATP or 30 μ M ATP, the ATP \rightarrow ADP conversion within 30 min is approx. 1% at a concentration of 100 μ g/ml MEK1 K97M* *detailed ATPase assay conditions on request

Storage buffer: 50 mM HEPES pH 7.5, 100 mM NaCl, 5 mM DTT, 20 % glycerol

Storage temperature: -80°C

For complete recovery, mix well and spin before use. Product must not be stored in diluted solutions, aliquots below 10µl are not advisable. Avoid repeated freeze-thaw cycles!

Protein concentration: 0.299 µg/µl

(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Coomassie stain



Phosphorylation of MEK1 K97M by RAF1 Y340D/Y341D

Radiometric filter binding assay



ADP-Glo[™] assay (Promega)

MSFC membrane (Millipore)



Assay conditions: 70 mM HEPES-NaOH, pH 7.5 3 mM MgCl2 3 µM Na-orthovanadate 1.2 mM DTT 50 µg/ml PEG20.000 ATP: variable concentration 1 % (v/v) DMSO Substrate: 80 µg/ml Kinase: 2 µg/ml **Recombinant Proteins**

3.0 µg MEK1 K97M, Lot049

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MEK1 K97M Recombinant Fusion Protein Amino Acid Sequence		
1	GPLAMTKKKP TPIQLNPAPD GSAVNGTSSA ETNLEALQKK LEELELDEQQ RKRLEAFLTQ	60
61	KQKVGELKDD DFEKISELGA GNGGVVFKVS HKPSGLVMAR MLIHLEIKPA IRNQIIRELQ	120
121	VLHECNSPYI VGFYGAFYSD GEISICMEHM DGGSLDQVLK KAGRIPEQIL GKVSIAVIKG	180
181	LTYLREKHKI MHRDVKPSNI LVNSRGEIKL CDFGVSGQLI DSMANSFVGT RSYMSPERLQ	240
241	GTHYSVQSDI WSMGLSLVEM AVGRYPIPPP DAKELELMFG CQVEGDAAET PPRPRTPGRP	300
301	LSSYGMDSRP PMAIFELLDY IVNEPPPKLP SGVFSLEFQD FVNKCLIKNP AERADLKQLM	360
361	VHAFIKRSDA EEVDFAGWLC STIGLNQPST PTHAAGV	420
1-4: legacy of tag cleavage blue: MEK1 boxed: variation from RefSeg		

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