

DAPK1

death associated protein kinase 1

Recombinant Human Active Protein Kinase

HGNC Symbol: DAPK1

Synonyms: DAPK

Product No.: 0332-0000-1

Lot: 002

Description: Human DAPK1, N-terminal fragment, amino acids M₁-L₃₆₃ (as in [NCBI/Protein](#) entry NP_004929.1), N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: DAPK1 Lot 002, was confirmed as DAPK1 by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 71,227 Da

Expression host: Sf9 insect cells

Purification: GST-Affinity Chromatography

Activation: This kinase was not activated by special procedures

Storage buffer: 50 mM TRIS-HCl pH 8.0, 100 mM NaCl, 5 mM DTT, 4 mM reduced glutathione, 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: 0.680 µg/µl

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

Biochemical Parameters:

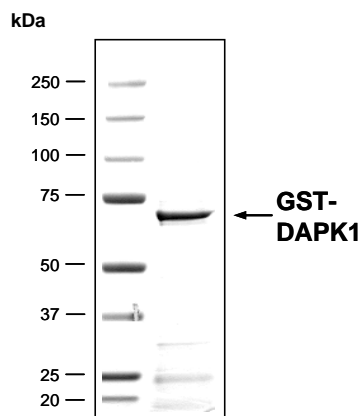
Specific kinase activity (P_i transfer): 85 pmol/µg × min

ATP-K_M: 0.82 µM

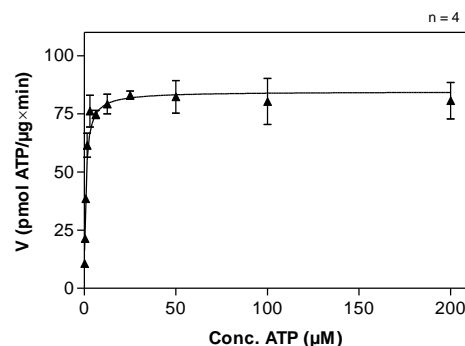
Additional assay technology:

DAPK1 Lot 002 was also successfully tested by ProQinase for the use with the ADP-Glo™ Kinase assay from Promega. ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details

DAPK1 Lot 002: Coomassie stain



DAPK1 Lot 002: Determination of V_{max} and K_M value for ATP



Determination of K_M value & Specific activity:

- Assay conditions:
 - 60 mM HEPES-NaOH, pH 7.5
 - 3 mM MgCl₂
 - 3 mM MnCl₂
 - 3 µM Na-orthovanadate
 - 1.2 mM DTT
 - 50 µg/ml PEG_{20,000}
 - ATP (variable)
 - Substrate: S6-derived peptide 200 µg/ml
 - Kinase: 4 µg/ml
- Filter binding assay
- MSPH membrane (Millipore)

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GST-DAPK1 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPOID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHG	RRRASVAAGI	240
241	LVPRGSPGLD	GICSRNSMTV	FRQENVDDYY	DTGEEELGSGQ	FAVVKCKREK	STGLQYAAKF	300
301	IKKRRTKSSR	RGVSREDIER	EVSILKEIQH	PNVITLHEVY	ENKTDVILIL	ELVAGGELFD	360
361	FLAEKESLTE	EEATEFLKQI	LNGVYYLHSL	QIAHFDLKE	NIMLLDRNVP	KPRIKIIDFG	420
421	LAHKIDFGNE	FKNIFGTPEF	VAPEIVNYEP	LGLEADMWSI	GVITYILLSG	ASPFLGDTKQ	480
481	ETLANVSAVN	YEFEDYFSN	TSALAKDFIR	RLLVKDPKKR	MTIQDSLQHP	WIKPKDTQQA	540
541	LSRKASAVNM	EKFKKFAARK	KWKQSVRLIS	LCQRLSRSFL	SRSNMSVARS	DDTLDEEDSF	600
601	VMKAIHAIN	DDNVPGLQHL					660

1-218: GST **Red:** HIS6-tag **Pink:** Thrombin cleavage site **blue:** DAPK1 fragment

DAPK1 wt ¹ Amino Acid Sequence							
1	MTVFRQENV	DYYDTGEELG	SGQFAVVKKC	REKSTGLQYA	AKFIKKRRTK	SSRRGVSRED	60
61	IEREVSILKE	IQHPNVITLH	EVYENKTDVI	LILELVAGGE	LFDFLAEKES	LTEEEATEFL	120
121	KQILNGVYYL	HSLQIAHFDL	KPENIMLLDR	NVPKPRIKII	DFGLAHKIDF	GNEFKNIFGT	180
181	PEFVAPEIVN	YEPLGLEADM	WSIGVITYIL	LSGASPFGLD	TKQETLANVS	AVNYEFEDEY	240
241	FSNTSALAKD	FIRLLLVKDP	KKRMTIQDSL	QHPWIKPKDT	QQALSRSKASA	VNMEKFKKFA	300
301	ARKKWKQSVR	LISLCQRLSR	SFLSRSNMSV	ARSDDTLDEE	DSFVMKAIH	AINDDNVPLG	360
361	QHLLGSLSNY	DVNQPNKHGT	PPLLIAAGCG	NIQILQLLIK	RGSRIDVQDK	GGSSNAVYWAA	420
421	RHGHVDTLKF	LSENKCPLDV	KDKSGEMALH	VAARYGHADV	AQVTCASAQA	IPISRTKEEE	480
481	TPLHCAAWHG	YYSVAKALCE	AGCNVNIKNR	EGETPLLTAS	ARGYHDIVC	LAEHGADLNA	540
541	CDKDGHIHALH	LAVRRCQMEV	IKTLLSQGCF	VDYQDRHGNT	PLHVACKDGN	MPIVVALCEA	600
601	NCNLDISNKY	GRTPLHLAAN	NGILDVVRYL	CLMGASVEAL	TTDGKTAEDL	ARSEQHEHVA	660
661	GLLARLRKDT	HRGLFIQQLR	PTQNLQPRIK	LKLFHGHS	SGSGKTTLVESLKC	GLLRSFFRRR	720
721	RPRLSSTNSS	RFPSPPLASK	PTVSVSINNL	YPGCENSVR	SRSMMFEPGL	TKGMLEVFVA	780
781	PTHHPHCSAD	DQSTKAIDIQ	NAYLNGVGDF	SVWEFSGNPV	YFCCYDYFAA	NDPTSIHVVV	840
841	FSLEEPYEQ	LNPVIFWLSF	LKSLVPVEEP	IAFGGKLNK	LQVVLVATHA	DIMNVPRPAG	900
901	GEFGYDKDTS	LLKEIRNRFG	NDLHISNKL	VLDAGASGSK	DMKVLNRNLQ	EIRSQIVSVC	960
961	PPMTHLCEKI	ISTLPSWRKL	NGPNQLMSLQ	QFVYDVQDQL	NPLASEEDLR	RIAQQHLSTG	1020
1021	EINIMQSETV	QDVLVLLDPRW	LCTNVLGKLL	SVETPRALHH	YRGRYTVEDI	QRLVPSDVE	1080
1081	ELLQILDAMD	ICARDLSSGT	MVDVPALIKT	DNLHRSWADE	EDEVVMVYGGV	RIVPVEHLTP	1140
1141	FPCGIFHKVQ	VNLCRWIHQQ	STEGDADIRL	WVNGCKLANR	GAELLVLLVN	HGQIEVQVR	1200
1201	GLETEKIKCC	LLLDVSVCTI	ENVMATTLPG	LLTVKHLYSP	QQLREHHEPV	MIYQPRDFFR	1260
1261	AQTLKETSLT	NTMGYKESF	SSIMCFGCHD	VYSQASLGMD	IHASDLNLLT	RRKLSRLDLP	1320
1321	PDPLGKDWCL	LAMNLGLPDL	VAKYNTNNGA	PKDFLPSPLH	ALLREWTTYP	ESTVGTLMASK	1380
1381	LRELGRDAA	DLLLKASSVF	KINLDGNGQE	AYASSCNSGT	SYNSISSVVS	R	1440

blue: DAPK1 sequence expressed in recombinant protein

¹[NCBI/Protein](https://www.ncbi.nlm.nih.gov/Protein/NP_004929.1) accession number NP_004929.1

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