

CDK1/CycE1

cyclin dependent kinase 1

Recombinant Human Active Protein Kinase

HGNC Symbol: CDK1

Synonyms: CDC2

Product No.: 0134-0055-1

Lot: 001

Description: Human CDK1, full length, amino acids M₁-M₂₉₇ (as in [NCBI/Protein](#) entry NP_001777.1) and human CycE1, amino acids M₁-A₃₉₅ (as in [NCBI/Protein](#) entry NP_476530.1), both N-terminally fused to GST-Thrombin cleavage site, coexpressed in Sf9 insect cells

Product identity: CDK1/CycE1 Lot 001, was confirmed as CDK1/CycE1 by specific Western Blotting using anti CDK1 and CycE1 antibodies

Theoretical MW_{CDK1}: 63,882 Da

Theoretical MW_{CycE1}: 72,041 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: 0.166 µg/µl

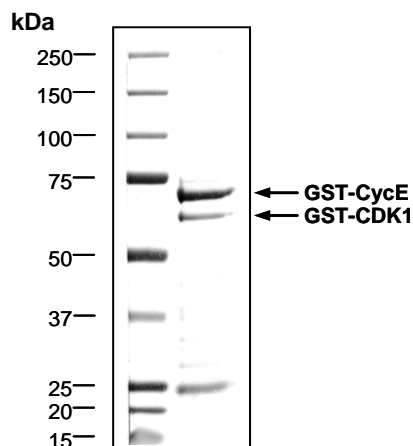
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

Specific kinase activity (P_i transfer): 53 pmol/µg × min

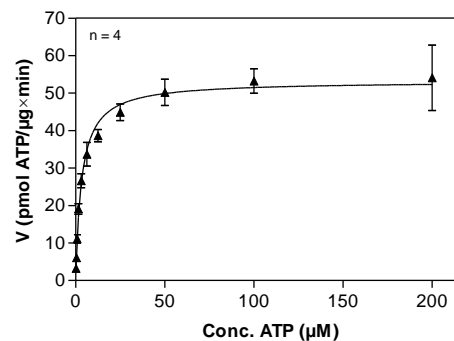
ATP-K_M: 3.3 µM

CDK1/CycE1 Lot 001:
Coomassie stain



2.0 µg CDK1/CycE

CDK1/CycE1 Lot 001:
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: RB-CTF, 100 µg/ml
 - CDK1/CycE1: 4 µg/ml
- Filter binding assay
- MSFC membrane (Millipore)

Additional assay technology:

CDK1/CycE1 Lot 001 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

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CDK1/CycE1

Product No.: 0134-0055-1

GST-CDK1 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	KRIEAIQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMG HHHHH G	RRRASVAAGI	240
241	LVPRGS PGLD	GICSIIEFTM	EDYTKIEKIG	EGTYGVVYKG	RHKTGQVVA	MKKIRLES GE	300
301	EGVPSTAIRE	ISLLKELRHP	NIVSLQDVL	QDSRLYLIFE	FLSMDLKKYL	DSIPPGQYMD	360
361	SSLVKSILYQ	ILQGIIVFCHS	RRVLHRDLK	QNLIDDKGT	IKLADFGLAR	AFGIPIRVYT	420
421	HEVVTWLWYRS	PEVLLGSARY	STPVDIWSIG	TIFAELATKK	PLFHGDSEID	QLFRIFRALG	480
481	TPNNEVWPEV	ESLQDYKNTF	PKWKPGSLAS	HVKNLDEGL	DLLSKMLIYD	PAKRISGKMA	540
541	LNHPYFNLLD	NQIKKM					600

1-218: GST **Red**: HIS6-tag **Pink**: Thrombin cleavage site **blue**: CDK1 **boxed**: variation from RefSeq

CDK1 wt ¹ Amino Acid Sequence							
1	MEDYTKIEKI	GEGTYGVVYK	GRHKTGQV	AMKKIRLES E	EEGVPSTAIR	EISLLKELRH	60
61	PNIVSLQDVL	MQDSRLYLIF	EFLSMDLKKY	LDSIPPGQYM	DSSLVKSILY	QILQGIIVFCH	120
121	SRRVLHRDLK	PQNLIDDKG	TIKLADFGLA	RAFGIPIRVY	THEVVTWLWYR	SPEVLLGSAR	180
181	YSTPVDIWSI	GTIFAELATK	KPLFHGDSEI	DQLFRIFRAL	GTPNNEVWPE	VESLQDYKNT	240
241	FPKWKPGSLA	SHVKNLDEGL	LDLLSKMLIY	DPAKRISGKM	ALNHPYFNLD	DNQIKKM	300

blue: CDK1 sequence expressed in recombinant protein **Red**: variant in recombinant protein

¹[NCBI/Protein](#) accession number NP_001777.1

GST-CycE1 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	KRIEAIQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	LVPRGS PERD	TMKEDGGAEF	240
241	SARSRKRKAN	VTVFLQDPDE	EMAKIDRTAR	DQCGSQPDN	NAVCADPCSL	IPTPKEDDD	300
301	RVYPNSTCKP	RIIAPSRGSP	LPVLSWANRE	EVWKIMLNKE	KTYLRDQHFL	EQHPLLQPKM	360
361	RAILLDWLME	VCEVYKLHRE	TFYLAQDFD	RYMATQENVV	KTLLQLIGIS	SLFIAAKLEE	420
421	IYPLLHQFA	YVTDGACSGD	EILTMELMIM	KALKWRLSPL	TIVSWNLVYM	QVAYLNDLHE	480
481	VLLPQYPQOI	FIQIAELLDL	CVLDVDCLEF	PYGILAASAL	YHFSSSELMQ	KVSGYQWCID	540
541	ENCVKWMPVF	AMVIRETGSS	KLKHFGRVAD	EDAHNIQTHR	DSL DLLDKAR	AKKAMLSEQN	600
601	RASPLPSGLL	TPPQSGKKQS	SGPEMA				660

1-218: GST **Pink**: Thrombin cleavage site **blue**: CycE1

Kinase wt ² Amino Acid Sequence							
1	MKEDGGAEFS	ARSRKRKANV	TVFLQDPDEE	MAKIDRTARD	QCGSQPDN	AVCADPCSLI	60
61	PTPKEDDDR	VYPNSTCKPR	IIAPSRGSP	PVLSWANREE	VWKIMLNKEK	TYLRDQHFL	120
121	QHPLLQPKMR	AILLDWLMEV	CEVYKLHRET	FYLAQDFD	YMATQENVV	TLLQLIGISS	180
181	LFIAAKLEEI	YPPKLHQFAY	VTDGACSGDE	ILTMELMIMK	ALKWRLSPLT	IVSWNLVYMQ	240
241	VAYLNDLHEV	LLPQYPQOIF	IQIAELLDLC	VLDVDCLEFP	YGILAASALY	HFSSSELMQK	300
301	VSGYQWCID	NCVKWMPVFA	MVIRETGSSK	LKHFRGVADE	DAHNIQTHRD	SLDLLDKARA	360
361	KKAMLSEQNR	ASPLPSGLLT	PPQSGKKQSS	GPEMA			420

blue: CycE1 sequence expressed in recombinant protein

²[NCBI/Protein](#) accession number NP_476530.1
HGNC symbol: CCNE1

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