

CDK8/CycC

cyclin-dependent kinase 8 / cyclin C

Recombinant Human Active Protein Kinase

HGNC Symbol: CDK8

Synonyms: K35

Product No.: 0376-0390-1

Lot: 004

Description: Human CDK8, full length, amino acids M₁-Y₄₆₃ (as in [NCBI/Protein](#) entry XP_005266272.1), N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site and human CycC, full length, amino acids M₁-S₂₈₃ (as in [NCBI/Protein](#) entry NP_005181.2), N-terminally fused to HIS₆-Thrombin cleavage site, coexpressed in Sf9 insect cells

Product identity: CDK8/CycC Lot004 was confirmed as CDK8/CycC by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{GST-CDK8}: 83,051 Da

Theoretical MW_{His-CycC} : 37,999 Da

Expression host: Sf9 insect cells

Purification: Immobilized Metal Affinity Chromatography

Activation: This kinase was not activated by special procedures

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

Storage temperature: -80°C

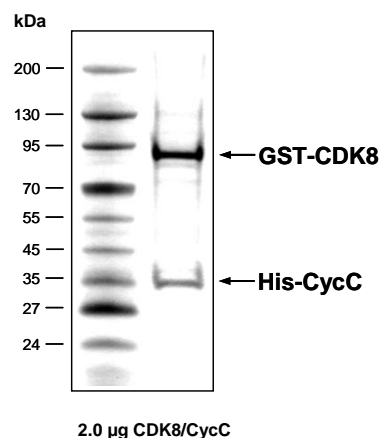
For complete recovery, mix well and spin before use. Product must not be stored in diluted solutions, aliquots below 10µl are not advisable. Avoid repeated freeze-thaw cycles!

Protein concentration: 0.262 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

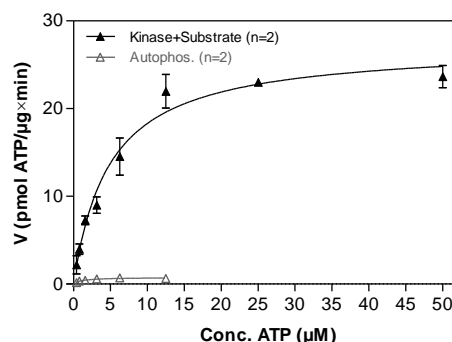
Biochemical Parameters:

Specific kinase activity (P_i transfer): 27 pmol/µg x min
ATP-K_M: 4.9 µM

CDK8/CycC Lot 004: Coomassie stain



CDK8/CycC Lot 004: 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: RBER-IRStide, 100 µg/ml
 - CDK8/CycC: 2 µg/ml
- Filter binding assay
- MSFC membrane (Millipore)

CDK8/CycC

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GST-CDK8 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	LVPKSGPGLD	GIYARGIQAS	MDYDFKVKLS	SERERVEDLF	EYEGCKVGRG	TYGHVYKAKR	300
301	KDGKDDKDYA	LKQIEGTGIS	MSACREIALL	RELKHPNVIS	LQKVFLSHAD	RKVWLLFDYA	360
361	EHDLWHIIFK	HRASKANKKP	VQLPRGMVKS	LLYQILDGIH	YLHANWVLRH	DLKPANILVM	420
421	GEGPERGRVK	IADMGFARLF	NSPLKPLADL	DPVVVTFWYR	APELLLGARH	YTKAIDIWAI	480
481	GCIFAEELLS	EPIFHCQED	IKTSNPYHHD	QLDRIFNVMG	FPADKDWEDI	KKMPEHSTLM	540
541	KDFRRNTYTN	CSLIKMEKH	KVKPDSKAFH	LLQKLLTMDP	IKRITSEQAM	QDPYFLEDPL	600
601	PTSDVFAGCQ	IPYPKREFLT	EEEPDDKGDGK	NQQQQQGNH	TNGTGHPGNQ	DSSHTQGPPL	660
661	KKVRVVPPTT	TSGGLIMTSD	YQRSNPAAAY	PNPGPSTSQP	QSSMGYSATS	QPPQYSHQT	720
721	HRY						780

1-218: GST Red: HIS6-tag Pink: Thrombin cleavage site blue: CDK8

CDK8 wt ¹ Amino Acid Sequence							
1	MDYDFKVKLS	SERERVEDLF	EYEGCKVGRG	TYGHVYKAKR	KDGKDDKDYA	LKQIEGTGIS	60
61	MSACREIALL	RELKHPNVIS	LQKVFLSHAD	RKVWLLFDYA	EHDLWHIIFK	HRASKANKKP	120
121	VQLPRGMVKS	LLYQILDGIH	YLHANWVLRH	DLKPANILVM	GEGPERGRVK	IADMGFARLF	180
181	NSPLKPLADL	DPVVVTFWYR	APELLLGARH	YTKAIDIWAI	GCIFAEELLS	EPIFHCQED	240
241	IKTSNPYHHD	QLDRIFNVMG	FPADKDWEDI	KKMPEHSTLM	KDFRRNTYTN	CSLIKMEKH	300
301	KVKPDSKAFH	LLQKLLTMDP	IKRITSEQAM	QDPYFLEDPL	PTSDVFAGCQ	IPYPKREFLT	360
361	EEEPDDKGDGK	NQQQQQGNH	TNGTGHPGNQ	DSSHTQGPPL	KKVRVVPPTT	TSGGLIMTSD	420
421	YQRSNPAAAY	PNPGPSTSQP	QSSMGYSATS	QPPQYSHQT	HRY		480

blue: CDK8 sequence expressed in recombinant protein

¹[NCBI/Protein](#) accession number XP_005266272.1

HIS-CycC Recombinant Fusion Protein Amino Acid Sequence							
1	MSPIDPMGHH	HHHHGRRRAS	VAAGILVPRG	SPGLDGIYAR	GIQASMAGNF	WQSSHYLQWI	60
61	LDKQDLLKER	QKDLKFLSEE	EYWKLQIFFT	NVIQALGEHL	KLRQQVIATA	TVYFKRFYAR	120
121	YSLKSIDPVL	MAPTCVFLAS	KVEEFGVVS	TRLIAAATSV	LKTRFSYAFP	KEFPYRMNHI	180
181	LECEFYLLEL	MDCCLIVYHP	YRPLLQYVQD	MGQEDMLLPL	AWRIVNDTYR	TDLCLLYPPF	240
241	MIALACLHVA	CVVQKQDARQ	WFAELSVDME	KILEIIRVIL	KLYEQWKNFD	ERKEMATILS	300
301	KMPKPKPPPN	SEGEQGPNGS	QNSSYSQS				360

1-218: GST Red: HIS6-tag Pink: Thrombin cleavage site blue: CycC

CycC wt ¹ Amino Acid Sequence							
1	MAGNFWQSSH	YLQWILDKQD	LLKERQKDLK	FLSEEEYWKL	QIFFTNVIQA	LGEHLKLRQQ	60
61	VIATATVYFK	RFYARYSLKS	IDPVLMAPTC	VFLASKVEEF	GVSNTRLIA	AATSVLKTRE	120
121	SYAFPKEFPY	RMNHILECEF	YLLELMDCC	IVYHPYRPLL	QYVQDMGQED	MLLPLAWRIV	180
181	NDTYRTDLCL	LYPPFMIALA	CLHVACVVQQ	KDARQWFAEL	SVDMEKILEI	IRVILKLYEQ	240
241	WKNFDERKEM	ATILSKMPKP	KPPNSEGEQ	GPNGSQNSSY	SQS		300

blue: CycC sequence expressed in recombinant protein

¹[NCBI/Protein](#) accession number NP_005181.2

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