

ProQinase™ CDK6/CycD3

cyclin dependent kinase 6

Recombinant Human Active Protein Kinase

HGNC Symbol: CDK6

Synonyms: PLSTIRE

Product No.: 0051-0373-1

Lot: 007

Description: Human CDK6, amino acids M1-A326 (as in [NCBI/Protein](#) entry NP_001250.1), N-terminal GST fusion protein with a Thrombin cleavage site and human CycD3, amino acids M1-L292 (as in [NCBI/Protein](#) entry NP_001751.1), N-terminally fused to HIS6-Thrombin cleavage site, coexpressed in Sf9 insect cells

Product identity: CDK6/CycD3 Lot 007, has been verified by mass spectrometry LC-ESI-MS/MS

Theoretical MW _{GST-CDK6} :	63279	Da
Theoretical MW _{HIS-CycD3} :	37260	Da

Expression host: Sf9 insect cells

Purification: GST-Affinity Chromatography

Activation: With CAK

Storage buffer: 50 mM HEPES pH 7.5, 100 mM NaCl, 5 mM DTT, 15 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!

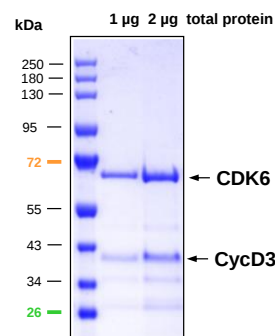
Protein concentration: 0.729 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

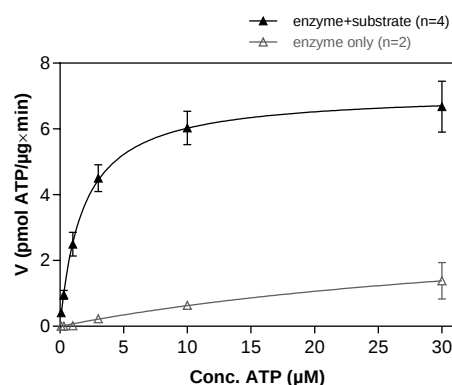
Specific kinase activity (P_i transfer): 7.1 pmol/µg*min

ATP-K_M: 1.8 µM

CDK6/CycD3 Lot 007:
Coomassie stain



CDK6/CycD3 Lot 007:
Determination of V_{max} and K_M value for ATP



- 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-CHKtide 40 µg/ml
- Kinase: 2 µg/ml

Assay technology:
Radiometric filter binding assay
MSFC membrane (96 well plate, Millipore)

Sequence information

GST-CDK6 Recombinant Fusion Protein Amino Acid Sequence

1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLIERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	LVPRGSMKED	GLCRADQQYE	240
241	CVAEIGEGAY	GKVFKARDLK	NGGRFVALKR	VRVQTGEEGM	PLSTIREVAV	LRHLETFEHP	300
301	NVRLFDVCT	VSRTDRETKL	TLVFEHVDQD	LTTYLDKVPE	PGVPTETIKD	MMFQLLRGLD	360
361	FLHSHRVVHR	DLKPQNILVT	SSGQIKLADF	GLARIYSFQM	ALTSVVVTIW	YRAPEVLLQS	420
421	SYATPVDLWS	VGCIFAEMFR	RKPLFRGSSY	VDQLGKILDV	IGLPGEEDWP	RDVALPRQAF	480
481	HSKSAQPIEK	FVTDIDELGK	DLLKCLTFN	PAKRISAYSA	LSHPYFQDLE	RCKENLDSHL	540
541	PPSQNTSELN	TA					600

1-218: GST Red: HIS6-tag Pink: Thrombin cleavage site blue: CDK6 boxed: variation from RefSeq

CDK6 wt¹ Amino Acid Sequence

1	MEKDGLCRAD	QQYECVAEIG	EGAYGKVFKA	RDLKNGGRFV	ALKRVRVQTG	EEGMPLSTIR	60
61	EVAVLRHLET	FEHPNVVRLF	DVCTVSRTDR	ETKLTIVFEH	VDQDLTTYLD	KVPEPGVPTE	120
121	TIKMMFQQL	RGLDFLHSHR	VVHRDLKPQN	ILVTSSGQIK	LADFGGLARIY	SFQMALTSVV	180
181	VTLWYRAPEV	LLQSSYATPV	DLWSVGCIFA	EMFRRKPLFR	GSSDQDLGK	ILDVIGLPGE	240
241	EDWPRDVALP	RQAFHSKSAQ	PIEKFTVDID	ELGKDLLKLC	LTFNPAKRIS	AYSALSHPYF	300
301	QDLERCKENL	DSHLPPSQNT	SELNTA				360

blue: CDK6 sequence expressed in recombinant protein Red: variant in recombinant protein

¹[NCBI/Protein](#) accession number NP_001250.1

HIS-CycD3 Recombinant Fusion Protein Amino Acid Sequence

1	MSPIDPMGHH	HHHHGRRRAS	VAAGILVPRG	SPGLDGIYAR	GIQASMELL	CEGTRHAPRA	60
61	GPDPRLGDO	RVLQSLRL	ERYVPRASYF	QCQVREIKPH	MRKMLAYWML	EVCEEQRCEE	120
121	EVFPLAMNYL	DRYLSCVPTR	KAQLQLLGA	CMLLASKLRE	TTPLTIEKLC	IYTDHAVSPR	180
181	QLRDWEVLVL	GKLKWDLA	IAHDFLAFIL	HRLSLPRDRQ	ALVKKHAQTF	LALCATDYTF	240
241	AMYPSPMIAT	GSIGAAVQGL	GACSMGDEL	TELLAGITGT	EVDCLRACQE	QIEAALRESL	300
301	REAAQTSSSP	APKAPRGSSS	QGSPQTSTPT	DVTAIHL			360

Red: HIS6-tag Pink: Thrombin cleavage site blue: CycD3 boxed: variation from RefSeq

CycD3 wt² Amino Acid Sequence

1	MELLCEGTR	HAPRAGPDPR	LLGDQRLVQS	LLRLEERYVP	RASYFQCVQR	EIKPHMRKML	60
61	AYWMLEVCEE	QRCEEEVFPL	AMNYLDRLS	CVPTRKAQLQ	LLGAVCMLLA	SKLRETTPLT	120
121	IEKLCIYTDH	AVSPRQLRDW	EVLVLGKLKW	DLAAVIAHDF	LAFILHRLSL	PRDRQALVKK	180
181	HAQTFLALCA	TDYTFAMYPP	SMIATGSIGA	AVQGLGACSM	SGDELTELLA	GITGTEVDCL	240
241	RACQEQIEAA	LRESLREASQ	TSSSPAPKAP	RGSSSQGPSQ	TSTPTDVTAI	HL	300

blue: CycD3 sequence expressed in recombinant protein Red: variant in recombinant protein

²[NCBI/Protein](#) accession number NP_001751.1

S259A: SNP variation see [NCBI/dbSNP](#) ID: rs386700585

HGNC identifier CycD3: CCND3