

Certificate of Analysis



CDK17/p35NCK

Cyclin-dependent kinase 17

Recombinant Human Active Protein Kinase

HGNC Symbol: CDK17

Synonyms: PCTAIRE2, PCTK2

Product No.: 1526-0355-1

Lot: 003

Description: Co-expression of human CDK17, amino acids M₁-F₅₂₃ (as in NCBI/Protein entry NP_002586.2), N-terminal GST-HIS₆ fusion protein with a 3C cleavage site and human p35NCK, amino acids M₁-R₃₀₇ (as in NCBI/Protein entry NP_003876.1), N-terminally fused to GST-Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: CDK17/p35NCK Lot 003, was confirmed as CDK17/p35NCK by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{GST-CDK17}: 87,672 Da

Theoretical MW_{GST-p35NCK}: 63,956 Da

Expression: Baculovirus infected Sf9 cells

Purification: Immobilized Metal Affinity Chromatography

Activation: This kinase was activated by coexpression with its physiological cofactor p35NCK

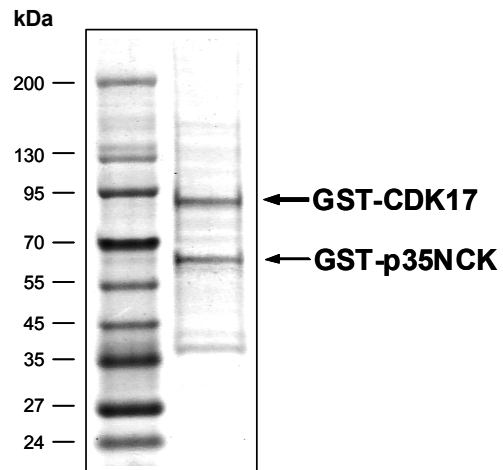
Storage buffer: 50 mM HEPES pH 7.5, 100 mM NaCl, 5 mM DTT, 20% glycerol

Storage temperature: -80°C
Avoid repeated freeze-thaw cycles!

Protein concentration: 0.286 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

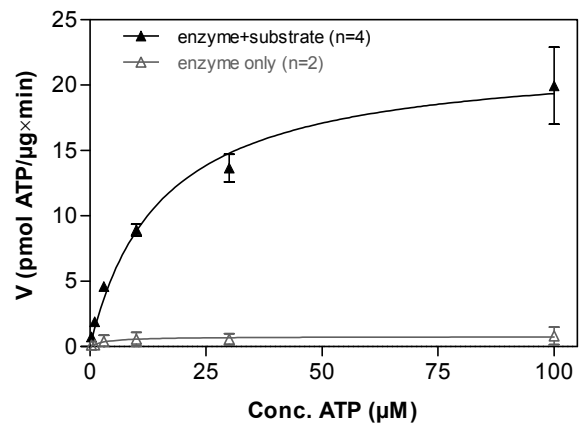
Biochemical Parameters:
Specific kinase activity (P_i transfer): 22 pmol/µg×min
ATP-K_M: 15.0 µM

CDK17/p35NCK Lot 003: Coomassie stain



2.0 µg CDK17/p35NCK

CDK17/p35NCK Lot 003: 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: GSK3-derived peptide (R11-SGRARTSSFAEPGGK), 100 µg / ml
 - CDK17/p35NCK: 4.0 µg / ml
- Filter binding assay
 - MSFC membrane (Millipore)

Additional assay technology: CDK17/p35NCK Lot 003

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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CDK17/p35NCK

Product No.: 1526-0355-1

CDK17 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQ SMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP EML	KMFKDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQG WQATF	GGGDHPPKSD	PMGHHHHHHG	RDSLEVL FQG	240
241	PMKKFKRRLS	LTLRGSQTID	ESLSELAEQM	TIEENSSKDN	EPIVKNGRPP	TSHMHSFLH	300
301	QYTG SFKKPP	LRRPHSVIGG	SLGSFMAMPR	NGSRLDIVHE	NLKMGS DGES	DQASGTSSDE	360
361	VQSPTGVCLR	NRIHRRISME	DLNKRLSLPA	DIRIPDGYLE	KLQINSPPFD	QPMSRRSRRRA	420
421	SLSEIGFGKM	ETYIKLEKLG	EGTYATVYKG	RSKLTENLVA	LKEIRLEHEE	GAPCTAIREV	480
481	SLLKDLKHAN	IVTLHDIVHT	DKSLTLVFEY	LDKDLKQYMD	DCGNIMSMHN	VKFLYQILR	540
541	GLAYCHRRKV	LHRDLKPQNL	LINEKGELKL	ADFGLARAKS	VPTKTYSNEV	VTLWYRPPDV	600
601	LLGSSEYSTQ	IDMWGVGCIF	FEMASGRPLF	PGSTVEDELH	LIFRLLGTPS	QETWPGISSN	660
661	EEFKNYNFPK	YKPQPLINHA	PRLDSEGIEL	ITKFLQYESK	KRVSAAEAMK	HVYFRSLGPR	720
721	IHALPESVSI	FSLKEIQLQK	DPGFRNSSYP	ETGHGKNRRQ	SMLF		780

1-218: GST Red: HIS6-tag Green: 3C cleavage site blue: CDK17

CDK17 wt ¹ Amino Acid Sequence							
1	MKKFKRRLSL	TLRGSQTIDE	SLSELAEQMT	IEENSSK DNE	PIVKNGRPPT	SHSMHSFLHQ	60
61	YTGSFKK PPL	RRPHSVIGGS	LG SFMAMPRN	GSRLDIVHEN	LKMGS DGESD	QASGTSSDEV	120
121	QSPTGVCLRN	RIHRRISMED	LNKRLSLPAD	IRIPDGYLEK	LQINSPPFDQ	PMSRRSRRAS	180
181	LSEIGFGKME	TYIKLEKLGE	GTYATVYKGR	SKLTENLVAL	KEIRLEHEEG	APCTAIREVS	240
241	LLKDLKHANI	VTLHDIVHTD	KSLTLVFEYL	DKDLKQYMD	CGNIMSMHNV	KLFLYQILRG	300
301	LAYCHRRKVL	HRDLKPQNLL	INEKGELKLA	DFGLARAKSV	PTKTYSNEVV	TLWYRPPDVL	360
361	LGSSEYSTQI	DMWGVGCIFF	EMASGRPLFP	GSTVEDELHL	IFRLLGTPSQ	ETWPGISSNE	420
421	EFKNYNFPKY	KPQPLINHAP	RLDSEGIELI	TKFLQYESKK	RVSAAEAMKH	VYFRSLGPRI	480
481	HALPESVSIF	SLKEIQLQKD	PGFRNSSYPE	TGHGKNRRQS	MLF		540

blue: CDK17 sequence expressed in fusionprotein

¹NCBI/Protein accession number NP_002586.2

p35NCK Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQ SMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP EML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQG WQATF	GGGDHPPKSD	PMGHHHHHHG	RRRASVAAGI	240
241	LVPRGSPGLD	GIYARGIQAS	MGTVLSLSPS	YRKATL F E D G	AATVGHY TAV	QNSKNAKDKN	300
301	LKRHSIISVL	PWK RIVAVSA	KKKNSKKVQP	NSSYQNNITH	LNNENLKKSL	SCANLSTFAQ	360
361	PPPAQPPAPP	ASQLSGSQTG	GSSSVK KAPH	PAVTSAGTPK	RVIVQASTSE	LLRCLGEFLC	420
421	RRCYRLK HLS	PTDPVLW LRS	VDRSLLLQGW	QDGFITPAN	VVFLYMLCRD	VISSEVGS DH	480
481	ELQAVLLTCL	YLSYSYMGNE	ISYPLK PFLV	ESCKEAFWDR	CLSVINLMSS	KMLQINADPH	540
541	YFTQVFS DLK	NESGQEDK KR	LLLGLDR				600

1-218: GST Red: HIS6-tag Pink: Thrombin blue: p35NCK boxed: variation from RefSeq

p35NCK wt ² Amino Acid Sequence							
1	MGTVLSLSPS	YRKATL F E D G	AATVGHY TAV	QNSKNAKDKN	LKRHSIISVL	PWK RIVAVSA	60
61	KKKNSKKVQP	NSSYQNNITH	LNNENLKKSL	SCANLSTFAQ	PPPAQPPAPP	ASQLSGSQTG	120
121	GSSSVK KAPH	PAVTSAGTPK	RVIVQASTSE	LLRCLGEFLC	RRCYRLK HLS	PTDPVLW LRS	180
181	VDRSLLLQGW	QDGFITPAN	VVFLYMLCRD	VISSEVGS DH	ELQAVLLTCL	YLSYSYMGNE	240
241	ISYPLK PFLV	ESCKEAFWDR	CLSVINLMSS	KMLQINADPH	YFTQVFS DLK	NESGQEDK KR	300
301	LLLGLDR						360

blue: p35NCK sequence expressed in fusionprotein Red: variant in fusionprotein

²NCBI/Protein accession number NP_003876.1
Q193K: SNP variation see NCBI/dbSNP ID: rs17852832

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