Certificate of Analysis

REACTION BIOLOGY

ProQinase™ CDK15/CycA2

cyclin dependent kinase 15

Recombinant Human Active Protein Kinase

HGNC Symbol: CDK15

Synonyms: PFTAIRE2; ALS2CR7; PFTK2

Product No.: 1889-0054-1

Lot: 002

Description: Human CDK15, internal fragment, amino acids Y_{103}-F_{387} (as in NCBI/Protein entry NP_001248364.1), N-terminal GST-HIS_6 fusion protein with a 3C cleavage site and human cyclin A2, full length, amino acids M1-L432 (as in NCBI/Protein entry NP_001228.1), N-terminal GST-HIS_6 fusion protein with a Thrombin cleavage site, co-expressed in Sf9 insect cells

Product identity: CDK15/CycA2 Lot 002, has been verified by mass spectrometry LC-ESI-MS/MS

Theoretical $MW_{Fusion\ Protein\ CDK15}$: 60,389 Da
Theoretical $MW_{Fusion\ Protein\ CycA2}$: 74,928 Da

Expression host: Sf9 insect cells

Purification: GST-Affinity Chromatography

Activation: This kinase was not activated by special procedures

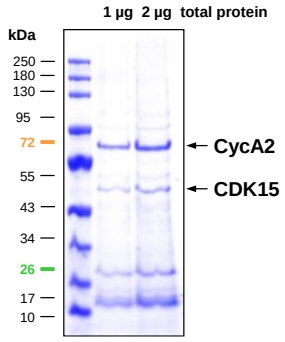
Storage buffer: 50 mM HEPES pH 7.5, 100 mM NaCl, 5 mM DTT, 15 mM reduced glutathione, 20 % glycerol

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!

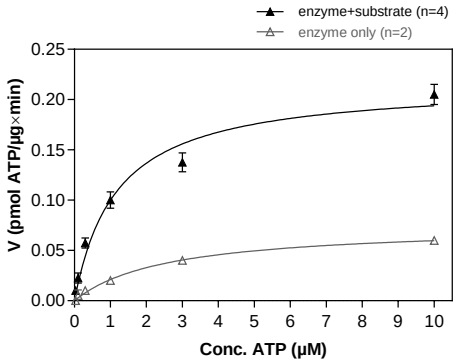
Protein concentration: 0.168 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:
Specific kinase activity (P_i transfer): 0.2 pmol/µg*min
ATP-K_M: 1.2 µM

CDK15/CycA2 Lot 002:
Coomassie stain



CDK15/CycA2 Lot 002:
Determination of V_{max} and K_M value for ATP



- Assay conditions:
  60 mM HEPES-NaOH, pH 7.5
  3 mM $MgCl_2$
  3 mM $MnCl_2$
  3 µM Na-orthovanadate
  1.2 mM DTT
  50 µg/ml $PEG_{20.000}$
  ATP (variable)
  Substrate: RBER-NTRK3tide 80 µg/ml
  Kinase: 4 µg/ml

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

Recombinant Proteins

Sequence information

GST-CDK15 Recombinant Fusion Protein Amino Acid Sequence

```
  1  MSPILGYWKI KGLVQPTRLL LEYLEEKYEE HLYERDEGDK WRNKKFELGL EFPNLPYYID  60
 61  GDVKLTQSMA IIRYIADKHN MLGGCPKERA EISMLEGAVL DIRYGVSRIA YSKDFETLKV 120
121  DFLSKLPEML KMFKDRLCHK TYLNGDHVTH PDFMLYDALD VVLYMDPMCL DAFPKLVCFK 180
181  KRIEAIPQID KYLKSSKYIA WPLQGWQATF GGGDHPPKSD PMGHHHHHHG RDSLEVLFQG 240
241  PYLNLEKLGE GSYATVYKGI SRINGQLVAL KVISMNAEEG VPFTAIREAS LLKGLKHANI 300
301  VLLHDIIHTK ETLTFVFEYM HTDLAQYMSQ HPGGLHPHNV RLFMFQLLRG LAYIHHQHVL 360
361  HRDLKPQNLL ISHLGELKLA DFGLARAKSI PSQTYSSEVV TLWYRPPDAL LGATEYSSEL 420
421  DIWGAGCIFI EMFQGQPLFP GVSNILEQLE KIWEVLGVPT EDTWPGVSKL PNYNPEWFPL 480
481  PTPRSLHVVW NRLGRVPEAE DLASQMLKGF PRDRVSAQEA LVHDYF              540
```

1-218: GST **Red**: HIS6-tag **Green**: 3C cleavage site **blue**: CDK15 fragment

CDK15 wt[1] Amino Acid Sequence

```
  1  MGQELCAKTV QPGCSCYHCS EGGEAHSCRR SQPETTEAAF KLTDLKEASC SMTSFHPRGL  60
 61  QAARAQKFKS KRPRSNSDCF QEEDLRQGFQ WRKSLPFGAA SSYLNLEKLG EGSYATVYKG 120
121  ISRINGQLVA LKVISMNAEE GVPFTAIREA SLLKGLKHAN IVLLHDIIHT KETLTFVFEY 180
181  MHTDLAQYMS QHPGGLHPHN VRLFMFQLLR GLAYIHHQHV LHRDLKPQNL LISHLGELKL 240
241  ADFGLARAKS IPSQTYSSEV VTLWYRPPDA LLGATEYSSE LDIWGAGCIF IEMFQGQPLF 300
301  PGVSNILEQL EKIWEVLGVP TEDTWPGVSK LPNYNPEWFP LPTPRSLHVV WNRLGRVPEA 360
361  EDLASQMLKG FPRDRVSAQE ALVHDYFSAL PSQLYQLPDV SGVRLKPEMC DLLASYQKGH 420
421  HPAQFSKCW                                                        480
```

blue: CDK15 sequence expressed in recombinant protein

[1]NCBI/Protein accession number NP_001248364.1

GST-CycA2 Recombinant Fusion Protein Amino Acid Sequence

```
1    MSPILGYWKI KGLVQPTRLL LEYLEEKYEE HLYERDEGDK WRNKKFELGL EFPNLPYYID  60
61   GDVKLTQSMA IIRYIADKHN MLGGCPKERA EISMLEGAVL DIRYGVSRIA YSKDFETLKV 120
121  DFLSKLPEML KMFEDRLCHK TYLNGDHVTH PDFMLYDALD VVLYMDPMCL DAFPKLVCFK 180
181  KRIEAIPQID KYLKSSKYIA WPLQGWQATF GGGDHPPKSD LVPRGSVMLG NSAPGPATRE 240
241  AGSALLALQQ TALQEDQENI NPEKAAPVQQ PRTRAALAVL KSGNPRGLAQ QQRPKTRRVA 300
301  PLKDLPVNDE HVTVPPWKAN SKQPAFTIHV DEAEKEAQKK PAESQKIERE DALAFNSAIS 360
361  LPGPRKPLVP LDYPMDGSFE SPHTMDMSIV LEDEKPVSVN EVPDYHEDIH TYLREMEVKC 420
421  KPKVGYMKKQ PDITNSMRAI LVDWLVEVGE EYKLQNETLH LAVNYIDRFL SSMSVLRGKL 480
481  QLVGTAAMLL ASKFEEIYPP EVAEFVYITD DTYTKKQVLR MEHLVLKVLT FDLAAPTVNQ 540
541  FLTQYFLHQQ PANCKVESLA MFLGELSLID ADPYLKYLPS VIAGAAFHLA LYTVTGQSWP 600
601  ESLIRKTGYT LESLKPCLMD LHQTYLKAPQ HAQQSIREKY KNSKYHGVSL LNPPETLNL  660
```

1-218: GST **Pink**: Thrombin cleavage site **blue**: CycA2

CycA2 wt[2] Amino Acid Sequence

```
1    MLGNSAPGPA TREAGSALLA LQQTALQEDQ ENINPEKAAP VQQPRTRAAL AVLKSGNPRG  60
61   LAQQQRPKTR RVAPLKDLPV NDEHVTVPPW KANSKQPAFT IHVDEAEKEA QKKPAESQKI 120
121  EREDALAFNS AISLPGPRKP LVPLDYPMDG SFESPHTMDM SIVLEDEKPV SVNEVPDYHE 180
181  DIHTYLREME VKCKPKVGYM KKQPDITNSM RAILVDWLVE VGEEYKLQNE TLHLAVNYID 240
241  RFLSSMSVLR GKLQLVGTAA MLLASKFEEI YPPEVAEFVY ITDDTYTKKQ VLRMEHLVLK 300
301  VLTFDLAAPT VNQFLTQYFL HQQPANCKVE SLAMFLGELS LIDADPYLKY LPSVIAGAAF 360
361  HLALYTVTGQ SWPESLIRKT GYTLESLKPC LMDLHQTYLK APQHAQQSIR EKYKNSKYHG 420
421  VSLLNPPETL NL                                                    480
```

blue: CycA2 sequence expressed in recombinant protein

[2]NCBI/Protein accession number NP_001228.1
HGNC identifier CycA2: CCNA2

Recombinant Proteins