

ProQinase™ ACK1

tyrosine kinase, non-receptor, 2

Recombinant Human Active Protein Kinase

HGNC Symbol: TNK2

Synonyms: p21cdc42Hs

Product No.: 0493-0000-1

Lot: 003

Description: Human ACK1, internal fragment, amino acids G₁₁₀-W₄₇₆ (as in NCBI/Protein entry NP_005772.3), N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: ACK1 Lot 003, was confirmed as ACK1 by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 71,130 Da

Expression: Baculovirus infected Sf9 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: 0.165 µg/µl

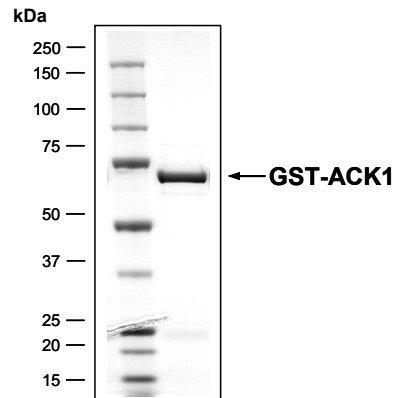
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

Specific kinase activity (P_i transfer): 72 pmol/µg×min

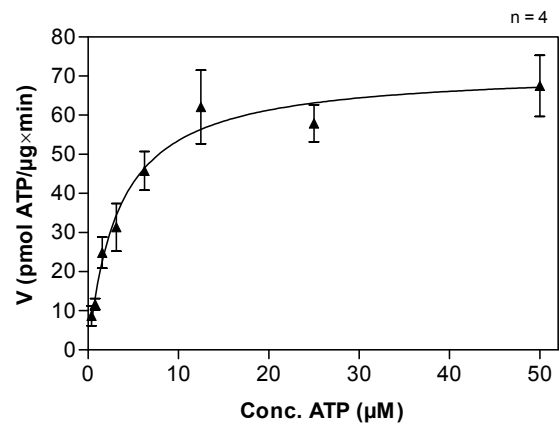
ATP-K_M: 3.4 µM

**ACK1 Lot 003:
Coomassie stain**



2.0 µg GST-ACK1

**ACK1 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: Poly(Glu:Tyr)_{4:1} 40 µg/ml
 - ACK1: 4.0 µg/ml
- Filter binding assay
 - MSFC membrane (Millipore)

Additional assay technology: ACK1 Lot 003

was also successfully tested by Reaction Biology for the use with the ADP-Glo™ Kinase assay from ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details



ProQinase™ ACK1

Product No.: 0493-0000-1

Recombinant Proteins

GST-ACK1 Recombinant Fusion Protein Amino Acid Sequence

| | | | | | | | |
|-----|--------------------|--------------------|-------------------|--------------------|-------------------|-------------------|-----|
| 1 | MSPILGYWKI | KGLVQPTRL | LEYLEEKYEE | HLIERDEGDK | WRNKKFELGL | EFPNLPYYID | 60 |
| 61 | GDVKLTQ SMA | IIRYIADKHN | MLGGCPKERA | EISMLEGAVL | DIRYGVSRIA | YSKDFETLKV | 120 |
| 121 | DFLSKLPEML | KMFEDRLCHK | TYLNGDHVTH | PDFMLYDALD | VVLYMDPMCL | DAFPKLVCFK | 180 |
| 181 | KRIEAIPOID | KYLKSSKYIA | WPLQGWQATF | GGGDHPPKSD | PMGHHHHHG | RRRASVAAGI | 240 |
| 241 | LVPRGS PGLD | GICSRGEGPL | QSLTCLIGEK | DLRLLEKLG D | GSFGVVRGE | WDAPSGKTVS | 300 |
| 301 | VAVKCLKPDV | LSQPEAMDDF | IREVNAMHSL | DHRNLIRLYG | VVLTPEMKMV | TELAPLGSLL | 360 |
| 361 | DRLRKHQGHF | LLGTLSTRYAV | QVAEGMGYLE | SKRFIHRDLA | ARNLLLATRD | LVKIGDFGLM | 420 |
| 421 | RALPQNDHY | VMQEHRKVPF | AWCAPESLKT | RTFSHASDTW | MFGVTLWEMF | TYGQEPWIGL | 480 |
| 481 | NGSQILHKID | KEGERLPRPE | DCPQDIYNVM | VQCAHAKPED | RPTFVALRDF | LLEAQPTDMR | 540 |
| 541 | ALQDFEEDPK | LHIQMNDVIT | VIEGRAENYW | WRGQNRTRLC | VGPFPRNVVT | SVAGLSAQDI | 600 |
| 601 | SQPLQNSFIH | TGHGSDPRH | CW | | | | 660 |

1-218: GST **Red**: HIS6-tag **Pink**: Thrombin cleavage site **blue**: ACK1 fragment

ACK1 wt¹ Amino Acid Sequence

| | | | | | | | |
|------|--------------------|--------------------|--------------------|-------------------|--------------------|--------------------|------|
| 1 | MQPEEGTGWL | LELLSEVQLQ | QYFLRLRDDL | NVTRLSHFEY | VKNEDLEKIG | MGRPGQRRWL | 60 |
| 61 | EAVKRRKALC | KRKSWMKVF | SGKRLEAEFF | PHHSQSTFRK | TSPAPGGPAG | EGPLQSLTCL | 120 |
| 121 | IGEKDLRLL E | KLGDGSFGVV | RRGEWDAPSG | KTVSVAVKCL | KPDVLSQPEA | MDDFIREVNA | 180 |
| 181 | MHSLDHRNLI | RLYGVVLTTP | MKMVTELA PL | GSLLDRLRKH | QGHFLLG TLS | RYAVQVAEGM | 240 |
| 241 | GYLESKRFIH | RDLAARNLLL | ATRDIVKIGD | FGLMRALPQN | DDHYVMQEHR | KVPFAWCAPE | 300 |
| 301 | SLKTRTFSHA | SDTWMFGVTL | WEMFTYQEP | WIGLNGSQIL | HKIDKEGERL | PRPEDCPQDI | 360 |
| 361 | YNVMVQCWAH | KPEDRPTFVA | LRDFLLEAQP | TDMRALQDFE | EPDKLHIQMN | DVITVIEGRA | 420 |
| 421 | ENYWRGQNT | RTL CVGPFPR | NVVT SVAGLS | AQDISQPLQN | SFIHTGHGDS | DPRHCW GFPD | 480 |
| 481 | RIDELYLGNP | MDPPDLSVE | LSTSRPPQHL | GGVKKPTYDP | VSEDQDPLSS | DFKRLGLRKP | 540 |
| 541 | GLPRGLWLAK | PSARVPGTKA | SRGSGAEVTL | IDFGEEPVPV | ALRPCAPSLA | QLAMDACSL | 600 |
| 601 | DETPPQSPTR | ALPRPLHPTP | VVDWDARPLP | PPPAYDDVAQ | DEDDFEICSI | NSTLVGAGVP | 660 |
| 661 | AGPSQGQNTY | AFVPEQARPP | PLEDNLFLP | PQGGGKPPSS | AQTAEIFQAL | QQECMRQLQA | 720 |
| 721 | PAGSPAPSPS | PGGDDKPQVP | PRVPIPPRPT | RPHVQLSPAP | PGEEETSQWP | GPASPPRVPP | 780 |
| 781 | REPLSPQGSR | TPSPLVPPGS | SPLPRLSSS | PGKTMPTTQS | FASDPKYATP | QVIQAPGPRA | 840 |
| 841 | GPCILPIVRD | GKKVSSTHY | LLPERPSYLE | RYQRFLREAQ | SPEEPTPLPV | PLLLPPPSTP | 900 |
| 901 | APAAPTATVR | PMPQAALDPK | ANFSTNNSNP | GARPPPPRAT | ARLPQRGCPG | DGPEAGR PAD | 960 |
| 961 | KIQMAMVHGV | TTEECQAALQ | CHGWSVQRAA | QYLKVEQLFG | LGLRPRGECH | KVLEMFDWNL | 1020 |
| 1021 | EQAGCHLLGS | WGAHHR | | | | | 1080 |

blue: ACK1 sequence expressed in fusionprotein

¹NCBI/Protein accession number NP_005772.3