Certificate of Analysis

REACTION BIOLOGY

ProQinase™ PAK2

p21 (CDKN1A)-activated kinase 2

Recombinant Human Active Protein Kinase

HGNC Symbol: PAK2

Synonyms: PAK65; PAKgamma; hPAK65; S6/H4K

Product No.: 0304-0000-1

Lot: 001

Description: Human PAK2, amino acids D_3-R_{524} (as in NCBI/Protein entry NP_002568.2), N-terminal GST-HIS_6 fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: PAK2 Lot 001, was confirmed as PAK2 by specific Western Blotting

Theoretical $MW_{Fusion\ Protein}$: 87,954 Da

Expression host: Sf9 insect cells

Purification: GST-Affinity Chromatography

Activation: This kinase was not activated by special procedures

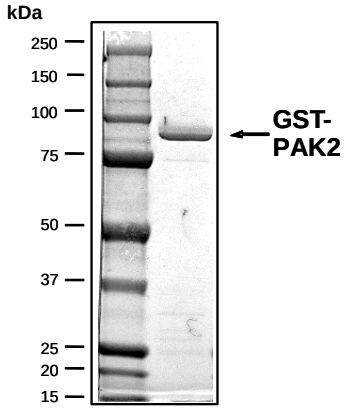
Storage buffer: 50 mM TRIS-HCl pH 8.0, 100 mM NaCl, 5 mM DTT, 15 mM reduced glutathione, 20 % glycerol

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

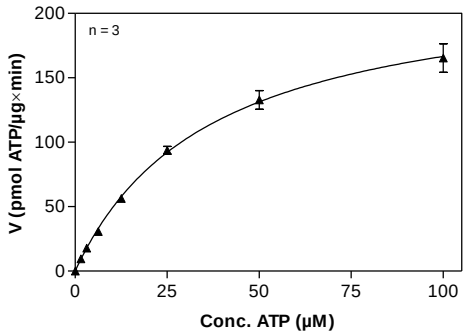
Biochemical Parameters:
Specific kinase activity (P_i transfer): 228 pmol/µg × min
ATP-K_M: 37 µM

PAK2 Lot 001:
Coomassie stain



2.0 µg GST-PAK2

PAK2 Lot 001:
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 $MgCl_2$
  3 mM $MnCl_2$
  3 µM Na-orthovanadate
  1.2 mM DTT
  50 µg/ml $PEG_{20.000}$
  ATP (variable)
  Substrate: Tetra(LRRWSLG), 100 µg/ml
  Kinase: 1 µg/ml
- Filter binding assay
  MSPH membrane (Millipore)

Additional assay technology:
PAK2 Lot 001 was also successfully tested by Reaction Biology 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

Recombinant Proteins

ProQinase™ PAK2

Product No.: 0304-0000-1

GST-PAK2 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 PMGHHHHHHG RRRASVAAGI	240
241	LVPRGSPGLD GIYARGIQAS MCDNGELEDK PPAPPVRMSS TIFSTGGKDP LSANHSLKPL	300
301	PSVPEEKKPR HKIISIFSGT EKGSKKKEKE RPEISPPSDF EHTIHVGFDA VTGEFTGMPE	360
361	QWARLLQTSN ITKLEQKKNP QAVLDVLKFY DSNTVKQKYL SFTPPEKDGF PSGTPALNAK	420
421	GTEAPAVVTE EEDDDEETAP PVIAPRPDHT KSIYTRSVID PVPAPVGDSH VDGAAKSLDK	480
481	QKKKTKMTDE EIMEKLRTIV SIGDPKKKYT RYEKIGQGAS GTVFTATDVA LGQEVAIKQI	540
541	NLQKQPKKEL IINEILVMKE LKNPNIVNFL DSYLVGDELF VVMEYLAGGS LTDVVTETCM	600
601	DEAQIAAVCR ECLQALEFLH ANQVIHRDIK SDNVLLGMEG SVKLTDFGFC AQITPEQSKR	660
661	STMVGTPYWM APEVVTRKAY GPKVDIWSLG IMAIEMVEGE PPYLNENPLR ALYLIATNGT	720
721	PELQNPEKLS PIFRDFLNRC LEMDVEKRGS AKELLQHPFL KLAKPLSSLT PLIMAAKEAM	780
781	KSNR	840

1-218: GST **Red**: HIS6-tag **Pink**: Thrombin cleavage site **blue**: PAK2

PAK2 wt[1] Amino Acid Sequence

1	MSDNGELEDK PPAPPVRMSS TIFSTGGKDP LSANHSLKPL PSVPEEKKPR HKIISIFSGT	60
61	EKGSKKKEKE RPEISPPSDF EHTIHVGFDA VTGEFTGMPE QWARLLQTSN ITKLEQKKNP	120
121	QAVLDVLKFY DSNTVKQKYL SFTPPEKDGF PSGTPALNAK GTEAPAVVTE EEDDDEETAP	180
181	PVIAPRPDHT KSIYTRSVID PVPAPVGDSH VDGAAKSLDK QKKKTKMTDE EIMEKLRTIV	240
241	SIGDPKKKYT RYEKIGQGAS GTVFTATDVA LGQEVAIKQI NLQKQPKKEL IINEILVMKE	300
301	LKNPNIVNFL DSYLVGDELF VVMEYLAGGS LTDVVTETCM DEAQIAAVCR ECLQALEFLH	360
361	ANQVIHRDIK SDNVLLGMEG SVKLTDFGFC AQITPEQSKR STMVGTPYWM APEVVTRKAY	420
421	GPKVDIWSLG IMAIEMVEGE PPYLNENPLR ALYLIATNGT PELQNPEKLS PIFRDFLNRC	480
481	LEMDVEKRGS AKELLQHPFL KLAKPLSSLT PLIMAAKEAM KSNR	540

blue: PAK2 sequence expressed in recombinant protein

[1]NCBI/Protein accession number NP_002568.2

Recombinant Proteins