

Certificate of Analysis



PAK7

p21 (CDKN1A)-activated kinase 7

Recombinant Human Active Protein Kinase

HGNC Symbol: PAK7

Synonyms: PAK5, KIAA1264

Product No.: 0410-0000-1

Lot: 001

Description: Human PAK7, full length, amino acids M₁-H₇₁₉ (as in NCBI/Protein entry NP_065074.1), N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: PAK7 Lot 001, was confirmed as human PAK7 by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 110,660 Da

Expression: Baculovirus infected Sf9 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, 4 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: 0.152 µg/µl

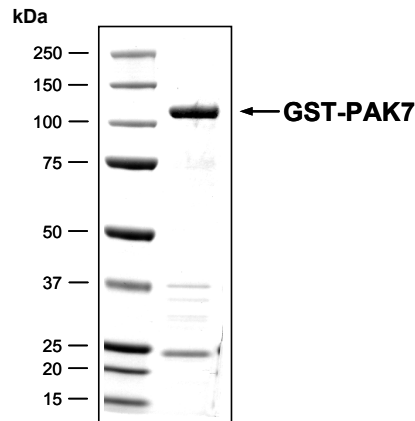
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

Specific kinase activity (P_i transfer): 18 pmol/µg×min

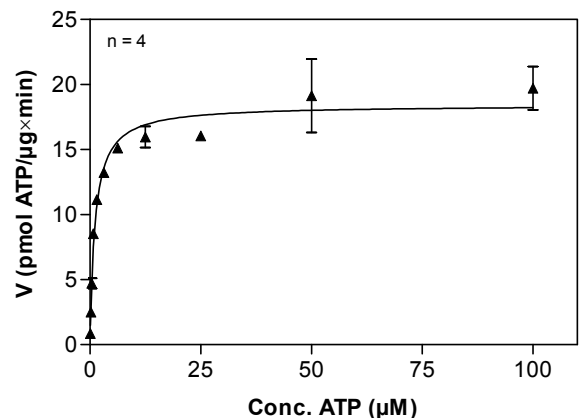
ATP-K_M: 1.1 µM

**PAK7 Lot 001:
Coomassie stain**



2.0 µg GST-PAK7

**PAK7 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 MgCl₂
 - 3 mM MnCl₂
 - 3 µM Na-orthovanadate
 - 1.2 mM DTT
 - 50 µg / ml PEG_{20,000}
 - ATP (variable)
 - Substrate: Tetra(LRRWSLG), 40 µg/ml
 - PAK7: 4.0 µg/ml
- Filter binding assay
 - MSFC membrane (Millipore)

Additional assay technology: PAK7 Lot 001

was also successfully tested by ProQinase for the use with the ADP-Glo™ Kinase assay from Promega



PAK7

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PAK7 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQ SMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQGQWATF	GGGDHPPKSD	PMGHHHHHHG	RRRASVAAGI	240
241	LVPRGSPGLD	GICSIEEFGT	MFGKKKKKIE	ISGPSNFEHR	VHTGFDPQEQ	KFTGLPQQWH	300
301	SLLADTANRP	KPMVDPSCIT	PIQLAPMKTI	VRGNKPKET	SINGLLEDFD	NISVTRSNSL	360
361	RKESPPTPDQ	GASSHGPGHA	EENGFITFSQ	YSESDDTTAD	YTTEKYREKS	LYGDDLD PYY	420
421	RGSHA AKQNG	HVMKMKHGEA	YYSEVKPLKS	DFARFSADYH	SHLDSLKSPS	EYSDLKWEYQ	480
481	RASSSSPLDY	SFQFTPSRTA	GTSGCSKESL	AYSESEWGPS	LDDYDRRPKS	SYLNQTS PQP	540
541	TMRQRSRSGS	GLQEPMPFPG	ASAFKTHPQG	HSYNSYTYPR	LSEPTMCIPK	VDYDRAQMVL	600
601	SPPLSGSDTY	PRGPAKLPQS	QSKSGYSSSS	HQYPSGYHKA	TLYHHPSLQS	SSQYISTASY	660
661	LSSLSLSSST	YPPPSWGSSS	DQQPSRV SHE	QFRAALQLV V	SPGDPREYLA	NFIKIGEGST	720
721	GIVCIATEKH	TGKQVAVKKM	DLRKQQRREL	LFNEVVIMRD	YHHDNVVDMY	SSYLVGDELW	780
781	VVMEFLE GGA	LTDIVTHTRM	NEEQIATVCL	SVLRALSYLH	NQGV IHRDIK	SDSILLTSDG	840
841	RIKLSDFGFC	AQVSKEVPKR	KSLVGT PYWM	APEVISRLPY	GTEVDIWSLG	IMVIEMIDGE	900
901	PPYFNEPPLQ	AMRRIRDSLP	PRVKDLHKVS	SVLRGF LDM	LVREPSQRAT	AQELLGHPFL	960
961	KLGPSPSCIV	PLMRQYRHH					1020

1-218: GST **Red**: HIS6-tag **Pink**: Thrombin cleavage site **blue**: PAK7

PAK7 wt ¹ Amino Acid Sequence							
1	MFGKKKKKIE	ISGPSNFEHR	VHTGFDPQEQ	KFTGLPQQWH	SLLADTANRP	KPMVDPSCIT	60
61	PIQLAPMKTI	VRGNKPKET	SINGLLEDFD	NISVTRSNSL	RKESPPTPDQ	GASSHGPGHA	120
121	EENGFITFSQ	YSESDDTTAD	YTTEKYREKS	LYGDDLD PYY	RGSHA AKQNG	HVMKMKHGEA	180
181	YYSEVKPLKS	DFARFSADYH	SHLDSLKSPS	EYSDLKWEYQ	RASSSSPLDY	SFQFTPSRTA	240
241	GTSGCSKESL	AYSESEWGPS	LDDYDRRPKS	SYLNQTS PQP	TMRQRSRSGS	GLQEPMPFPG	300
301	ASAFKTHPQG	HSYNSYTYPR	LSEPTMCIPK	VDYDRAQMVL	SPPLSGSDTY	PRGPAKLPQS	360
361	QSKSGYSSSS	HQYPSGYHKA	TLYHHPSLQS	SSQYISTASY	LSSLSLSSST	YPPPSWGSSS	420
421	DQQPSRV SHE	QFRAALQLV V	SPGDPREYLA	NFIKIGEGST	GIVCIATEKH	TGKQVAVKKM	480
481	DLRKQQRREL	LFNEVVIMRD	YHHDNVVDMY	SSYLVGDELW	VVMEFLE GGA	LTDIVTHTRM	540
541	NEEQIATVCL	SVLRALSYLH	NQGV IHRDIK	SDSILLTSDG	RIKLSDFGFC	AQVSKEVPKR	600
601	KSLVGT PYWM	APEVISRLPY	GTEVDIWSLG	IMVIEMIDGE	PPYFNEPPLQ	AMRRIRDSLP	660
661	PRVKDLHKVS	SVLRGF LDM	LVREPSQRAT	AQELLGHPFL	KLGPSPSCIV	PLMRQYRHH	720

blue: PAK7 sequence expressed in fusionprotein

¹NCBI/Protein accession number NP_065074.1