

Certificate of Analysis



SNK

Serum inducible kinase

Recombinant Human Active Protein Kinase

HGNC Symbol: PLK2

Synonyms: PLK2

Product No.: 0277-0000-1

Lot: 007

Description: Human SNK, amino acids M₁-N₆₈₅ (as in NCBI/Protein entry NP_006613.2), N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: SNK Lot 007, was confirmed as SNK by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 107,562 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, 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 10 µl are not advisable. Avoid repeated freeze-thaw cycles!

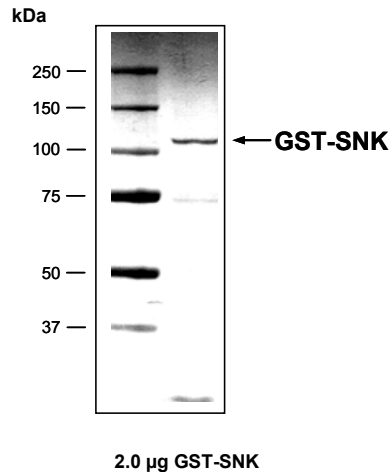
Protein concentration: 0.035 µg/µl

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

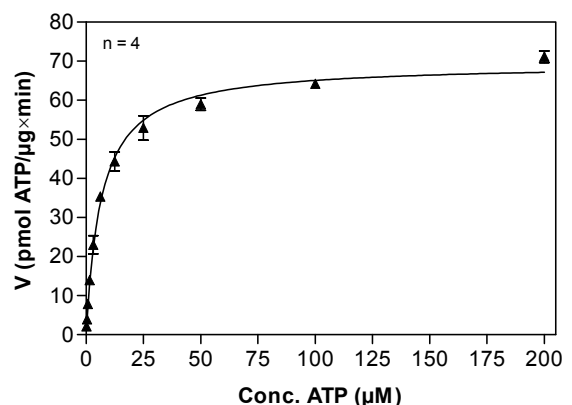
Biochemical Parameters:

Specific kinase activity (P_i transfer): 69 pmol/µg×min
ATP-K_M: 6.6 µM

SNK Lot 007: Coomassie stain



SNK Lot 007: 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: CDC25C-derived peptide (R₁₁-ISDELMDATFADQEA), 200 µg / 50 µl
 - SNK: 4.0 µg / ml
- Filter binding assay
 - MSPH membrane (Millipore)

Additional assay technology: SNK Lot 007

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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SNK Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSM	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHHG	RRRASVAAGI	240
241	LVPRGSPGLD	GICSSMELLR	TITYQPAAST	KMCEQALGKG	CGADSKKKRP	PQPPEESQPP	300
301	QSQAQVPPAA	PHHHHHSHS	GPEISRIIVD	PTTGKRYCRG	KVLGKGGFAK	CYEMTDLTNN	360
361	KVYAAKIIPH	SRVAKPHQRE	KIDKEIELHR	ILHHKHVVQF	YHYFEDKENI	YILLEYCSRR	420
421	SMAHILKARK	VLTEPEVRY	LRQIVSGLKY	LHEQEILHRD	LKLGNTFFINE	AMELKVGDG	480
481	LAARLEPLEH	RRRTICGTPN	YLSPEVLNKQ	GHGCESDIWA	LGCVMYTMLL	GRPPFETTNN	540
541	KETYRCIREA	RYTMPSSLLA	PAKHLIASML	SKNPEDRPSL	DDIIRHDFFL	QGFTPDLRSS	600
601	SCCHTVPDFH	LSSPAKNFFK	KAAAALFGGK	KDKARYIDTH	NRVSKEDEDI	YKLRHDLKKT	660
661	SITQQPSKHR	TDEELQPPTT	TVARSGTPAV	ENKQQIGDAI	RMIVRGTLGS	CSSSSECLD	720
721	STMGSVADTV	ARVLRGCLN	MPEADCIPKE	QLSTSFQWVT	KWVDYSNKYG	FGYQLSDHTV	780
781	GVLFNNGAHM	SLLPDKKTVH	YYAELGQCSV	FPATDAPEQF	ISQVTVLKYF	SHYMEENLMD	840
841	GGDLPSVTDI	RRPRLYLLQW	LKSDKALMML	FNDGTFQVNF	YHDHTKIIIC	SQNEEYLLTY	900
901	INEDRISTTF	RLTTLMSGC	SSELKNRMEY	ALNMLLQRCN			960

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

SNK wt ¹ Amino Acid Sequence							
1	MELLRTITYQ	PAASTKMCEQ	ALGKCGGADS	KKKRPPQPPE	ESQPPQSQAQ	VPPAAPHHHH	60
61	HHSHSGPEIS	RIIVDPTTGK	RYCRGKVLGK	GGFAKCYEMT	DLTNNKVYAA	KIIPHSRVAK	120
121	PHQREKIDKE	IELHRILHHK	HVVQFYHYFE	DKENIYILLE	YCSRRSMAHI	LKARKVLTEP	180
181	EVRYYLRLQIV	SGLKYLHEQE	ILHRDLKLG	FFINEAMELK	VGDFGLAARL	EPLHRRRTI	240
241	CGTPNYLSPE	VLNKQGHGCE	SDIWLGCVM	YTMLLGRPPF	ETTNLKETYR	CIREARYTMP	300
301	SLLAPAKHL	IASMLSKNPE	DRPSLDDIIR	HDFFLQGFTP	DRLSSSCCHT	VPDFHLSSPA	360
361	KNFFKKAAAA	LFGGKKDKAR	YIDTHNRVSK	EDEDIYKLRH	DLKKTSTITQQ	PSKHRTDEEL	420
421	QPPTTTTARS	GTPAVENKQQ	IGDAIRMIVR	GTLGSCSSSS	ECLEDSTMG	VADTVARVLR	480
481	GCLNMPHAD	CIPKEQLSTS	FQWVTKWVDY	SNKYGFGYQL	SDHTVGVLFN	NGAHMSLLPD	540
541	KKTVHYAEL	GQCSVFPATD	APEQFISQVT	VLKYFSHYME	ENLMDGGDLP	SVTDIRRPRL	600
601	YLLQWLKSDK	ALMMLFNDGT	FQVNFYHDHT	KIIICSQNEE	YLLTYINEDR	ISTTFRLLTL	660
661	LMSGCSSELK	NRMEYALNML	LQRCN				720

blue: SNK sequence expressed in fusionprotein

¹NCBI/Protein accession number NP_006613.2

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