

PKC-theta

protein kinase C theta

Recombinant Human Active Protein Kinase

HGNC Symbol: PRKCQ

Synonyms: PRKCT, nPKC-theta

Product No.: 0113-0000-1

Lot: 008

Description: Human PKC-theta, full length, amino acids M₁-S₇₀₆ (as in [NCBI/Protein](#) entry NP_006248.1), N-terminal GST-, C-terminal HIS₆ fusion protein with a FactorXa cleavage site, expressed in Sf9 insect cells

Product identity: PKC-theta Lot 008, was confirmed as PKC-theta by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 109,667 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, 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: 0.273 µg/µl

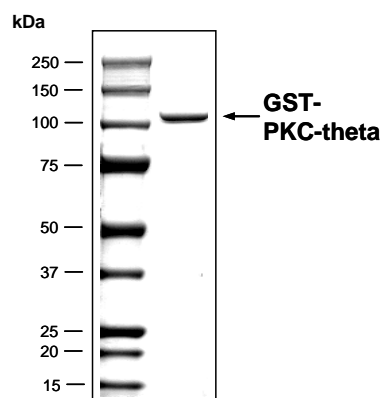
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

Specific kinase activity (P_i transfer): 166 pmol/µg × min

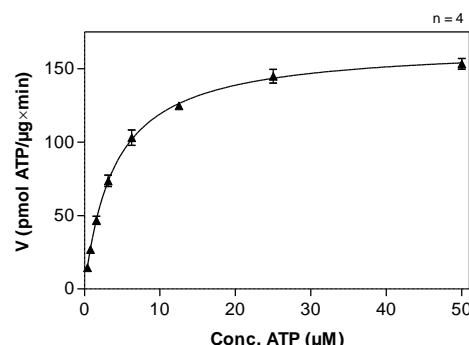
ATP-K_M: 3.9 µM

PKC-theta Lot 008:
Coomassie stain



2.0 µg GST-PKC-theta

PKC-theta Lot 008:
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
 - 5 µg/ml phosphatidyl-serine
 - 1 µg/ml 1,2 dioleoyl-glycerol
 - 1.2 mM DTT
 - 50 µg/ml PEG_{20,000}
 - ATP (variable)
 - Substrate: Histone H2B 30 µg/ml
 - Kinase: 4 µg/ml
- Filter binding assay
- MSFC membrane (Millipore)

Additional assay technology:

PKC-theta Lot 008 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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GST-PKC-theta Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPIQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	L IEGR GIPLQ	GATMSPFLRI	240
241	GLSNFDCGSC	QSCQGEAVNP	YCAVLVKEYV	ESENGQMYIQ	KKPTMYPPWD	STFDAHINKG	300
301	RVMQIIVKKG	NVDLISETTV	ELYSLAERCR	KNNGKTEIWL	ELKPQGRMLM	NARYFLEMSD	360
361	TKDMNEFETE	GFFALHQRRG	AIQAKVHHV	KCHEFTATFF	PQPTFCSVCH	EFVWGLNKQG	420
421	YQCRQCNAAI	HKKCIDKVIA	KCTGSAINSR	ETMFHKERFK	IDMPHRFKVY	NYKSPTFCEH	480
481	CGTLLWGLAR	OGLKCDACGM	NVHRCQTKV	ANLCGINQKL	MAEALAMIES	TQQARCLRDT	540
541	EQIFREGPVE	IGLPCSIKNE	ARPPCLPTPG	KREPQGISWE	SPLDEVDKMC	HLPEPELNKE	600
601	RPSLQIKLKI	EDFILHKMLG	KGSFGKVFLA	EFKKTNQFFA	IKALKKDVVL	MDDDVECTMV	660
661	EKRVLSLAWE	HPFLTHMFCT	FQTKENLFFV	MEYLNGGDLM	YHIQSCHKFD	LSRATFYAAE	720
721	IILGLQFLHS	KGIVYRDLKL	DNILLDKDGH	IKIADFGMCK	ENMLGDAKTN	TFCGTPDYIA	780
781	PEILLGQKYN	HSVDWWSFGV	LLYEMLIGQS	PFHGQDEEEL	FHSIRMDNPF	YPRWLEKEAK	840
841	DLLVKLVFRE	PEKRLGVRGD	IRQHPLFREI	NWEELERKEI	DPPFRPKVKS	PFDCSNFDKE	900
901	FLNEKPRLSF	ADRALINSMD	QNMFRNFSFM	NPGMERLISH	HHHHH		960

1-218: GST **Red**: HIS6-tag **Pink**: Factor Xa cleavage site **blue**: PKC-theta

PKC-theta wt ¹ Amino Acid Sequence							
1	MSPFLRIGLS	NFDCGSCQSC	QGEAVNPYCA	VLVKEYVESE	NGQMYIQKKP	TMYPWDSTF	60
61	DAHINKGRVM	QIIVKGNVD	LISETTVELY	SLAERCRCNN	GKTEIWLELK	PQGRMLNAR	120
121	YFLEMSDTKD	MNEFETEGFF	ALHQRRGAIK	QAKVHHVKCH	EFTATFFPQP	TFCVSCHEFV	180
181	WGLNKQGYQC	RQCNAAIHKK	CIDKVIKCT	GSAINSRTEM	FHKERFKIDM	PHRFKVYNYK	240
241	SPTFCEHCGT	LLWGLARQGL	KCDACGMNVH	HRCQTKVANL	CGINQKMAE	ALAMIESTQQ	300
301	ARCLRDTEQI	FREGPVEIGL	PCSIKNEARP	PCLPTPGKRE	PQGISWESPL	DEVDKMCHLP	360
361	EPELNKERPS	LQIKLKIEDF	ILHKMLGKGS	FGKVFLAEFK	KTNQFFAIKA	LKKDVVLMDD	420
421	DVECTMVEKR	VLSLAWHEPF	LTHMFCTFQT	KENLFFVMEY	LNGGDLMYHI	QSCHKFDLSR	480
481	ATFYAAEIIIL	GLQFLHSGKI	VYRDLKLDNI	LLDKDGHIKI	ADFGMCKENM	LGDAKTNTFC	540
541	GTPDYIAPEI	LLGQKYNHSV	DWWSFGVLLY	EMLIGQSPFH	GQDEEELFHS	IRMDNPFYPR	600
601	WLEKEAKDLL	VKLVFREPEK	RLGVRGDIRQ	HPLFREINWE	ELERKEIDPP	FRPKVKSPPD	660
661	CSNFDKEFLN	EKPRLSFADR	ALINSMDQNM	FRNFSFMNPG	MERLIS		720

blue: PKC-theta sequence expressed in recombinant protein

¹[NCBI/Protein](https://www.ncbi.nlm.nih.gov/Protein) accession number NP_006248.1

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