

ProQinase™ PDGFR-alpha V561D

platelet-derived growth factor receptor, alpha polypeptide

Recombinant Human Active Protein Kinase

HGNC Symbol: PDGFRA

Synonyms: PDGFR2, CD140a, RHEPDGFRA

Product No.: 1056-0000-1

Lot: 006

Description: Human PDGFR-alpha, C-terminal fragment, amino acids Q₅₅₁-L₁₀₈₉ (as in NCBI/Protein entry NP_006197.1) with a V561D mutation, N-terminal GST-HIS₆ fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

Product identity: PDGFR-alpha V561D Lot 006, was confirmed as PDGFR-alpha by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 89,908 Da

Expression host: Sf9 insect 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: 0.148 µg/µl

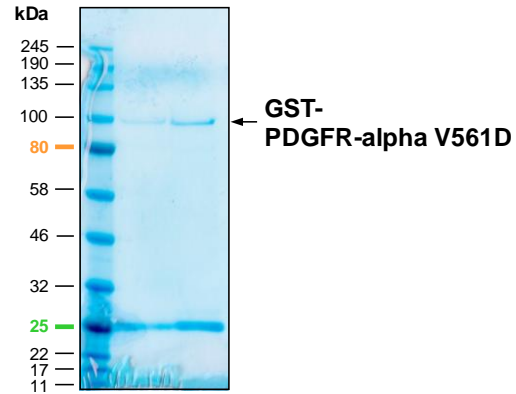
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

Specific kinase activity (P_i transfer): 20 pmol/µg × min

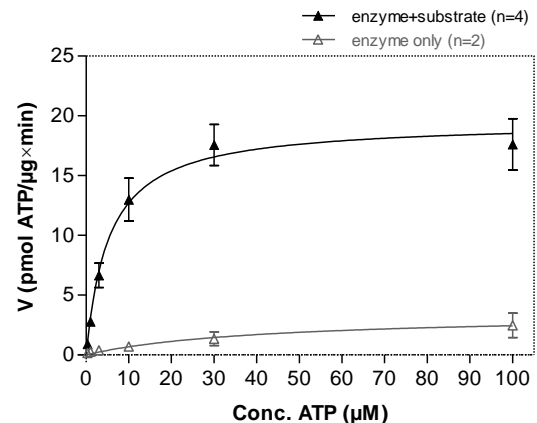
ATP-K_M: 5.3 µM

**PDGFR-alpha V561D LOT006:
Coomassie stain**



1 µg GST-PDGFR-alpha V561D
2 µg GST-PDGFR-alpha V561D

**PDGFR-alpha V561D LOT006:
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: TRK-C-derived peptide, 80 µg / ml
 - PDGFR-alpha V561D: 1.0 µg / ml
- Filter binding assay
 - MSPH membrane (Millipore)

Additional assay technology: PDGFR-alpha V561D LOT006

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



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Recombinant Proteins

GST-PDGFR-alpha V561D Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPOID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMG HHHHHH G	RDS LEVLFGQ	240
241	PLAMGQKPRY	EIRWRDIESI	SPDGHEYIYV	DPMQLPYDSR	WEFPRDGLVL	GRVLGSGAFG	300
301	KVVEGTAYGL	SRSQPVMKVA	VKMLKPTARS	SEKQALMSEL	KIMTHLGPLH	NIVNLLGACT	360
361	KSGPIYIITE	YCFYGDLVNY	LHKNRDSFLS	HHPEKPKKEL	DIFGLNPAD	STRSYVILSF	420
421	ENNGDYMDMK	QADTTQYVPM	LERKEVSKYS	DIQRSLYDRP	ASYKKKSMLE	SEVKNLLSDD	480
481	NSEGLTLLDL	LSFTYQVARG	MEFLASKNCV	HRDLAARNVL	LAQGKIVKIC	DFGLARDIMH	540
541	DSNYVSKGST	FLPVKWMAP	SIFDNLTYTL	SDVWSYGILL	WEIFSLGGTP	YPGMMVDSTF	600
600	YNKIKSGYRM	AKPDHATSEV	YEIMVKCWN	EPEKRPSFYH	LSEIVENLLP	GQYKKSYEKI	660
661	HLDFLKSDFH	AVARMRVDSD	NAYIGVITYKN	EEDKLDWEG	GLDEQRLSAD	SGYIIPLPDI	720
721	DPVPEEEDLG	KRNRHSSQTS	EESALETGSS	SSTFIKREDE	TIEDIDMDD	IGIDSSDLVE	780
781	DSFL						840

1-218: GST **Red**: HIS6-tag **Green**: 3C cleavage site **blue**: PDGFR-alpha fragment **boxed**: V561D mutation

PDGFR-alpha wt ¹ Amino Acid Sequence							
1	MGTSHPAFLV	LGCLLTGLSL	ILCQLSLPSI	LPNENEKVVQ	LNSSFSLRCF	GESEVSWQYP	60
61	MSEEESSDVE	IRNEENNSGL	FVTVLEVSSA	SAAHTGLYTC	YNNHTQTEEN	ELEGRHIYIY	120
121	VPDPDVAFVP	LGMTDYLIVV	EDDDSAIIPC	RTDPETPVT	LHNSEGVPVA	SYDSRQGFNG	180
181	TFTVGPYICE	ATVKGKFKQT	IPFNVYALKA	TSELDLEMEA	LKTVYKSGET	IVVTCAVFNN	240
241	EVVDLQWTYP	GEVKGKGITM	LEEIKVPSIK	LVYTLTVPEA	TVKDSGDYEC	AARQATREVK	300
301	EMKKVTISVH	EKGFIKPT	FSQLEAVNLH	EVKHFVVEVR	AYPPPRISWL	KNNLTLIENL	360
361	TEITTDVEKI	QEIRYRSKPK	LIRAKEEDSG	HYTIVAQNED	AVKSYTFELL	TQVPSILDLD	420
421	VDDHHGSTGG	QTVRCTAEGT	PLPDIEWMIC	KDIKKCNET	SWTILANNVS	NIITEIHSRD	480
481	RSTVEGRVTF	AKVEETIAVR	CLAKNLLGAE	NRELKLVAPT	LRSELTVA	VLVLLVIVII	540
541	SLIVLVVIWK	QKPRYEIRWR	VIESISPDGH	EYIYVDPML	PYDSRWEFPR	DGLVLRVLG	600
600	SGAFGKVEG	TAYGLSRSQP	VMKVAVKMLK	PTARSSEKQA	LMSELKIMTH	LGPLNIVNL	660
661	LGACTKSGPI	YIITEYCFYG	DLVNYLHKNR	DSFLSHHPEK	PKKELDIFGL	NPADSTRSY	720
721	VILSFENNGD	YMDMKQADTT	QYVPLMERKE	VSKYSDIQRS	LYDRPASYKK	KSMLESEVKN	780
781	LLSDDNSEGL	TLLDLLSFTY	QVARGMEFLA	SKNCVHRDLA	ARNVLLAQGK	IVKICDFGLA	840
841	RDIMHDSNYV	SKGSTFLPVK	WMAPEIFDN	LYTTLSDVWS	YGILLWEIFS	LGSTPYPGMM	900
901	VDSTFYNKIK	SGYRMAKPDH	ATSEVYEIMV	KCWNSEPEKR	PSFYHLSEIV	ENLLPGQYKK	960
961	SYEKIHLDFL	KSDHPAVARM	RVSDNAYIG	VTYKNEEDKL	KDWEGLDEQ	RLSADSGYII	1020
1021	PLPDIDPVPE	EEDLGKRNHR	SSQTSEESAI	BTGSSSSTFI	KREDETIEDI	DMDDIGIDS	1080
1081	SDLVEDSFL						1140

blue: PDGFR-alpha sequence expressed in recombinant protein **Red**: variant in recombinant protein

¹NCBI/Protein accession number NP_006197.1