

ProQinase™ FLT3 wt

fms-related tyrosine kinase 3

Recombinant Human Active Protein Kinase

HGNC Symbol: FLT3

Synonyms: CD135, FLK2, STK-1

Product No.: 0776-0000-1

Lot: 016

Description: Human FLT3, C-terminal fragment, amino acids R₅₇₁-S₉₉₃ (as in [NCBI/Protein](#) entry NP_004110.2), N-terminal GST-HIS₆ fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

Product identity: FLT3 wt Lot 011 was confirmed as FLT3 by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 77,198 Da 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: 0.147 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

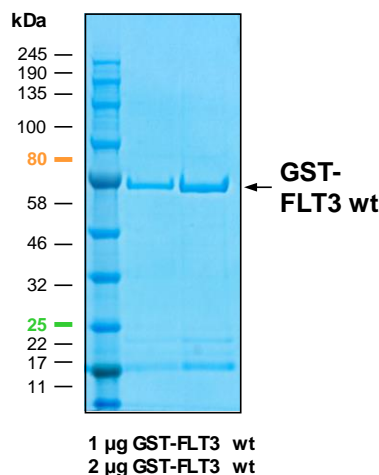
Biochemical Parameters:

Specific kinase activity (P_i transfer): 158 pmol/µg × min
ATP-K_M: 12 µM

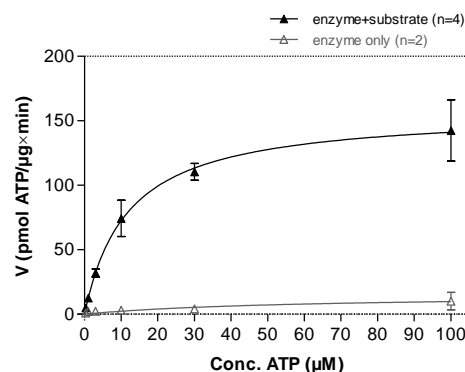
Additional assay technology:

FLT3 wt Lot 016 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

FLT3 wt Lot 016:
Coomassie stain



FLT3 wt Lot 016:
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: Poly(Ala,Glu,Lys,Tyr)_{6:2:5:1}, 20 µg/ml
 - FLT3 wt: 1 µg/ml
- Filter binding assay
- MSFC membrane (Millipore)

ProQinase™ FLT3 wt

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GST-FLT3 wt Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDKVLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPIQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMG HHHHHG	RDS LEVLFQG	240
241	PLAMGRYESQ	LQMVQVTGSS	DNEYFYVDFR	EYEYDLKWEF	PRENLEFGKV	LGSGAFGKVM	300
301	NATAYGISKT	GVSIQVAVKM	LKEKADSSER	EALMSELKMM	TQLGSHENIV	NLLGACTLSG	360
361	PIYLIFEYCC	YGDLLNYLRS	KREKFHRTWT	EIFKEHNFSF	YPTFQSH PNS	SMPGSREVQI	420
421	HPDSDQISGL	HGNSFHSEDE	IEYENQKRLE	EEEDLNVLTF	EDLLCFAYQV	AKGMEFLEFK	480
481	SCVHRDLAAR	NVLVTHGKVV	KICDFGLARD	IMSDSNYVVR	GNARLPVKWM	APESLFEGIY	540
541	TIKSDVWSYG	ILLWEIFSLG	VNPYPGIPVD	ANFYKLIQNG	FKMDQPFYAT	EEIYIIMQSC	600
600	WAFDSRKRPS	FPNLTSFLGC	QLADAEAMY	QNVDRVSEC	PHTYQNRPF	SREMDLGLS	660
661	PQAQVEDS						720

1-218: GST **Red**: HIS6-tag **Green**: 3C cleavage site **blue**: FLT3 fragment

FLT3 wt ¹ Amino Acid Sequence							
1	MPALARDGGQ	LPLLVVFSAM	IFGTITNQDL	PVIKCVLINH	KNNDSSVGKS	SSYPMVSESP	60
61	EDLGCALRPQ	SSGTVYEAAA	VEVDVSASIT	LQVLVDAPGN	ISCLWVFKHS	SLNCQPHFDL	120
121	QNRGVVSMVI	LKMTETQAGE	YLLFIQSEAT	NYTILFTVSI	RNTLLYTLRR	PYFRKMNQD	180
181	ALVCISESVP	EPIVEWVLCD	SQGESCKEES	PAVVKKEEKV	LHELFGTDIR	CCARNELGRE	240
241	CTRLFTIDLN	QTPQTTLPL	FLKVGPELWI	RCKAVHVNHG	FGLTWELENK	ALEEGNYFEM	300
301	STYSTNRTMI	RILFAFVSSV	ARNDTGYTTC	SSSKHPSQSA	LVTIVEKGF	NATNSSEDEYE	360
361	IDQYEEFCFS	VRFKAYPQIR	CTWTFSRKSF	PCEQKGLDNG	YSISKFCNHNK	HQPGEYIFHA	420
421	ENDDAQFTKM	FTLNIRRKPK	VLAEASASQA	SCFSDGYPLP	SWTWKCKSDK	SPNCTEEITE	480
481	GVWNRKANRK	VFGQWVSSST	LMNSEAIKGF	LVKCCAYNSL	GTSCETILLN	SPGPFPIQD	540
541	NISFYATIGV	CLLFIVVLT	LICHKYKKQF	RYESQLQMVQ	VTGSSDNEYF	YVDFREYEYD	600
600	LKWEFPRENL	EFGKVLGSGA	FGKVMNATAY	GISKTGVSIG	VAVKMLKEKA	DSSEREALMS	660
661	ELKMMTQLGS	HENIVNLLGA	CTLSGPIYLI	FEYCCYGDL	NYLRSKREKF	HRTWTEIFKE	720
721	HNFSFYPTFQ	SHPNSSMPGS	REVQIHPDS	QISGLHGNSF	HSEDEIEYEN	QKRLEEEEDL	780
781	NVLTFFEDLLC	FAYQVAKGME	FLEFKSCVHR	DLAARNVLVT	HGKVVKICDF	GLARDIMSDS	840
841	NYVVRGNARL	PVKWMAPESE	FEGIYTIKSD	VWSYGILLWE	IFSLGVNYP	GIPVDANFYK	900
901	LIQNGFKMDQ	PFYATEEIIYI	IMQSCWAFDS	RKRPSFPNLT	SFLGCQLADA	EEAMYQNVGD	960
961	RVSECPHTYQ	NRRPFSREMD	LGLLSPQAQV	EDS			

blue: FLT3 sequence expressed in recombinant protein

¹[NCBI/Protein](#) accession number NP_004110.2