

ProQinase™ FAK (non activated)

focal adhesion kinase

Recombinant Human Active Protein Kinase

HGNC Symbol: PTK2

Synonyms: FADK, FADK1, FAK1, FRNK, pp125FAK

Product No.: 0165-0000-1

Lot: 007

Description: Human FAK, amino acids A₂-H₁₀₅₂ (as in NCBI/Protein entry NP_722560.1), N-terminal GST fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

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

Theoretical MW_{Fusion Protein}: 145,394 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

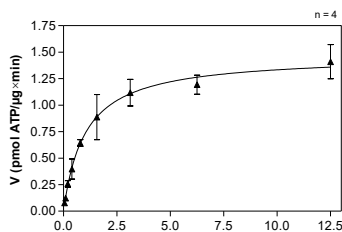
Storage temperature: -80°C

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

Biochemical Parameters:

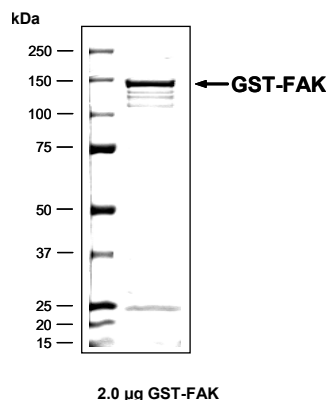
Specific kinase activity (P_i transfer): 1.5 pmol/µg × min
ATP-K_M: 1 µM



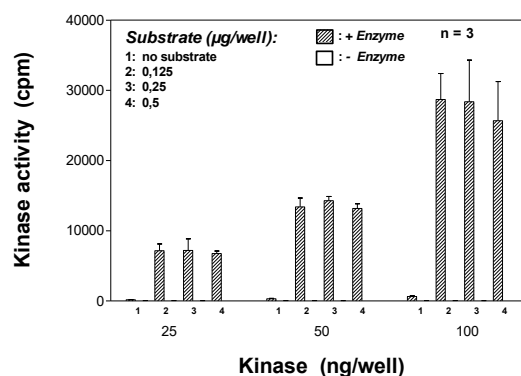
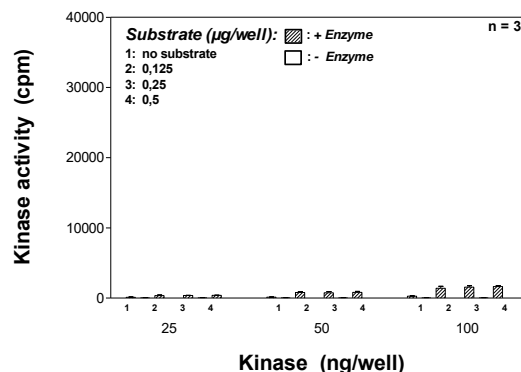
Determination of K_M value & Specific activity:

- 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(Glu:Tyr)_{4,1}, 5 µg/ml
 - Kinase: 4 µg/ml
- Filter binding assay
 - MSFC membrane (Millipore)

FAK Lot 007: Coomassie stain



Kinase activity FAK (non activated) vs. active FAK:



Final assay concentrations:

- 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}
 - 1 µM ATP
 - Substrate (variable): Poly(Glu:Tyr)_{4,1}
 - Recombinant FAK (non activated) or active FAK: 4 µg/ml
- Assay:** ³³PanQinase® Assay

ProQinase™ FAK

Product No.: 0165-0000-1

Recombinant Proteins

GST-FAK Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQ SMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP EML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPQID	KYLKSSKYIA	WPLQG WQATF	GGGDHPPKSD	LVPRGSAAAY	LDPNLNHTPN	240
241	SSTKTHLGTG	MERSPGAMER	VLKVFHYFES	NSEPTTWASI	IRHG DATDVR	GIIQKIVDSH	300
301	KVKHVACYGF	RLSHLRSEEV	HWLHVDMGVS	SVREKYELAH	PPEEWKYELR	IRYLPKGFLN	360
361	QFTEDKPTLN	FFYQQVKS DY	MLEIADQVDQ	EIALKLGCLE	IRRSYWEMRG	NALEKKS NYE	420
421	VLEKDVGLKR	FFPKSLLDSV	KAKTLRKL IQ	QTFRQFANLN	REESILKFFE	ILSPVYRFDK	480
481	ECFKCALGSS	WIISVELAIG	PEEGISY LTD	KGCNPTH LAD	FTQVQTIQYS	NSEDKDRKGM	540
541	LQLKIAGAPE	PLTVTAPSLT	IAENMADLID	GYCRLVNGTS	QSFIIRPQKE	GERALPSI PK	600
601	LANSEKQ GMR	THAVSVSETD	DYAEI IDEED	TYTMPSTRDY	EIQRERIE LG	RCIGEGQFGD	660
661	VHQGIYMSPE	NPALAVA IKT	CKNCTSDSVR	EKFLQEAL TM	RQFDHPHIVK	LIGVITENPV	720
721	WIIMELCTLG	ELRSFLQVRK	YSLDLASLIL	YAYQLSTALA	YLESKRFVHR	DIAARNVLVS	780
781	SNDCVKLGDF	GLSRYMEDST	YKASKGKLP	IKWMAPE SIN	FRRFTSASDV	WMFGVCMWEI	840
841	LMHG VKPFQG	VKNNDVIGRI	ENGERLPMPP	NCPPTLYSLM	TKCWAYDPSR	RPRFTELKAQ	900
901	LSTILEEEKA	QQEERMRES	RRQATVSWDS	GGSD EAPPKP	SRPGYSPRS	SEGFYPSPOH	960
961	MVQTNHYQVS	GYPGSHGITA	MAGSIYPGQA	SLLDQTD SWN	HRPQEIAMWQ	PNVEDSTVLD	1020
1021	LRGIGQVLP T	HLMEERLIRQ	QQEMEEDQRW	LEKEERFLPK	DVRLSRGSI D	REDGSLQPTI	1080
1081	GNQHIYQPVG	KPDPAAPPK K	PPRPGAPGHL	GSLASLSSPA	DSYNEG VKLO	PQEISPPPTA	1140
1141	NLDRSNDKVG	ENVTGLVKAV	IEMSSKIQA P	PPEEYVPMVK	EVGLALRTLL	ATVDETIPLL	1200
1201	PASTHREIEM	AQKLLNSDLG	ELINKMKLAQ	QYVMTSLQQE	YKKQMLTA AH	ALAVDAKNLL	1260
1261	DVIDQARLKM	LGQTRPH					1320

1-218: GST **Pink**: Thrombin cleavage site **blue**: FAK **boxed**: variation from RefSeq

FAK wt ¹ Amino Acid Sequence							
1	MAAYLDPNL	NHTPNSSTKT	HLGTGMERSP	GAMERVLKVF	HYFESNSEPT	TWASIIRHGD	60
61	ATDVRGIIQK	IVDSHKVKHV	ACYGFRLSHL	RSEEVHWLHV	DMGVSSVREK	YELAHPPEEW	120
121	KYELRIRYLP	KGFLNQFTED	KPTLNFFYQQ	VKSDYMLEIA	DQVDQEI ALK	LGCLEIRRSY	180
181	WEMRGNALEK	KSNEYVLEKD	VGLKRFFPKS	LLDSVKAKTL	RKLIQQTFRQ	FANLNREESI	240
241	LKFFEILSPV	YRFDKECFKC	ALGSSWIISV	ELAIGPEEGI	SYLTDKGCNP	THLADFTQVQ	300
301	TIQYSNSEDK	DRKGM LQLKI	AGAPEPLT VT	APSLTIAENM	ADLIDGYCRL	VNGTSQS FII	360
361	RPQKEGERAL	PSIPKLANSE	KQGMRTHAVS	VSETDDYAEI	IDEEDTYTMP	STRDYEIQRE	420
421	RIELGRCIGE	GQFGDVHQGI	YMSPENPALA	VAIKTCKNCT	SDSVREKFLQ	EALTMRFQDH	480
481	PHIVKLVIGVI	TENPVWIIME	LCTLGELRSF	LQVRKYS LDL	ASLILYAYQL	STALAYLESK	540
541	RFVHRDIAAR	NVLVSSNDCV	KLGD FGLSRY	MEDSTYYKAS	KGKLP IKWMA	PESINFRRFT	600
601	SASDVVMFGV	CMWEILMHGV	KPFQGVKNND	VIGRIENGER	LPMPNPCPPT	LYSLMTKCWA	660
661	YDPSRRPRFT	ELKAQLSTIL	EEKAQQEER	MRMESRRQAT	VSWDSSGSD E	APPKPSRPGY	720
721	PSPRSSEGFY	PSPQHMVQTN	HYQVSGYPGS	HGITAMAGSI	YPGQASLLDQ	TDSWNHRPQE	780
781	IAMWQP NVED	STVLDLRGIG	QVLP THLMEE	RLIRQQQE ME	EDQRWLEKEE	RFLKPDVRLS	840
841	RGSIDREDGS	LQPIGNQHI	YQPVGKPDPA	APPKPPRPG	APGHLGSLAS	LSSPADSYNE	900
901	GVKLQPQEIS	PPPTANLDRS	NDKVYENV TG	LVKAVIEMSS	KIQPAPPEEY	VPMVKEVGLA	960
961	LRTLLATVDE	TIPLLPASTH	REIEMAQKLL	NSDLGELINK	MKLAQQYVMT	SLQQEYKQOM	1020
1021	LTAHALAVD	AKNLLDVIDQ	ARLKM LQGTR	PH			1080

blue: FAK sequence expressed in recombinant protein

¹NCBI/Protein accession number NP_722560.1