# Certificate of Analysis

REACTION BIOLOGY

# ProQinase™ VRK1

**VRK serine/threonine kinase 1**

**Recombinant Human Active Protein Kinase**

**HGNC Symbol:** VRK1

**Synonyms:** n/a

**Product No.:** 0302-0000-1

**Lot:** 001

**Description:** Human VRK1, full length, amino acids $M_1$-$K_{396}$ (as in NCBI/Protein entry NP_003375.1), N-terminal GST-$HIS_6$ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

**Product identity:** VRK1 Lot 001, was confirmed as VRK1 by mass spectroscopy LC-ESI-MS/MS

**Theoretical $MW_{Fusion\ Protein}$:** 75,142 Da

**Expression host:** Sf9 insect cells

**Purification:** GST-Affinity Chromatography

**Activation:** This kinase was not activated by special procedures

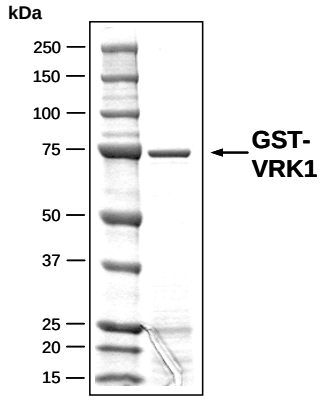
**Storage buffer:** 50 mM TRIS-HCl pH 8.0, 100 mM NaCl, 5 mM DTT, 15 mM reduced glutathione, 20 % glycerol

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

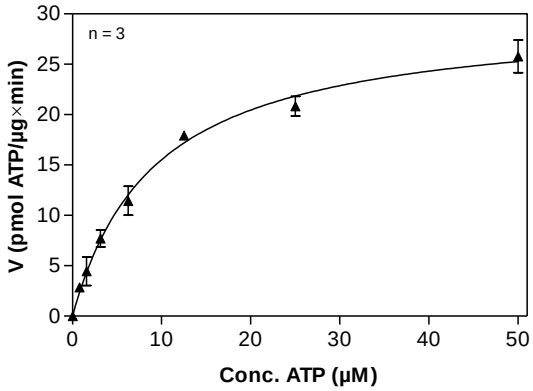
**Biochemical Parameters:**
Specific kinase activity ($P_i$ transfer): 30 pmol/µg × min
ATP-$K_M$: 9.3 µM

**VRK1 Lot 001:**
**Coomassie stain**




**Lot: SP001, 2.0 µg**

**VRK1 Lot 001:**
**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 $MgCl_2$
  3 mM $MnCl_2$
  3 µM Na-orthovanadate
  1.2 mM DTT
  50 µg / ml $PEG_{20.000}$
  ATP (variable)
  Substrate: $R_{11}$-S6-Peptide, 200 µg/ml
  VRK1: 4 µg/ml
- Filter binding assay

MSFC membrane (Millipore)

**Additional assay technology:**
VRK1 Lot 001 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

Recombinant Proteins

# ProQinase™ VRK1

**Product No.:** 0302-0000-1

**GST-VRK1 Recombinant Fusion Protein Amino Acid Sequence**

| | | |
|---|---|---|
| 1 | MSPILGYWKI KGLVQPTRLL LEYLEEKYEE HLYERDEGDK WRNKKFELGL EFPNLPYYID | 60 |
| 61 | GDVKLTQSMA IIRYIADKHN MLGGCPKERA EISMLEGAVL DIRYGVSRIA YSKDFETLKV | 120 |
| 121 | DFLSKLPEML KMFEDRLCHK TYLNGDHVTH PDFMLYDALD VVLYMDPMCL DAFPKLVCFK | 180 |
| 181 | KRIEAIPQID KYLKSSKYIA WPLQGWQATF GGGDHPPKSD PMGHHHHHHG RRRASVAAGI | 240 |
| 241 | LVPRGSPGLD GIYARVKMPR VKAAQAGRQS SAKRHLAEQF AVGEIITDMA KKEWKVGLPI | 300 |
| 301 | GQGGFGCIYL ADMNSSESVG SDAPCVVKVE PSDNGPLFTE LKFYQRAAKP EQIQKWIRTR | 360 |
| 361 | KLKYLGVPKY WGSGLHDKNG KSYRFMIMDR FGSDLQKIYE ANAKRFSRKT VLQLSLRILD | 420 |
| 421 | ILEYIHEHEY VHGDIKASNL LLNYKNPDQV YLVDYGLAYR YCPEGVHKEY KEDPKRCHDG | 480 |
| 481 | TIEFTSIDAH NGVAPSRRGD LEILGYCMIQ WLTGHLPWED NLKDPKYVRD SKIRYRENIA | 540 |
| 541 | SLMDKCFPEK NKPGEIAKYM ETVKLLDYTE KPLYENLRDI LLQGLKAIGS KDDGKLDLSV | 600 |
| 601 | VENGGLKAKT ITKKRKKEIE ESKEPGVEDT EWSNTQTEEA IQTRSRTRKR VQK | 660 |

1-218: GST **Red**: HIS6-tag **Pink**: Thrombin cleavage site **blue**: VRK1

**VRK1 wt[1] Amino Acid Sequence**

| | | |
|---|---|---|
| 1 | MPRVKAAQAG RQSSAKRHLA EQFAVGEIIT DMAKKEWKVG LPIGQGGFGC IYLADMNSSE | 60 |
| 61 | SVGSDAPCVV KVEPSDNGPL FTELKFYQRA AKPEQIQKWI RTRKLKYLGV PKYWGSGLHD | 120 |
| 121 | KNGKSYRFMI MDRFGSDLQK IYEANAKRFS RKTVLQLSLR ILDILEYIHE HEYVHGDIKA | 180 |
| 181 | SNLLLNYKNP DQVYLVDYGL AYRYCPEGVH KEYKEDPKRC HDGTIEFTSI DAHNGVAPSR | 240 |
| 241 | RGDLEILGYC MIQWLTGHLP WEDNLKDPKY VRDSKIRYRE NIASLMDKCF PEKNKPGEIA | 300 |
| 301 | KYMETVKLLD YTEKPLYENL RDILLQGLKA IGSKDDGKLD LSVVENGGLK AKTITKKRKK | 360 |
| 361 | EIEESKEPGV EDTEWSNTQT EEAIQTRSRT RKRVQK | 420 |

**blue**: VRK1 sequence expressed in recombinant protein

[1]NCBI/Protein accession number NP_003375.1

Recombinant Proteins