

## ProKinase™ RET S891A

RET proto-oncogene

Recombinant Human Active Protein Kinase

HGNC Symbol: RET

**Synonyms:** CDHF12, CDHR16, HSCR1, MEN2A, MEN2B, MTC1, PTC, RET51, RET-ELE1

**Product No.:** 1098-0000-1

**Lot:** 006

**Description:** Human RET, C-terminal fragment, amino acids H<sub>658</sub>-S<sub>1114</sub> (as in NCBI/Protein entry NP\_066124.1) with a S891A mutation, N-terminal GST-HIS<sub>6</sub> fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

**Product identity:** RET S891A Lot 006, was confirmed as RET by mass spectroscopy LC-ESI-MS/MS

**Theoretical MW<sub>Fusion Protein</sub>:** 80,420 Da

**Expression:** Baculovirus infected Sf9 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:** 0.187 µg/µl (Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

**Biochemical Parameters:**

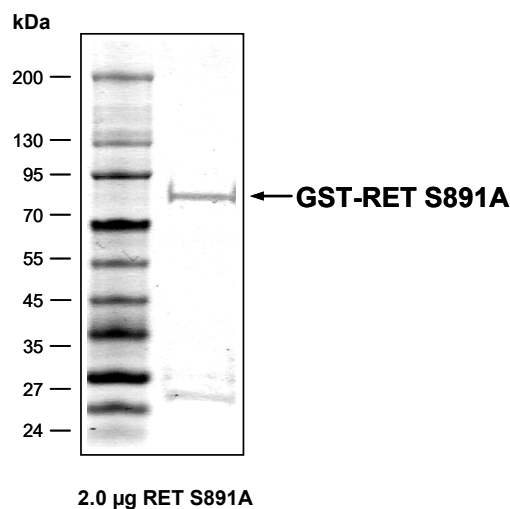
Specific kinase activity (P<sub>i</sub> transfer): 80 pmol/µg×min  
ATP-K<sub>M</sub>: 3.7 µM

**Additional assay technology:** RET S891A Lot 006

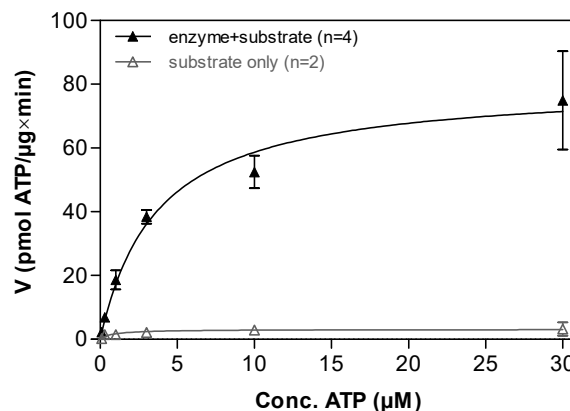
was also successfully tested by Reaction Biology for the use with the ADP-Glo™ Kinase assay from ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details



**RET S891A Lot 006:  
Coomassie stain**



**RET S891A Lot 006:  
Determination of V<sub>max</sub> and K<sub>M</sub> value for ATP**



**Determination of K<sub>M</sub> value & Specific activity:**

- Assay conditions:
  - 60 mM HEPES-NaOH, pH 7.5
  - 3 mM MgCl<sub>2</sub>
  - 3 mM MnCl<sub>2</sub>
  - 3 µM Na-orthovanadate
  - 1.2 mM DTT
  - 50 µg / ml PEG<sub>20,000</sub>
  - ATP (variable)
  - Substrate: TRK-C-derived Peptide (R11-VYSTDYRFLNPS), 80 µg / ml
  - RET S891A: 1.0 µg / ml
- Filter binding assay
  - MSFC membrane (Millipore)

Recombinant Proteins

# ProQinase™ RET S891A

Product No.: 1098-0000-1

## RET S891A Recombinant Fusion Protein Amino Acid Sequence

1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSM	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPPE	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPIQ	KYLKSSKYIA	WPLQGWQATF	GGGDHPKSD	PMGHHHHHG	RDSL <del>EV</del> LFQG	240
241	PLAMGARGR	CYHKFAHKPP	ISSAEMTFR	PAQAFFVSYS	SSGARRPSLD	SMENQVSVDA	300
301	FKILEDPKWE	FPRKNLVLGK	TLGEGEFGKV	VKATAFHLKG	RAGYTTVAVK	MLKENASPSE	360
361	LRDLLSEFNV	LKQVNHPIVI	KLYGACSDQG	PLLLIVEYAK	YGSRLRGFLRE	SRKVGPGYLG	420
421	SGGSRNSSSL	DHPDERALTM	GDLISFAWQI	SQGMQYLAEM	KLVHRDLAAR	NILVAEGRKM	480
481	KIDDFGLSRD	VYEEDSYVKR	SQGRIPVKWM	AIESLFDHIY	TTQSDVWSFG	VLLWEIVTLG	540
541	GNPYPGIPPE	RLFNLKGTGH	RMERPDNCSE	EMYRLMLQCW	KQEPDKRPVF	ADISKDLEKM	600
600	MVKRRDYLDL	AASTPSDSL	YDDGLSEET	PLVDCNNAPL	PRALPSTWIE	NKLYGMSDPN	660
661	WPGESPVPLT	RADGTNTGFP	RYPNDSVYAN	WMLSPSAAKL	MDTFDS		720

1-218: GST Red: HIS6-tag Green: 3C blue:RET fragment A: S891A point mutation

## RET wt<sup>1</sup> amino acid sequence

1	MAKATSGAAG	LRLLLLLLLP	LLGKVALGLY	FSRDAYWEKL	YVDQAAGTPL	LYVHALRDAP	60
61	EEVPSFRLGQ	HLYGTYRTRL	HENNWICIQE	DTGLLYLNRS	LDHSSWEKLS	VRNRGFPLLT	120
121	VYLKVFLSPT	SLREGECQWP	GCARVYFSFF	NTSFPACSSL	KPRELCFPET	RPSFRIRENR	180
181	PPGTFHQFRL	LPVQFLCPNI	SVAYRLLEGE	GLPFRCAPDS	LEVSTRWALD	REQREKYELV	240
241	AVCTVHAGAR	EEVVMVFPFV	TVYDEDDSD	TFPAGVDTAS	AVVEFKRKED	TVVATLRVFD	300
301	ADVVPASGEL	VRRYTSTLLP	GDTWAQQTFR	VEHWPNETSV	QANGSFVRAT	VHDYRLVLNR	360
361	NLSISENRTM	QLAVLVNDS	FQGPAGAVLL	LHFNVSVLPV	SLHLPSTYSL	SVSRRARRFA	420
421	QIGKVCVENC	QAFSGINVQY	KLHSSGANCS	TLGVVTS AED	TSGILFVNDT	KALRRPKCAE	480
481	LHYMVVATDQ	QTSRQAQQL	LVTVEGSYVA	EEAGCPLSCA	VSKRRLECEE	CGGLGSPTGR	540
541	CEWRQGDGKG	ITRNFSTCSP	STKTCPDGHC	DVVETQDINI	CPQDCLRCSI	VGGHEPGEPR	600
600	GIKAGYGTCTN	CFPEEEKCFC	EPEDIQDPLC	DEL CRTVIAA	AVLFSFIVSV	LLSAFCIHCY	660
661	HKFAHKPPIS	SAEMTFRRPA	QAFFVSYS	GARRPSLDSM	ENQVSVDAFK	ILEDPKWEFP	720
721	RKNLVLGKTL	GEGEFGKVVK	ATAFHLKGRA	GYTTVAVKML	KENASPSELR	DLLSEFNVLK	780
781	QVNHPIVIKL	YGACSDGQPL	LLIVEYAKYG	SLRGFLRESR	KVGPGYLGSG	GSRNSSSLDH	840
841	PDERALTMGD	LISFAWQISQ	GMQYLAEMKL	VHRDLAARNI	LVAEGRKMKI	SDFGLSRDVI	900
901	EEDSYVKRSQ	GRIPVKWMAI	ESLFDHIYTT	QSDVWSFGVL	LWEIVTLGGN	PYPGIPPERL	960
961	FNLKLTGHRM	ERPDCNSEEM	YRLMLQCWKQ	EPDKRPVFAD	ISKDLEKMMV	KRRDYLDLAA	1020
1021	STPSDSLIIYD	DGLSEEETPL	VDCNNAPLPR	ALPSTWIENK	LYGMSDPNWP	GESPVPLTRA	1080
1081	DGTNTGFP	PNDVSVYANWM	LSPSAAKLMD	TFDS			1140

blue: RET sequence expressed in fusionprotein Red: variant in fusionprotein

<sup>1</sup>NCBI/Protein accession number NP\_066124.1

Recombinant Proteins