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Certificate of Analysis

Flt1, 10 µg

Recombinant Human FMS-related Tyrosine Kinase 1, Histidine-tagged
Cat Number: BIOTK-Flt1
Lot Number: 3-08

Description:
Recombinant Human protein, Catalytic Domain (amino acids 783-1338), Histidine-tagged, expressed in insect cells. No special measures were taken to activate this kinase.

Specific Activity:
30.8 nmole of phosphate transferred to Poly E_4Y peptide substrate (Sigma, P0275) per minute per mg of total protein at 30°C. Activity determined at a final protein concentration of 4 µg/mL.

Protein Concentration:
0.19 mg/mL total protein as measured using the Bradford protein assay with BSA as a standard.

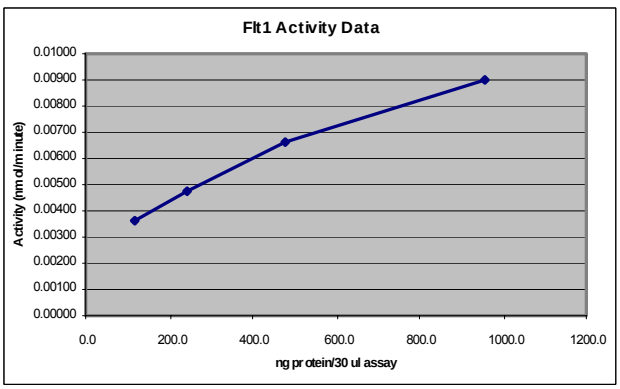
Storage and Handling:
Store at -80°C for long time. At first use, aliquot and to avoid multiple freeze-thaw cycles. If properly stored at -80°C, this product is guaranteed for 12 months from date of purchase. **Protein stable for 6 months at -20°C, for 3-5 h at 4°C and not stable at room temperature.**

Storage Buffer:
50 mM Tris (pH 7.5), 150 mM NaCl, 0.02% Triton® X-100, 2 mM DTT, 50% Glycerol

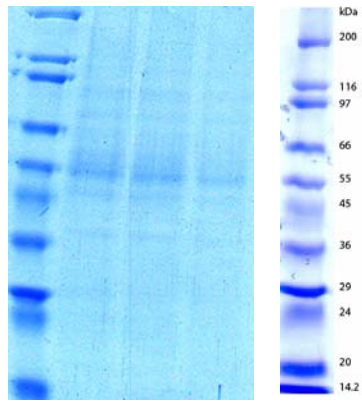
Enzyme Dilution Buffer:
50 mM Tris (pH 7.5), 10% Glycerol, 0.02% Triton® X-100, 0.1 mg/mL BSA, 0.5 mM Na_3VO_4, 2 mM DTT

Quality Assurance

Activity Graph:



PAGE Description:



The SDS-PAGE was run on a 10 well 4-20% Tris-Glycine Novex gel (Catalog #: EC6025BOX).

Lane 1: SigmaMarker, Wide Range, Molecular Weight (Catalog#: S8445, Sigma)
Lane 2: 2.8 µg Flt1
Lane 3: 1.4 µg Flt1
Lane 4: 0.7 µg Flt1

Assay Conditions:
The enzyme was pre-diluted in enzyme dilution buffer and assayed in 50 mM Tris (pH 7.5), 0.01% Tween-20, 5 mM $MgCl_2$, 5 mM $MnCl_2$, 0.5 mM Na_3VO_4, 2 mM DTT, 10 µM ATP, 50 µg Poly E_4Y peptide substrate per reaction and trace [^{32}P]-γ-ATP for 10 minutes at 30°C.

Purity:
≥60% as determined by a Coomassie blue-stained SDS-PAGE gel.

Molecular Weight:
68 kDa calculated from the sequence below.

Protein Sequence:

MSYYHHHHHH DYDIPTTENL YFQGAMGS**MK RSSSEIKTDY LSIIMDPDEV PLDEQCERLP YDASKWEFAR ERLKLGKSLG RGAFGKVVQA SAFGIKKSPT** 100
CRTVAVKMLK EGATASEYKA LMTELKILTH IGHHLNVVNL LGACTKQGGP LMVIVEYCKY GNLSNYLKSK RDLFFLNKDA ALHMEPKKEK MEPGLEQGKK 200
PRLDSVTSSE SFASSGFQED KSLSDVEEEE DSDGFYKEPI TMEDLISYSF QVARGMEFLS SRKCIHRDLA ARNILLSENN VVKICDFGLA RDIYKNPDYV 300
RKGDTRLPLK WMAPESIFDK IYSTKSDVWS YGVLLWEIFS LGGSPYPGVQ MDEDFCSRLR EGMRMRAPEY STPEIYQIML DCWHRDPKER PRFAELVEKL 400
GDLLQANVQQ DGKDYIPINA ILTGNSGFTY STPAFSEDFF KESISAPKFN SGSSDDVRYV NAFKFMSLER IKTFEELLPN ATSMFDDYQG DSSTLLASPM 500
LKRFTWTDSK PKASLKIDLR VTSKSKESGL SDVSRPSFCH SSCGHVSEGK RRFTYDHAEL ERKIACCSPP PDYNSVVLYS TPPISRHAVP SLSRSTRGS. 600

783-1338 amino acids from Flt1 (GenBank Accession Number NP_002010) in bold.