

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of Flag-FancI (Xenopus Laevis)

<u>Enzyme description:-</u>	FLAG-Fanc I (Xenopus (laevis) 2-1321 FL [S557D S560D S566D S597D])
<u>Clone number:-</u>	DU25095
<u>Source:-</u>	Recombinant
<u>Tag:-</u>	N-terminal FLAG
<u>Purification method:-</u>	FLAG-Ab-Sepharose
<u>Expression level:-</u>	0.8mg/L
<u>Calculated molecular mass:-</u>	
Monoisotopic	150760 Da
Average Mass	150851 Da
[cysteines reduced, methionines have not been oxidised]	
<u>Theoretical pI:-</u>	5.86
<u>Purity:-</u>	90%
<u>Enzyme storage buffer:-</u>	
50 mM HEPES pH 7.5, 150mM NaCl, 1mM DTT, 0.1mg/ml FLAG peptide	
<u>Storage temperature:-</u>	-80°C
<u>Assay:-</u>	

Division of Signal Transduction Therapy

Clone Data Sheet

Protein name

Protein FLAG-Fanc I (Xenopus (laevis) 2-1321 FL
[S557D S560D S566D S597D])

Synonyms

Clone Number DU25095

Species Human

Accession Number NP_001164682

Tags N-terminal FLAG

Amino acid sequence of
the expressed protein

MDYKDDDDKD**Q**KILSLAAEE**Q**NDGL**Q**SYL**Q**NLKETELSEIITKHAVKGK
DCGALLRGVFKGSPCSHDVAVRRRLAVYRHCIQLVESGDLQREVASEIM
GLLMLEVHHFPGASLIELANLFADAVKAGTSLNGKSLDLFPTVLTALSS
KESLAYGKGELTGEEFK**Q**LINSLCSSRWDPQSVIFLTSMFRDVSLSAE
ELQFVVEKVLRLMSKLDLQEMPPLVY**Q**LLLLSAKGSKKNIVEGIITIFN
DLD**Q**K**Q**L**V****Q**QENSESLDLEDATIP**Q**D**Q**LHHVEGTIIILHIVFAIKLD**Q**EL
GRELLKYLKAG**Q**QGDSSKILCPFSAALLSVSRIRHF**Q**EQVDFLKSTI
LRDYKDL**Q**FH**Q**SSKFL**Q**DLVPQPVCVSSILLETVKNSVYGWDHVT**Q**GLV
ELGFILMDSFGPKAAFNGKVVELISVHSRTP**Q**QAC**Q**LGSRIILETFKV
HEPIRSEILE**Q**VLNRVITKAATPVTHFIDLLSDIVVSAPLIL**Q**NSSSKV
TEAFDHL**S**FLPLTT**V****Q**GLLKAV**Q**PLLKISMSMRDSLILVLRKAMFSS**Q**I
DARKSAVAGFLLLLRNFKVLGSLSSD**Q**CD**Q**AIGAD**Q**I**Q**VDVHMRYNAAA
NEAFCL**E**ILGSLRRCLD**Q**QADVRLMLYEGFYDVLRRNS**Q**LASSVM**Q**TLL
S**Q**LKRY**Y**EPEDLLPPLKLEGCIT**Q**GDHIFL**Q**EPLAHLCCIHHC**Q**LW
YKSSL**Q**QHRNPDEDDDD**Q**TGC**Q**QDLNDIMESITRM**I**KCDLED**F**ELD
KSAD**F**SLASGVGVKNNIYAVLVMGICEV**L**IEYNFI**I**ANFSKSKFED**I**LG
LFKCCSKLSDILKEKAAG**R**QPGNNKTARSLISMTFVSTLLTALFRDST
RSHEESLSILRANVDFMRY**S**VCVAL**Q**KI**Q**QLEETGVTDGPDG**Q**NSEKMF
RSLCEITRVL**M**WRYTSIPAAENPGKKDKGKTISLLCLEGLLRVFNT**V****Q**
QRYPSKIP**Q**FLTALDALGDEDEEGSREINVTEKAAF**Q**IK**Q**FORS**L**IN**Q**L
SGGEDDFNSKEALLVSILSTLSRLLAPSS**Q**QF**V**QMLSWTVKICKETNI
ED**V****Q**FCKGIMNLLFSLHV**Q**FKSPVSVLREL**Q**DIHGHLGDID**Q**IEVEK
QSHFATVSLKTAAPT**V**TLLVLGQA**A**KVLEEVDWLI**I**IKLKGLLGSEKLST
EDLT**Q**TSNARVPIEK**A**IIL**Q**LGTLLTACHELV**Q**TALPAGSCTDTLLKEL
AKMYTILTSVVKYYL**Q**ICSSHG**Q**ISARLEKLVKLSGSHLTP**Q**CYAFIT
YV**Q**NIHSESLSLAAEKKKKKKEEALTVTS**A**KILRDTKPIPNLIFAIE**Q**Y
EKFLIHLSKSKSVNLM**Q**YMKLSTSRDFRINAATLDAAL**Q**EKGSEDEENE
PDNE**Q**AVTEES**Q**EPKKKRRRKZLE

Native sequence in bold

Protease cleavage n/a

Cloning sites

**DNA sequence of
insert**

ATGGACTACAAAGACGATGACGACAAGGACCAAAAAATCTGTCTTTGG
CTGCTGAAGAACAAAATGATGGACTTCAGAGTTATCTTCAAAACCTGAA
AGAAACCGAGCTTTCAGAAATCATAACAAAGCATGCAGTTAAGGGAAAA
GACTGTGGAGCGCTATTGCGAGGCGTCTTTAAAGGGTCTCCGTGCTCTC
ATGATGTAGCCGTACGCCGAAGGCTCGCGGTTTATAGACACTGCATACA
GCTTGTTGAGTCCGGGGACTTGCAACGAGAGGTTGCATCTGAGATTATG
GGTTTATTAATGCTGGAGGTCCATCATTTTCCGGGTGCTTCGCTCATTG
AACTGGCTAACTTATTTGCGGATGCTGTAAAGCTGGTACTCTGTCCAA
TGGAAAATCTTTAGATTTGTTTCCACAGTACTTACTGCGCTTTCTTCA
AAAGAAAGCTTAGCATATGGTAAAGGTGAAGTACTGGAGAGGAATTTA
AGAAACAAGCTTATAAATAGCCTTTGTTCTAGCAGATGGGATCCACAGAG
TGTGATTTTTCTTACTTCAATGTTTAGGGATGTTTCCCTGAGTGCCGAA
GAGCTGCAGTTTGTGTGGAAAAGGTTTTAAGGATGCTTTCTAAGTTAG
ACCTCCAGGAAATGCCCCCTTAGTTTACCAATTGCTACTCCTGTCTGC
TAAGGGTAGTAAAAAGAATATTGTGGAAGGAATTATCACTATATTTAAT
GATCTTGATCAGAAGCAGTTAGTACAGCAAGAGAACTCTGAATCATTAG
ACTTGGAAGATGCAACCATCCACAAAGACCAGCTGCACCACGTGGAGGG
CACAATCATTTTACACATCGTTTTTGC AATTAAGTTGGACCAGGAATTA
GGAAGAGAGCTTCTGAAGTATCTCAAGGCTGGCCAACAAGGGGACTCAA
GCAAGATCCTTTGCCCTTTCAGTGCTGCCCTCTTGTTATCTGTATCAAG
GATTCATAGATTCCAAGAACAGGTATTTGATTTTCTCAAATCTACTATA
CTGAGAGATTATAAAGACCTTCAGTTTCATCAAAGCTCAAAATTCCTTC
AGGACTTAGTTCCCCAGCCTGTATGCGTATCTTCAATTCCTCTGGAAAC
TGTTAAAAACAGTGTTTATGGATGGGACCATGTCACTCAAGGGCTTG
GAACTTGGTTTTATTTAATGGATTCTTTTGGACCCAAAGCAGCCTTTG
GAAATAAAGTGTTGAGCTTATTTCCGTGCATTCCAGGACACCTTCCCA
GCAAGCTTGTCAGCTTGATCTAGAATCTTGCTAGAAACTTTTAAAGGTC
CATGAGCCTATCAGAAGTGAAATTCCTGGAGCAGGTCTTGAATCGTGTTA
TCACAAAAGCTGCAACTCCAGTTACACATTTTATAGATCTTTTATCAGA
TATTGTGGTTTCTGCTCCGCTCATCCTTCAGAATTCATCATCTAAAGTT
ACTGAAGCATTTGACCACTTGTCCTTCTGCCTCTTACAAGTGTTCAGG
GGCTCCTGAAAGCTGTGCAGCCCCCTTCTTAAATCAGTATGTCCATGAG
GGATTCCCTTATTCTTGTTCTAAGAAAAGCCATGTTCTCAAGTCAAATA
GATGCAAGAAAGTCGGCTGTCGCTGGGTTTCTTCTCCTACTGAGGAACT
TTAAAGTCTTAGGAAGTCTGTCCTCAGATCAGTGCGACCAAGCTATTGG
TGCTGACCAGATCCAGGTGGATGTCCATATGCGTTACAATGCTGCAGCA
AATGAGGCCTTTTGTCTGGAAATCCTTGGCAGTCTGAGGAGGTGCTTAG
ACCAGCAAGCAGATGTTAGGCTCATGCTTTACGAGGGTTTCTATGATGT
CCTGCGACGAAATTTCCAGCTTGCCAGTTCTGTGCATGCAGACTCTGTTG
TCTCAGTTAAAGCGATATTATGAACCCGAACCTGATCTTTTGGCCACCAT
TGAAGTTGGAGGGCTGTATTACTGCTCAAGGGGATCATATATTTTTGCA
GGAACCTCTGGCTCACCTGTTGTGCTGTATCCACCATTGCTTGCAGTGG
TATAAGAGCAGTTTACAGCAGCATAGGAATCCTGATGAAGATGATGACG
ATGATGACCAAACGGGATGTCAACAGGACCTTAATGATATAATGGAGTC
CATCACAAGGAGAATGATTAAATGTGACCTGGAGGACTTTGAATTAGAC
AAGTCTGCAGACTTTTCTTTGGCGTCTGGTGTGGAGTGAAAAATAACA
TATATGCTGTTTTGGTAATGGGGATTTGTGAAGTCCTAATAGAATACAA
CTTTATCATTGCAAATTTTTCAGCAAGAGCAAATTTGAGGACATTTTGGGC
CTGTTTAAAGTGCTGCAGCAAACGTCCGATATCTTAAAGGAGAAAGCTG
CTAAAGGCAGACAGCCTGGGAACAATAAAACGGCCAGAAGCCTCATATC
AATGACTTTTGTCTTACACTCTTAAGTCTTTTATTCAGAGACAGCACA
CGCTCGCACGAGGAGAGTCTTTCTATTCTTCGCGCAAATGTGGATTCA
TGCGCTATTTCGGTGTGTGTGTCCTTCAAAAATTCAGCAGCTTGAAGA
AACTGGTGTACAGATGGACCTGATGGACAGAACAGCGAGAAAATGTTT
CGAAGTCTATGTGAAATTACCAGGGTACTTATGTGGAGATACACCTCAA
TTCCAGCTGCAGCTGAGAATCCTGGGAAGAAGGACAAAGGCCAAAACCAT

TTCCTTGCTGTGCCTGGAGGGATTGCTGAGAGTGTTTAACACCGTTCAG
CAACGATATCCATCAAAGATCCCACAATTCCTTACTGCGTTGGATGCCC
TAGGGGATGAGGATGAAGAAGGCTCAAGAGAAATCAATGTGACCGAGAA
GGCAGCTTTTCAGATTAAACAGTTTCAAAGATCTCTTATTAACCAACTC
AGTGGTGGAGAGGATGACTTTAACAGCAAAGAAGCTCTACTGTTAGTCA
GCATCTTGCTACATTATCCAGGTTACTGGCCCCCTCTTCTCAACAGTT
CGTGCAGATGCTGTCTTGGACTGTTAAAATCTGTAAAGAGACAAACATT
GAGGATGTCCAGTTTTTGCAAGGGAATAATGAACCTTCTCTTTAGCCTCC
ATGTTTCAGTTCAGAGCCCAGTGAGTGTGCTTCGAGAAGTGTGCCAGGA
CATCCATGGGCATCTGGGTGATATTGATCAGGATATTGAAGTGGAGAAG
CAGTCTCATTTTGCTACTGTAAGTCTCAAGACTGCAGCCCCGACTGTTA
CTTTGTTGGTTCTGGGTCAAGCTGCAAAGGTCCTGGAAGAAGTGGACTG
GCTGATCATAAAGCTGAAAGGACTACTGGGTTTCAGAGAAGCTCTCTACG
GAAGATTTAACTCAGACTTCAAATGCAAGGGTTCCAATAGAAAAAGCTA
TCATTCTTCAGTTGGGCACACTGCTGACTGCCTGCCATGAGTTAGTGCA
GACTGCTCTACCTGCTGGGAGTTGTACTGACACTCTTCTGAAGGAGCTG
GCAAAGATGTACACCATCCTCACGTCCGTTGTCAAATACTACTTACAGA
TCTGCAGCAGTCACGGGGGACAGATTTCTGCTCGGTTGGAGAAGCTGGT
GAAGCTTTCGGGTTCTCATCTTACCCCAATGTTATGCCTTCATCACA
TATGTACAGAACATCCACAGTGAAAGTCTAAGCCTTGCTGCAGAAAAGA
AGAAGAAAAAGAAGGAAGAGGCATTGACTGTACCTCAGCCAAAATCCT
GCGTGATACTAAGCCCATACCAAACCTGATCTTTGCCATTGAACAGTAT
GAGAAGTTTCTCATTCATCTCTCCAAGAAGTCCAAGGTGAAGTTGATGC
AATACATGAAGCTGAGCACCTCAAGGGACTTCAGGATAAATGCAGCCAC
ACTAGACGCAGCTTTACAAGAGAAAGGGTCTGAAGATGAAGAGAATGAG
CCAGATAATGAACAGGCTGTAACAGAAGAAGAAAGTCAAGAACCAAAGA
AGAAGAGGCGCAGAAAATAACTCGAG