

Standard Operating Procedure

Preparation of active EPHB4 [561 - 987]

Substrate:-
Poly Glu Tyr (4:1) Final concentration: 0.1 mg/ml

Division of Signal Transduction Therapy

Clone Data Sheet

EPHB4 [561 - 987]

<u>Protein</u>	EPHB4 [561 - 987]
<u>Clone number</u>	DU 9182
<u>Species</u>	Human
<u>Accession number</u>	NM_004444.4
<u>Tags</u>	N-terminal His6
<u>Bacterially expressed protein</u>	MSYYHHHHHDYDIPTTENLYFQGAMGSLCLRKQSNGREAEYSKD HGQYLIGHGTKVYIDPFTYEDPNEAVREFAKEIDVSIVKIEEVIG AGEFGEVCRGLKAPGKKESCVAIKTLKGGYTERQRREFLSEASI MGQFEHPNIIIRLEGVVTNSMPVMILTEFMENGALDSFLRLNDGQF TVIQLVGMLRGIASGMRYLAEMSYVHRDLAARNILVNSNLVCKVS DFGLSRFLEENSSDPTYTSSLGGKIPIRWTAPEAIAFRKFTSASD AWSYGIVMWEVMSFGERPYWDMNQDVINAIEQDYRLPPPPDCPT SLHQLMLDCWQKDRNARPRFPQVVSALDKMIRNPASLKIVARENG GASHPLLDQRPQPHYSAFGSVGEWLRAIKMGRYEESFAAAGFGSFE LVSQISAEDLLRIGVTLAGHQKKILASVQHMKSQAKPGTPGGTGG PAPQY
<u>Native sequence</u>	Amino acids L561 – Y987 (end) of human EPHB4. Residue L29 of the fusion protein is equivalent to L561 of the native enzyme. The His6 tag is located at residues 5 - 10.
<u>Protease cleavage</u>	rTEV (ENLYFQG) residues 18 - 24
<u>Cloning sites</u>	<i>Bam</i> H1 and <i>Not</i> I sites of pFastBac HTb.

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Nucleotide

sequence of insert

ggatcccTCTGCCTCAGGAAGCAGAGCAATGGGAGAGAAGCAGAA
TATTCGGACAAACACGGACAGTATCTCATCGGACATGGTACTAAG
GTCTACATCGACCCCTTCACTTATGAAGACCCTAATGAGGCTGTG
AGGGAATTTGCAAAAGAGATCGATGTCTCCTACGTCAAGATTGAA
GAGGTGATTGGTGCAGGTGAGTTTGGCGAGGTGTGCCGGGGGCGG
CTCAAGGCCCCAGGGAAGAAGGAGAGCTGTGTGGCAATCAAGACC
CTGAAGGGTGGCTACACGGAGCGGCAGCGGCGTGAGTTTCTGAGC
GAGGCCTCCATCATGGGCCAGTTCGAGCACCCCAATATCATCCGC
CTGGAGGGCGTGGTCACCAACAGCATGCCCGTCATGATTCTCACA
GAGTTCATGGAGAACGGCGCCCTGGACTCCTTCCTGCGGCTAAAC
GACGGACAGTTCACAGTCATCCAGCTCGTGGGCATGCTGCGGGGC
ATCGCCTCGGGCATGCGGTACCTTGCCGAGATGAGCTACGTCCAC
CGAGACCTGGCTGCTCGCAACATCCTAGTCAACAGCAACCTCGTC
TGCAAAGTGTCTGACTTTGGCCTTTCCCGATTCTTGAGGAGAAC
TCTTCCGATCCCACCTACACGAGCTCCCTGGGAGGAAAGATTCCC
ATCCGATGGACTGCCCCGGAGGCCATTGCCTTCCGGAAGTTCACT
TCCGCCAGTGATGCCTGGAGTTACGGGATTGTGATGTGGGAGGTG
ATGTCATTTGGGGAGAGGCCGTACTGGGACATGAGCAATCAGGAC
GTGATCAATGCCATTGAACAGGACTACCGGCTGCCCCGCCCCCA
GACTGTCCACCTCCCTCCACCAGCTCATGCTGGACTGTTGGCAG
AAAGACCGGAATGCCCGGCCCCGCTTCCCCCAGGTGGTCAGCGCC
CTGGACAAGATGATCCGGAACCCCGCCAGCCTCAAAATCGTGGCC
CGGGAGAATGGCGGGGCTCACACCCTCTCCTGGACCAGCGGCAG
CCTCACTACTCAGCTTTTGGCTCTGTGGGCGAGTGGCTTCGGGCC
ATCAAAATGGGAAGATACGAAGAAAGTTTCGCAGCCGCTGGCTTT
GGCTCCTTCGAGCTGGTCAGCCAGATCTCTGCTGAGGACCTGCTC
CGAATCGGAGTCACTCTGGCGGGACACCAGAAGAAAATCTTGGCC
AGTGTCCAGCACATGAAGTCCCAGGCCAAGCCGGAACCCCGGGT
GGGACAGGAGGACCGGCCCCGCAGTACTgagcggccgc