

Division of Signal Transduction Therapy

Standard Operating Procedure

Preparation of active SHIP2 [418 - 884]

Enzyme description:-	SHIP2 [418 - 884]
<u>Clone number:-</u>	DU 616
<u>Source:-</u>	Recombinant
<u>Expression system:-</u>	Baculovirus expression vector system
<u>Tag:-</u>	N-terminal His(6)
<u>Purification method:-</u>	Ni ²⁺ -NTA agarose
<u>Expression level:-</u>	3-5 mg/L
<u>Calculated molecular mass:-</u>	56, 281 daltons
<u>Purity:-</u>	80 %
<u>Activation protocol:-</u>	Constitutively active
<u>Enzyme storage buffer:-</u>	
	50 mM HEPES pH 7.5, 10 % glycerol, 150 mM NaCl, 5 mM MgCl ₂
<u>Storage temperature:-</u>	-20 °C
<u>Assay:-</u>	

Currently being configured by Dr Alex Gray.

Please contact Alex directly for further details

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Clone Data Sheet

SHIP2 [418 - 884]

Protein SHIP2 [418 - 884]

Clone Number DU 616

Species Human

Accession number HSY14385

Tags N-terminal His(6)

Baculovirus expressed protein
MHHHHHHDYDIPTTENLYFQ~~Q~~GAMGSQDEPDMISVFIGTWNMGSVPPPKN
VTSWFTSKGLGKTLDEVTVTIPHDIYVFGTQENSVGDREWLDLLRGGLK
ELTDLDYRPIAMQSLWNIKVAVLVKPEHENRISHVSTSSVKTGIANTLG
NKGAVGVSFMFNGTSFGFVNCHLTSGNEKTARRNQNYLDILRLLSLGDR
QLNAFDISLRFTHLFWFGDLNYRLDMDIQEILNYISRKEFEPLLRVDQL
NLEREKHKVFLRFSEEEISFPPTYRYERGSRDYAWHKQKPTGVRTNVP
SWCDRILWKSYPETHIICNSYGCTDDIVTSDHSPVFGTFEVEGVT SQFIS
KKGLSKTSDQAYIEFESIEAIVKTASRTKFFIEFYSTCLEEYKKS FEND
AQSSDNINFLKVQWSSRQLPTLKPILADIEYLQDQHLLLVKSM DGYES
YGECVVALKSMIGSTAQQFLTFLSHRGEETGNIRGSMKVRVPTERL GTR
ER

Native sequence Amino acids Q418 - R884 of human SHIP2.
[Full length protein ends at residue K1258]
Residue Q26 of the fusion protein is equivalent to Q418 of the native enzyme. The His(6) tag is located at residues 2 - 7

Protease cleavage rTEV site (ENLYFQG) is residues 15 – 21

Cloning sites *Bam*HI site of pFastBAC HTb

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**Nucleotide
sequence of insert**

CAGGACGAGCCCGACATGATCTCAGTCTTCATAGGCACCTGGAACATGG
GAAGTGTACCACCTCCAAAAACGTGACATCCTGGTTCACATCGAAGGG
TCTGGGGAAGACCCTGGACGAGGTCACAGTGACCATACCCCATGACATC
TATGTCTTTGGGACCCAGGAGAACTCAGTGGGCGACCGCGAGTGGCTGG
ACCTACTGCGCGGGGGCCTCAAGGAGCTTACGGATCTGGATTACCGCCC
GATTGCCATGCAATCACTGTGGAATATCAAGGTGGCAGTGCTGGTCAAG
CCAGAGCACGAGAACCGTATCAGCCATGTCAGTACGTCCAGTGTGAAGA
CTGGCATCGCCAACACCCTGGGGAACAAGGGGGCTGTGGGCGTCTCCTT
CATGTTTAATGGCACCTCATTGTTGGCTTTGTGAATTGTCACCTCACCTCG
GGAAATGAGAAGACGGCTCGGAGGAACCAAACTACTTGGACATCCTGC
GGCTGCTCTCGCTGGGCGACCGGCAGCTCAATGCCTTTGACATCTCTCT
GCGTTTCACACACCTCTTCTGGTTTGGGACCTCAACTACCGCCTGGAC
ATGGATATCCAGGAGATCCTGAACTACATCAGCAGGAAAGAGTTTGAGC
CCCTCCTCAGGGTGGACCAGCTCAACCTGGAGCGGGAGAAGCACAAAGGT
CTTCCTTCGATTCAGTGAGGAGGAGATCTCCTTCCCACCCACCTACCGC
TATGAGCGGGTTCCCGGACACATATGCCTGGCACAAGCAGAAGCCAA
CTGGGGTCCGGACCAATGTGCCCTCATGGTGTGACCGGATTCTGTGGAA
ATCCTACCCTGAACTCACATCATCTGCAATTCTTATGGTTGCACTGAT
GACATCGTCACCAGCGACCATTCCCCGTGTTTGGGACATTTGAGGTTG
GAGTTACCTCCCAGTTCATCTCCAAGAAAGGGCTCTCAAAGACTTCAGA
CCAGGCCTACATTGAGTTTGAGAGCATCGAGGCCATTGTGAAGACAGCC
AGCCGCACCAAGTTCTTCATCGAGTTCTACTCTACCTGCCTGGAGGAAT
ACAAGAAGAGCTTTGAGAATGATGCCCAGAGCAGTGACAACATCAACTT
CCTCAAAGTGCAGTGGTCTTCACGCCAGCTGCCACGCTCAAACCAATT
CTGGCTGATATCGAGTACCTGCAGGACCAGCACCTCCTGCTCACAGTCA
AGTCCATGGATGGCTATGAATCCTATGGGGAGTGTGTGGTTGCACTCAA
ATCCATGATCGGCAGCACGGCCCAACAGTTCCCTGACCTTCCTATCCCAC
CGTGGCGAGGAGACAGGCAATATCAGAGGCTCCATGAAGGTGCGGGTGC
CCACGGAGCGCTGGGCACCCGTGAGCGG