

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of GST-BAP1

<u>Enzyme description:-</u>	GST-BAP1
<u>Clone number:-</u>	DU12809
<u>Source:-</u>	BL21 Recombinant
<u>Tag:-</u>	N-terminal GST
<u>Purification method:-</u>	GSH sepharose
<u>Expression level:-</u>	2 mg/L

Calculated molecular mass:-

Monoisotopic	107118 Da
Average Mass	107182 Da
[cysteines reduced, methionines have not been oxidised]	

Theoretical pI:- 6.50

Purity:- 70%

Enzyme storage buffer:-

50 mM HEPES pH 7.5, 10% glycerol, 150mM NaCl, 1mM DTT

Storage temperature:- -80°C

Assay:-

Ub-Rho110-Gly cleavage assay monitored by Ex/Em 485/535 nm

Assay buffer:-

40 mM Tris pH 7.5, 100 mM NaCl, 5 mM DTT, 0.01% Triton X-100, 0.005% Ovalbumin, 0.5 µM Ub-Rho110-Gly

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Clone Data Sheet

GST-BAP1

<u>Protein</u>	GST-BAP1
<u>Synonyms</u>	hucep-6
<u>Clone Number</u>	DU12809
<u>Species</u>	Human
<u>Accession Number</u>	Protein: Q92560 DNA: AAH01596.1
<u>Tags</u>	N-terminal GST
<u>Amino acid sequence of expressed protein</u>	<p>MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELGL EFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGAVL DIRYGVSR IAYS KDFETLKVDFLSKLP EMLKMFEDRLCHKTYLNGDHVTH PDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSSKYIA WPLQGWQATFGGGDHPPKSDLEVLVFOGPLGSMNKGWLELESDPGLFTLLV EDFGVKGVQVEEIYDLQSKCQGPVYGFIFLFWIEERRSRKRVSTLVDDT SVIDDDIVNNMFFAHQLIPNSCATHALLSVLLNCSSVDLGPTLSRMKDF T KGFSPEKGYAIGNAPELAKAHNSHARPEPRHLPEKQNGLSAVRTMEAFH FVSYVPITGRLFELDGLKVYPIDHGPWGEDEEWTDKARRVIMERIGLATA GEPYHDIRFNLMAVVPDRRIKYEARLHVLKVNROTIVLEALQQLIRVTQPE LIQTHKSQESQLPEESKSASNKSPLVLEANRAPAASEGNHTDGAEAAAGS CAQAPSHSPPNKPKLVVKKPPGSSLNGVHPNPTPIVQRLPAFLDNHNYAKS PMQEEEDLAAGVGRSRVPVRPPQOYSDEDDYEDDEEDDVQNTNSALRYK GKGTGKPGALSGSADGQLSVLQPNNTINVLAEKLKESQKDLIPLSIKTSS GAGSPAVAVPTHSQPSPTPSNESTDTASEIGSAFNSPLRSPIRSANPTRP SSPVTSHISKVLFGEDDSLRLVDCIRYNRAVRDLGPVISTGLLHLAEDGV LSPLALTEGGKSSPSIRPIQGSQSSSPVEKEVVEATDSREKTGMVRPG EPLSGEKYSPKELLALLKCVEAEIANYEACLKEEVEKRRKFKIDDQRRTH NYDEFICTFISMLAQEGMLANLVEQNISVRRRQGVSIGRLHKQRKPD RRK RSRPYKAKRQ</p>
<u>Native sequence</u>	in bold
<u>Protease cleavage</u>	Precision site underlined
<u>Cloning sites</u>	BamH1 / Not1

DNA sequence of insert

GGATCCATGAATAAGGGCTGGCTGGAGCTGGAGAGCGACCCAGGCCTCTT
CACCCCTGCTCGTGGAAGATTTTCGGTGTCAAGGGGGTGC AAGTGGAGGAGA
TCTACGACCTTCAGAGCAAATGTCAGGGCCCTGTATATGGATTTATCTTC
CTGTTCAAATGGATCGAAGAGCGCCGGTCCCGGCGAAAGGTCTCTACCTT
GGTGGATGATACGTCCGTGATTGATGATGATATTGTGAATAACATGTTCT
TTGCCACCAGCTGATACCCAACCTTTGTGCAACTCATGCCTTGCTGAGC
GTGCTCCTGAACTGCAGCAGCGTGGACCTGGGACCCACCTGAGTCGCAT
GAAGGACTTCACCAAGGGTTTTAGCCCTGAGAGCAAAGGATATGCGATTG
GCAATGCCCCGGAGTTGGCCAAGGCCATAATAGCCATGCCAGGCCCGAG
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CAGTGGTGGCCGACCGCAGGATCAAGTATGAGGCCAGGCTGCATGTGCTG
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GTGGAGAAGGAGTTCGTGGAAGCCACGGACAGCAGAGAGAAGACGGGGAT
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TGCTGGCACTGCTGAAGTGTGTGGAGGCTGAGATTGCAAACATGAGGCG
TGCCCTCAAGGAGGAGGTAGAGAAGAGGAAGAAGTTCAAGATTGATGACCA
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CGGCGGCGCAAGGGGTGAGCATCGGCCGGCTCCACAAGCAGCGGAAGCC
TGACCGGCGGAAACGCTCTCGCCCTACAAGGCCAAGCGCCAGTGAGCGG
CCGC