

*Division of Signal Transduction Therapy*

**Standard Operating Procedure**

**Preparation of active BAP1 [1 – 240]**

**Enzyme description:-** BAP1 [1 - 240]

**Clone number:-** DU 63658

**Source:-** Recombinant

**Expression system:-** *E.coli*

**Tag:-** N-terminal GST

**Purification method:-** GSH Sepharose

**Calculated molecular mass:-**

Monoisotopic 53, 980.44 daltons

Average Mass 54, 015.21 daltons

[cysteines reduced, methionines have not been oxidised]

**Theoretical pI:-** 5.76

**Purity:-** 80 %

**Activation protocol:-** Constitutively active

**Enzyme storage buffer:-**

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

**Storage temperature:-** -70 °C

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**Clone Data Sheet**

**BAP1 [1 - 240]**

<b><u>Protein</u></b>	BAP1 [1 - 240]
<b><u>Clone number</u></b>	DU 63658
<b><u>Species</u></b>	Human
<b><u>Accession number</u></b>	Q92560
<b><u>Tags</u></b>	N-terminal GST
<b><u>Bacterially expressed protein</u></b>	<p>MSPILGYWKIKGLVQPTRLLEYLEEKYEEHLYERDEGDKWRNKKFELG LEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAIEISMLEGA VLDIRYGVSR IAYS KDFETLKVDFLSKLP EMLKMFEDRLCHKTYLNGDH VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSS KYIAWPLQGWQATFGGGDHPPKSDLEVL FQGPLGSMNKGWLELESDPGL <b>F TLLVEDFGVKGVQVEEIYDLQSKCQGPVYGFIFLFWIEERRSRKVS</b> <b>TLVDDTSVIDDDIVNNMFFAHQLIPNSCATHALLSVLLNCSSVDLGPTL</b> <b>SRMKDFTKGFSPESKGYAIGNAPELAKAHNSHARPEPRHLPEKQNGLSA</b> <b>VRTMEAFHFVSYVPITGRLFELDGLKVYPIDHGPGWGEDEEWTDKARRVI</b> <b>MERIGLATAGEPYHDIRFNLMAVVPDRRIK</b></p>
<b><u>Native sequence</u></b>	<p>Amino acids M1 – K240 (end residue is Q729) of human BAP1. Residue M232 of the fusion protein is equivalent to M1 of the native enzyme. The GST tag is located at residues 1 – 220.</p>
<b><u>Protease cleavage</u></b>	PreScission ( <u>LEVLFQGP</u> ) residues 221 - 229
<b><u>Cloning sites</u></b>	<i>Bam</i> H1 and <i>Not</i> 1 sites of pGEX 6P-1

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### Nucleotide Sequence

ATGTCCCCTATACTAGGTTATTGGAAAATTAAGGGCCTTGTGCAACCCA  
CTCGACTTCTTTTGGAAATATCTTGAAGAAAAATATGAAGAGCATTTGTA  
TGAGCGCGATGAAGGTGATAAATGGCGAAACAAAAAGTTTGAATTGGGT  
TTGGAGTTTCCCAATCTTCCTTATTATATTGATGGTGATGTTAAATTAA  
CACAGTCTATGGCCATCATAACGTTATATAGCTGACAAGCACAACATGTT  
GGGTGGTTGTCCAAAAGAGCGTGCAGAGATTTCAATGCTTGAAGGAGCG  
GTTTTGGATATTAGATACGGTGTTCGAGAATTGCATATAGTAAAGACT  
TTGAAACTCTCAAAGTTGATTTTCTTAGCAAGCTACCTGAAATGCTGAA  
AATGTTTGAAGATCGTTTATGTCATAAACATATTTAAATGGTGATCAT  
GTAACCCATCCTGACTTCATGTTGTATGACGCTCTTGATGTTGTTTTAT  
ACATGGACCCAATGTGCCTGGATGCGTTCCCAAATTAGTTTGTTTTAA  
AAAACGTATTGAAGCTATCCACAAATTGATAAGTACTTGAAATCCAGC  
AAGTATATAGCATGGCCTTTGCAGGGCTGGCAAGCCACGTTTGGTGGTG  
GCGACCATCCTCCAAAATCGGATCTGGAAGTTCTGTTCCAGGGGCCCT  
GGGATCCATGAATAAGGGCTGGCTGGAGCTGGAGAGCGACCCAGGCCTC  
TTCACCCTGCTCGTGAAGATTTTCGGTGTCAAGGGGGTGCAAGTGGAGG  
AGATCTACGACCTTCAGAGCAAATGTCAGGGCCCTGTATATGGATTTAT  
CTTCCTGTTCAAATGGATCGAAGAGCGCCGGTCCCGGCGAAAGGTCTCT  
ACCTTGGTGGATGATACGTCCGTGATTGATGATGATATTTGTGAATAACA  
TGTTCTTTGCCCACCAGCTGATACCCAACCTTTGTGCAACTCATGCCTT  
GCTGAGCGTGCTCCTGAACTGCAGCAGCGTGGACCTGGGACCCACCCTG  
AGTCGCATGAAGGACTTCACCAAGGGTTTTAGCCCTGAGAGCAAAGGAT  
ATGCGATTGGCAATGCCCCGGAGTTGGCCAAGGCCATAATAGCCATGC  
CAGGCCCCGAGCCACGCCACCTCCCTGAGAAGCAGAATGGCCTTAGTGCA  
GTGCGGACCATGGAGGCGTTCCACTTTGTCAGCTATGTGCCTATCACAG  
GCCGGCTCTTTGAGCTGGATGGGCTGAAGGTCTACCCATTGACCATGG  
GCCCTGGGGGAGGACGAGGAGTGGACAGACAAGGCCCGGCGGGTCATC  
ATGGAGCGTATCGGCCTCGCCACTGCAGGGGAGCCCTACCACGACATCC  
GCTTCAACCTGATGGCAGTGGTGCCCGACCGCAGGATCAAGtga