

*Division of Signal Transduction Therapy*

**Standard Operation Procedure**

**Preparation of His-Halo-Mud1 UBA domain trimer**

<b><u>Enzyme description:-</u></b>	Mud1 291-332 = UBA domain trimer
<b><u>Clone number:-</u></b>	DU23827
<b><u>Source:-</u></b>	bacteria
<b><u>Tag:-</u></b>	N-terminal His-Halo-
<b><u>Purification method:-</u></b>	Ni <sup>++</sup> -Sephrose
<b><u>Expression level:-</u></b>	15 mg/L
<b><u>Calculated molecular mass:-</u></b>	
Monoisotopic	49734
Average Mass	49736
[cysteines reduced, methionines have not been oxidised]	
<b><u>Theoretical pI:-</u></b>	4.66
<b><u>Purity:-</u></b>	90%
<b><u>Enzyme storage buffer:-</u></b>	
50 mM HEPES pH 7.5, 10% glycerol, 150mM NaCl, 1mM DTT	
<b><u>Storage temperature:-</u></b>	-80°C
<b><u>Assay:-</u></b>	

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### **Clone Data Sheet**

#### **Protein name His-Halo-Mud1 UBA domain trimer**

<b><u>Protein</u></b>	His-Halo-Mud1 UBA 291-332 = UBA domain trimer
<b><u>Synonyms</u></b>	
<b><u>Clone Number</u></b>	DU23827
<b><u>Species</u></b>	Schizosaccharomyces pombe
<b><u>Accession Number</u></b>	Protein: Q10256, NP_001018195.1 DNA:NM_001018619.1
<b><u>Tags</u></b>	N-terminal His-Halo-
Aminoacid sequence of the expressed protein	MGSSHHHHHENLYFQMAEIGTGFPFDPHYVEVLGERMHYVDVGPRDGPV LFLHGNPTSSYVWRNIIPHVAPTHRCIAPDLIGMGKSDKPDLYFFDDHVR FMDAFIEALGLEEVVLVIHDWGSALGFHWAKRNP ERVKGIAFMEFIRPIPT WDEWPEFARETFOAFRTTDDVGRKLIIDQNVFIEGTLPMGVVRPLTEVEMDH YREFFLNPVDREPLWRFPNELPIAGEPANIVALVEEYMDWLHQSPVPKLLF WGTPGVLIPPAEAAARLAKSLPNCKAVDIGPGLNLLQEDNPDIGSEIARWL STLEISGG <b>SGGGPTDPGLNSKIAQLVSMGFDPLEAAQALDAANGDLDAAS</b> <b>FLLGGGRSGGGPTDPGLNSKIAQLVSMGFDPLEAAQALDAANGDLDAASF</b> <b>LLGGGRSPTDPGLNSKIAQLVSMGFDPLEAAQALDAANGDLDAASFLLZA</b> <b>A</b>
Native sequence	in bold
Protease cleavage	TEV site does not work

#### **DNA sequence of the expression cassette**

ATGGGCAGCAGCCATCATCATCATCACGAAAACCTGTATTTTCAGATG  
GCAGAAATCGGTACTGGCTTTCCATTCGACCCCATATGTGGAAGTCCTG  
GGCGAGCGCATGCACTACGTTCGATGTTGGTCCGCGCATGGCACCCCTGTG  
CTGTTCCCTGCACGGTAACCCGACCTCCTCCTACGTGTGGCGCAACATCATC  
CCGCATGTTGCACCGACCCATCGCTGCATTGCTCCAGACCTGATCGGTATG  
GGCAAATCCGACAAACCAGACCTGGGTTATTTCTTCGACGACCACGTCCGC  
TTCATGGATGCCCTTCATCGAAGCCCTGGGTCTGGAAGAGGTCGTCTGGTC  
ATTCACGACTGGGGCTCCGCTCTGGGTTTCCACTGGGCCAAGCGCAATCCA  
GAGCGCGTCAAAGGTATTGCATTTATGGAGTTCATCCGCCCTATCCCGACC  
TGGGACGAATGGCCAGAATTTGCCCGGAGACCTTCCAGGCCTTCCGCACC  
ACCGACGTCGGCCGCAAGCTGATCATCGATCAGAACGTTTTTATCGAGGGT  
ACGCTGCCGATGGGTGTCGTCCGCCCGCTGACTGAAGTCGAGATGGACCAT  
TACCGCGAGCCGTTCCCTGAATCCTGTTGACCGCGAGCCACTGTGGCGCTTC  
CCAAACGAGCTGCCAATCGCCGGTGAGCCAGCGAACATCGTCGCGCTGGTC  
GAAGAATACATGGACTGGCTGCACCAGTCCCTGTCCCGAAGCTGCTGTTC  
TGGGGCACCCAGGCGTTCTGATCCCACCGGCCGAAGCCGCTCGCCTGGCC  
AAAAGCCTGCCTAACTGCAAGGCTGTGGACATCGGCCCGGGTCTGAATCTG  
CTGCAAGAAGACAACCCGGACCTGATCGGCAGCGAGATCGCGCGCTGGCTG  
TCGACGCTCGAGATTTCCGGCGGATCCGGCGGAGGCCCACTGACCCCGGT  
TTGAACTCCAAAATTGCACAGCTAGTCAGTATGGGATTTGATCCATTAGAG  
GCTGCGCAGGCGTTAGACGCTGCGAATGGAGATTTAGATGTAGCTGCTTCT  
TTTCTCCTTGGCGGAGGCAGATCCGGCGGAGGCCCACTGACCCCGGTTTG  
AACTCCAAAATTGCACAGCTAGTCAGTATGGGATTTGATCCATTAGAGCT  
GCGCAGGCGTTAGACGCTGCGAATGGAGATTTAGATGTAGCTGCTTCTTT

CTCCTTGGCGGAGGCAGATCCCCACTGACCCCGGTTTGAACTCCAAAATT  
GCACAGCTAGTCAGTATGGGATTTGATCCATTAGAGGCTGCGCAGGCGTTA  
GACGCTGCGAATGGAGATTTAGATGTAGCTGCTTCTTTTCCTTTAAGCG  
GCCGC