

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of HUWE1 [3670-4374]

Enzyme description:- GST-HUWE1 [3670-4374]

Clone number:- DU43501

Source:- human recombinant

Tag:- N-terminal GST-tag

Purification method:- GSH-Sepharose

Expression system:- E.coli

Calculated molecular mass:-

Monoisotopic 97426 Da

Average Mass 97486 Da

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 5.50

Purity:- 90%

Enzyme storage buffer:-

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

Storage temperature:- -80°C

Assay:-

Division of Signal Transduction Therapy

Clone Data Sheet

GST-HUWE1 [3760-4374]

<u>Protein</u>	GST-HUWE1 (3760-4374) (catalytic domain)
<u>Synonyms</u>	HECT, UBA and WWE domain-containing protein 1; ARF-binding protein 1; ARF-BP1; Homologous to E6AP carboxyl terminus homologous protein 9; HectH9; Large structure of UREB1; LASU1; Mcl-1ubiquitin ligase E3; Mule; Upstream regulatory element-binding protein 1; URE-B1; URE-binding protein1.
<u>Clone Number</u>	DU43501
<u>Species</u>	Human
<u>Accession Number</u>	Protein: Q7Z6Z7, DNA: NM_031407
<u>Tags</u>	N-terminal GST-tag
Aminoacid sequence of the expressed protein	MSPILGYWKIKGLVQPTRLLLEYLEEKYEELHYERDEGDKWRNKKFELGLEF PNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGAVLDIRY GVSRIAYSKDFETLKVDFLSKLPPEMLKMFEDRLCHKTYLNGDHVTHPDFMLY DALDVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSSKYIAWPLQGWQA TFGGGDHPPKSDENLYFOGGSSIQAAVRQLEAEADAIQMVREGQRARRQQQ AATSESSQSEASVRREESPMDVDQPSPSAQDTQSIASDGTPOGEKEKEERPP ELPLLSEQLSLDELWMLGECLKELEESHQHAVLVLOPAVEAFFLVHATER ESKPPVRDTRESQLAHIKDEPPPLSPAPLTPATPSSLDPPFFSREPSSMHISS SLPPDTQKFLRFAETHRTVLNQILRQSTTHLADGPFVAVLDYIRVLDVDFVCR KYFRQELERLDEGLRKEDMAVHVRRDHVFEDSYRELHRKSPEEMKNRLYIVF EGEEGQDAGLLREWYMIISREMFNPMYALFRTSPGDRVITYTINPSSHCPN HLSYFKFVGRIVAKAVYDNRLLECYFTRSFYKHILGKSVRYTDMESDYHFY QGLVYLLENDVSTLGYDLTFSTEVQEFVCEVRDLKPNGANILVTEENKKEY VHLVCQMRMTGAIRKQLAAFLEGFYEIIPKRLISIFTEQELELLISGLPTID IDDLKSNTYHXYQSNSIQIQWFRALRSFDQADRAKFLQFVTGTSKVPLQG FAALEGMNGIQKFQIHRDDRSTDRLPSAHTCFNQLDLPAYESFEKLRHMLLL AIQECSEGFGLA
Native sequence	This sequence covers the HUWE1 C-terminus incl. the catalytic domain in bold. Full length HUWE1 is a 4374 aminoacid protein.
Protease cleavage	TEV protease site underlined
Cloning sites	BamH1 Not1

DNA sequence of insert

ggatccagcatccaggcagctgttcggcagctggaggctgaggctgatgc
cattatacaaagtgtacgtgaggggtcaaagggcgcgagacagcaacaag
cagcaacgtcggagtctagccagtcagagggctctgtccggagggaggaa
tcacccatggatgtggaccagccatctcccagtgctcaagatactcaatc
cattgcctccgatggaacccccacagggggagaaggaaaaggaagaagac
cacctgagttaccctgtctcagcagcagctgagtttgagcagagctgtgg
gacatgcttggggagtgtctaaaggaactagaggaatcccatgaccagca
tgcggtgctagtgtacagcctgtgtcagaggccttctttctgggccatg
ccacagagcgggagagcaagcctcctgtccgagacaccctgtgagagccag
ctggcacacatcaaggacgagcctcctccactctcccctgccccctaac
cccagccacgccttctcctccttgaccattcttctcccgggagccctcat
ctatgcacatctcctcaagcctgccccctgacacacagaagttccttcgc
tttgagagactcaccgactgtgttaaaccagatcctacggcagctccac
gaccaccttgctgatgggccttttgctgtcctggtagactacattcgtg
tcctcgactttgatgtcaagcgcaaatatctccgccaagagctggagcgt
ttagatgaggggctccggaaagaagacatggctgtgcatgtccgctcgtga
ccatgtgtttgaagactcctatcgtgagctgcatcgcaaatccccgaag
aatgaagaatcgattgtatatagtatgtttgaaggagaagaagggcaggat
gctggtgggctcctgcgggagtggtatatgatcatctctcgagagatggt
taaccctatgtatgccttggtccgtacctcacctgggtgatcagtcacct
acaccatcaatccatcttcccactgcaacccccaccacctcagctacttc
aagtttgctggacgcattgtggccaaagctgtatatgacaaccgtcttct
ggagtgtactttactcgatccttttacaacacatcttgggcaagtcag
tcagatatacagatatggagagtgaagattaccacttctaccaaggctcgt
gtttatctgctggaaaatgatgtctccacactaggctatgacctcacctt
cagcactgaggtccaagagtttgagtttggaagttcgtgacctcaaac
ccaatggggccaacatcttggtaacagaggagaataagaaggagtatgta
cacctggtatgccagatgagaatgacaggagccatccgcaagcagttggc
ggcttctttagaaggcttctatgagatcattccaaagcgctcatttcca
tcttcaactgagcaggagttagagctgcttatatcaggactgccaccatt
gacatcgatgatctgaaatccaacactgaataaccacaagtaccagtccaa
ctctattcagatccagtggttctggagagcattgcttcttctcgatcaag
ctgaccgtgccaagttcctccagtttgctcacgggtacttccaaggtacc
ctgcaaggctttgctgcctcgaaggcatgaatggcattcagaagtttca
gatccatcgagatgacagggtccacagatcgctgccttcagctcacacat
gttttaatcagctggatctgcctgcctatgagagctttgagaagctccgc
cacatgctactgttggtatccaggagtgtctgaaggcttgggctggc
ctaagcggccgc