Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of His-Halo-SQSTM UBA domain tetramer

Enzyme description:- SQSTM 389-439 tetramer = UBA domain trimer

Clone number:- DU23922

Source:- bacteria

Tag:- N-terminal His-Halo-

Purification method:- Ni^{++}-Sepharose

Expression level:- 4 mg/L

Calculated molecular mass:-

Monoisotopic	58985
Average Mass	59021

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 5.08

Purity:- 90%

Enzyme storage buffer:-

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

Storage temperature:- -80ºC

Assay:-

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Clone Data Sheet

Protein name His-Halo-SQSTM UBA domain tetramer

Protein	His-Halo-SQSTM UBA 389-439 = UBA domain tetramer
Synonyms	ORCA, OSIL, EBIAP, p62
Clone Number	DU23922
Species	human
Accession Number	Protein: Q13501
Tags	N-terminal His-Halo-
Aminoacid sequence of the expressed protein	MGSSHHHHHHENLYFQMAEIGTGFPFDPHYVEVLGERMHYVDVGPRDGTPVLFLHGNPTSSYVWRNIIPHVAPTHRCIAPDLIGMGKSDKPDLGYFFDDHVRFMDAFIEALGLEEVVLVIHDWGSALGFHWAKRNPERVKGIAFMEFIRPIPTWDEWPEFARETFQAFRTTDVGRKLIIDQNVFIEGTLPMGVVRPLTEVEMDHYREPFLNPVDREPLWRFPNELPIAGEPANIVALVEEYMDWLHQSPVPKLLFWGTPGVLIPPAEAARLAKSLPNCKAVDIGPGLNLLQEDNPDLIGSEIARWLSTLEISGGSGGG**EADPRLIESLSQMLSMGFSDEGGWLTRLLQTKNYDIGAALDTIQYSKHGGGRSGGGEADPRLIESLSQMLSMGFSDEGGWLTRLLQTKNYDIGAALDTIQYSKHGGGRSGGGEADPRLIESLSQMLSMGFSDEGGWLTRLLQTKNYDIGAALDTIQYSKHGGGRSEADPRLIESLSQMLSMGFSDEGGWLTRLLQTKNYDIGAALDTIQYSKH**
Native sequence	in bold
Protease cleavage	TEV site does not work
DNA sequence of the expression cassette	ATGGGCAGCAGCCATCATCATCATCATCACGAAAACCTGTATTTTCAGatggcagaaatcggtactggctttccattcgacccccattatgtggaagtcctgggcgagcgcatgcactacgtcgatgttggtccgcgcgatggcacccctgtgctgttcctgcacggtaacccgacctcctcctacgtgtggcgcaacatcatcccgcatgttgcaccgacccatcgctgcattgctccagacctgatcggtatgggcaaatccgacaaaccagacctgggttatttcttcgacgaccacgtccgcttcatggatgccttcatcgaagccctgggtctggaagaggtcgtcctggtcattcacgactggggctccgctctgggtttccactgggccaagcgcaatccagagcgcgtcaaaggtattgcatttatggagttcatccgccctatcccgacctgggacgaatggccagaatttgcccgcgagaccttccaggccttccgcaccaccgacgtcggccgcaagctgatcatcgatcagaacgttttttatcgagggtacgctgccgatgggtgtcgtccgcccgctgactgaagtcgagatggaccattaccgcgagccgttcctgaatcctgttgaccgcgagccactgtggcgcttcccaaacgagctgccaatcgccggtgagccagcgaacatcgtcgcgctggtcgaagaatacatggactggctgcaccagtcccctgtcccgaagctgctgttctggggcaccccaggcgttctgatcccaccggccgaagccgctcgcctggccaaaagcctgcctaactgcaaggctgtggacatcggcccgggtctgaatctgctgcaagaagacaacccggacctgatcggcagcgagatcgcgcgctggctgtcgacgctcgagatttccggcGGATCCGGCGGAGGCGAGGCTGACCCGCGGCTGATTGAGTCCCTCTCCCAGATGCTGTCCATGGGCTTCTCTGATGAAGGCGGCTGGCTCACCAGGCTCCTGCAGACCAAGAACTATGACATCGGAGCGGCTCTGGACACCATCCAGTATTCAAAGCATggcggaggcAGATCCGGCGGAGGCGAGGCTGACCCGCGGCTGATTGAGTCCC

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TCTCCCAGATGCTGTCCATGGGCTTCTCTGATGAAGGCGGCTGGCTCACC
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CCAGTATTCAAAGCATggcggaggcAGATCCGGCGGAGGCGAGGCTGACC
CGCGGCTGATTGAGTCCCTCTCCCAGATGCTGTCCATGGGCTTCTCTGAT
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ccgc
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