

Division of Signal Transduction Therapy

Standard Operation Procedure

Preparation of USP6 [529 – 1406]

Enzyme description:- USP6 [529 – 1406]

Clone number:- DU37745

Source:- Sf21 Recombinant

Tag:- N-terminal GST tag

Purification method:- GSH sepharose

Expression level:- 4 mg/L

Calculated molecular mass:-

Monoisotopic 125215 Da

Average Mass 125292 Da

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 6.73

Purity:- 90%

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-USP6 [529 – 1406]

<u>Protein</u>	GST-USP6 [529-1406]
<u>Synonyms</u>	UBP6, HRP1, TRE2
<u>Clone Number</u>	DU37745
<u>Species</u>	Human
<u>Accession Number</u>	Protein: P35125 DNA: NM_004505.2
<u>Tags</u>	N-terminal GST tag
<u>Amino acid sequence of expressed protein</u>	<p>MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELGLEFP NLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGAVLDIRYGV SRIAYSKDFETLKVDFLSKLP EMLKMFEDRLCHKTYLNGDHVTHPDFMLYDAL DVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSSKYIAWPLOGWQATFGG GDHPPKSDLEVLFOGPLGSKGATGLSNLGNTCFMNSSIQCVSNTQPLTQYFIS GRHLYELNRTNPIGMKGHMAKCYGDLVQELWSGTQKSVAPLKLRRTI AKYAPK FDGFQQQDSQELLAFLLDGLHEDLNRVHEKPYVELKDS DGRPDWEVAAEAWDN HLRRNRSIIVDLFHGQLRSQVKCKTCGHSVRFDPFNFLSLPLPMDSYMDLEI TVIKLDGTTPVRYGLRLNMDEKYTGLKKQLRDL CGLNSEQILLAEVHDSNIKN FPQDNQKVQLSVSGFLCAFEIPVPSSPISASSPTQIDFSSSPSTNGMFTLTN GDLPKPIFIPNGMPNTVVPCGTEKNFTNGMVNGHMPSLPDSPFTGYIIAVHRK MMRTELYFLSPQENRPSLFGMPLIVPCTVHTRKKDLYDAVWIQVSWLARPLPP QEAS IHAQDRDNCMGYQYPFTLRVVQKDGNSCAWCPOYRFCRGCKIDCGEDRA FIGNAYIAVDWHPTALHLRYQTSQERVVDKHESVEQSRRQAEPINLDSCLRA FTSEEELGESEMYYCSKCKTHCLATKKLDLWRLPPFLIIHLKRFQFVNDQWIK SQKIVRFLRESFDPSAFLVPRDPALCQHKPLTPQDEL SKPRILAREVKKVDA QSSAGKEDMLLSKSPSSLSANISSSPKGPSSSSRKSGTSCPSSKNSSPNSSPR TLGRSKGRLRLPQIGSKNKPSSSKKNLDASKENGAGQICELADALSRGHMRGG SQPELVTPQDHEVALANGFLYEHEACGNGCGDGY SNGQLGNHSEEDSTDDQRE DTHIKPIYNLYAISCHSGILSGGHYITYAKNPCKWYCYNDSSCEELHPDEID TDSAYILFYEQQIDYAOFLPKIDGKKMADTSSTDEDS ESDYEKYSMLQ</p>
<u>Native sequence</u>	in bold
<u>Protease cleavage</u>	Prescission site underlined
<u>Cloning sites</u>	BamH1/Not1

DNA sequence of insert

GGATCCAAGGGAGCCACAGGTCTAAGCAACCTGGGAAACACATGCTTCATGAA
CTCAAGCATCCAGTGCCTTAGTAACACACAGCCACTGCACAGTATTTTATCT
CAGGGAGACATCTTTATGAACTCAACAGGACAAATCCCATTGGTATGAAGGG
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