

# *MRCPPU Reagents and Services*

## Standard Operating Procedure

### Preparation of active USP32 [732 - 1570]

<b><u>Enzyme description:-</u></b>	USP32 [732 - 1570]
<b><u>Clone number:-</u></b>	DU 76095
<b><u>Source:-</u></b>	Recombinant
<b><u>Expression system:-</u></b>	Baculovirus expression vector system
<b><u>Tag:-</u></b>	N-terminal GST
<b><u>Purification method:-</u></b>	GSH Agarose

#### **Calculated molecular mass:-**

Monoisotopic      120, 636.97 daltons  
Average Mass      120, 714.20 daltons  
[cysteines reduced, methionines have not been oxidised]

<b><u>Theoretical pI:-</u></b>	6.36
<b><u>Purity:-</u></b>	>80 %
<b><u>Activation protocol:-</u></b>	Constitutively active

#### **Enzyme storage buffer:-**

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

<b><u>Storage temperature:-</u></b>	-70 °C
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## Clone Data Sheet

USP32 [732 - 1570]

Protein USP32 [732 - 1570]

Clone number DU 76095

Species Human

Accession number Q8NFA0

Tags N-terminal GST

Baculovirus  
expressed protein

MSPILGYWKIKGLVQPTRLLEYLEEKYEEHLYERDEGDKWRNK  
KFELGLEFPNLPYYIDGDVKLTQSMATIRYIADKHNMLGGCPKE  
RAEISMLEGAVLDIRYGVSRRIAYSKDFETLKVDFLSKLPEMLKM  
FEDRLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKL  
VCFKKRIEAIPOIDKYLKSSKYIAWPLQGWQATFGGGDHPKSD  
LEVLFGGPLGS**GATGLSNLGNTCFMNSSIQCVSNTQPLTQYFIS**  
**GRHLYELNRTNPIGMKGHMAKCYGDLVQELWSGTQKNVAPLKLK**  
**WTIAKYAPRFNGFQQQDSQELLAFLLDGLHEDLNRVHEKPYVEL**  
**KDSGRPDWEVAAEAWDNHLRRNRSIVVDLFHGQLRSQVKCKTC**  
**GHISVRFDPFNFLSLPLPMSYMHLEITVIKLDGTTPVRYGLRL**  
**NMDEKYTGLKKQLSDLCLNSEQILLAEVHGSNIKNFQDNQKV**  
**RLSVSGFLCAFEIPVPVSPISASSPTQTFSSSPSTNEMFTLTT**  
**NGDLRPPIFIPNGMPNTVVPCGTEKNFTNGMVNGHMPSLPDSPF**  
**TGYIIAVHRKMMRTELYFLSSQKNRPSLFGMPLIVPCTVHTRKK**  
**DLYDAVWIQVSRLASPLPPQEASNHAQDCDDSMGYQYPFTLRVV**  
**QKDGNSCAWCPWYRFCRGCKIDCGEDRAFIGNAYIAVDWDPTAL**  
**HLRYQTSQERVVDEHESVEQSRRQAEPINLDSCLRAFTSEEL**  
**GENEMYCCKKTHCLATKLDLWRLPPIIHLKRFQFVNGRW**  
**IKSQKIVKFPRESFDPSAFLVPRDPALCQHKPLTPQGDELSEPR**  
**ILAREVKKVDAQSSAGEEDVLLSKSPSSLSANI ISSPKGSPSSS**  
**RKSGTSCPSSKNSSPNSSPRTLGRSKGRLRLPQIGSKNKLSSSK**  
**ENLDASKENGAGQICELADALSRGHVVGGSQPELVTPQDHEVAL**  
**ANGFLYEHEACNGYSNGQLGNHSEEDSTDDQREDTRIKPIYNL**  
**YAISCHSGILGGGHVVTYAKNPNCWKWYCYNDSSCKELHPDEIDT**  
**DSAYILFYEQQGID**

Native sequence Amino acids G732 – D1570 (end residue is Q1604) of human USP32. Residue G232 of the fusion protein is equivalent to G732 of the native enzyme. The GST tag is located at residues 1 - 220.

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**Protease cleavage**

PreScission (LEVLFQGP) residues 221 - 228