

*Division of Signal Transduction Therapy*

**Standard Operation Procedure [G1207S]**

**Preparation of UBE2O 1-1292 (end)**

<b><u>Enzyme description:-</u></b>	UBE2O
<b><u>Clone number:-</u></b>	DU45549
<b><u>Source:-</u></b>	insect recombinant
<b><u>Tag:-</u></b>	N-terminal His
<b><u>Purification method:-</u></b>	Ni <sup>++</sup> -NTA-Sepharose
<b><u>Expression level:-</u></b>	3 mg/L
<b><u>Calculated molecular mass:-</u></b>	
Monoisotopic	145487 Da
Average Mass	145573 Da
[cysteines reduced, methionines have not been oxidised]	
<b><u>Theoretical pI:-</u></b>	4.98
<b><u>Purity:-</u></b>	90%
<b><u>Enzyme storage buffer:-</u></b>	
50mM HEPES pH 7.5, 150mM NaCl, 10% glycerol, 1mM TCEP	
<b><u>Storage temperature:-</u></b>	-80°C
<b><u>Assay:-</u></b>	

## Division of Signal Transduction Therapy

### Clone Data Sheet

#### UBE2O [G1207S]

**Protein** UBE2O 1-1292 (end) [G1207S]  
**Synonyms** E2-230K  
**Clone Number** DU45549  
**Species** Human  
**Accession Number** NM\_022066.3  
GI:192449448 with silent change g6a (to kill BamH1) and coding change g3619a (G1207S) - common variant

**Tags** N-terminal His-

Aminoacid sequence of the expressed protein

MSYYHHHHHDYDIPTTENLYFQGAMGSLEVLFQGPMADPAAPTPAAPAP  
AQAPAPAPEAVPAPAAAPVPAPAPASDSASGPSSDSGPEAGSQRLLFSHD  
LVSGRYRGSVHFGLVRLIHGEDSDSEGESEGRSSGCSEAGGAGHEEGRA  
SPLRRGYVRVQWYPEGVKQHVKETKLEDRSVVPRDVVRHMRSTDSQCG  
TVIDVNIDCAVKLIGTNCIIYPVNSKDLQHIWPFMYGDYIAYDCWLGVY  
DLKNQIILKLSNGARCSMNTEGAKLYDVC PHVSDSGLFFDDSYGFYPGQ  
VLIGPAKIFSSVQWLSGVKPVLS TKSKFRVVVEEVQVVELKVTWITKSF  
CGGTDSVSPPSVITQENLGRVKRLGCFDHAQRQLGERCLYVFPKVEPA  
KIAWECPEKNCAQGE GSMAKKVKRLLKKQVVRIMSCSPDTQCSRDSMED  
PDKKGESKTKSEAESASPEETPDGSASPVEMQDEGAEPEHEAGEQLPFL  
LKEGRDDRLHSAEQDADDEAADD TDSSVTSSASSTSSQSGSGTSRKK  
SIPLSIKNLKRKHKRKNK ITRDFKPGDRVAVEVVTTMTSADVMWQDGSV  
ECNIRSNDLFPVHHLDNNEFCPGDFVVDKRVQSCDP PAVYGVVQSGDHIG  
RTCMVKWFKLRPSGDDVELIGEEEDVSVDIADHPDFRFR TTDIVIRIGN  
TEDGAPHKEDEPSVGQVARVDVSSKVEVWADNSKTIILPQHLYNIESEI  
EESDYDSVEGSTSGASSDEWEDSDSWETDNGLVEDEHPKIEEPP IPPLE  
QPVAPEDKGVVISEEAATAAVQGAVAMAAPMAGLMEKAGKDGP PKSFREL  
KEAIKILESLKNMTVEQLLTGSPTSPTVEPEKPTREKKFLDD IKKLQENL  
KKTLDNVAIVEEEKMEAVPDVERKEDKPEGQSPVKA EWSETPVLCQQCG  
GKPGVTFTSAKGEVFSVLEFAPS NHSFKKIEFQPEAKKFFSTVRKEMAL  
LATS LPEGIMVKTFEDRMDLFSALIKGPTRTPYEDGLYLFDIQLPNIYPA  
VPPHFCYLSQCSGRLNPNLYDNGKVCVSL LGTWIGKTERWTSKSSLLQV  
LISIQGLILVNEPYYNEAGFSDRGLQEGYENSRCYNEMALIRVVQ SMTQ  
LVRRPPEVFEQEIRQHFSTGGWRLVNRIESWLETHALLEKAQALP NGVPK  
ASSPEPPAVAELSDSGQEPEDGGPAPGEASQGS DSEGGAQSLASASRD  
HTDQTSETAPDASVPPSVKPKRRKSYRSFLPEKSGYPDIGFPLF PLSKG  
FIKSIRGVL TQFRAALLEAGMPECTEDK

Native sequence in bold

Protease site Precission Protease underlined

Cloning sites BamH1/NotI

**DNA sequence of the expression cassette**

GGATCCCTGGAAGTTCTGTTCAGGGGCCCATGGCAGATCCCGCAGCC  
CCCACGCCCCGAGCTCCCGCTCCAGCCCAGGCCCGGCTCCAGCCCCG  
GAGGCAGTCCCGGCCCCAGCCGAGCCCCCGTCCCGGCGCCGGCGCCC  
GCCTCGGACTCGGCCCTCCGGGCCGTCTCGGACTCCGGCCCAGAAGCC  
GGCTCGCAGCGCCTGCTGTTTTCTCACGACCTGGTGTGGGCCGTTAC  
CGTGGCTCCGTGCACTTCGGGCTGGTGCGCCTCATCCACGGCGAGGAC  
TCGGACTCGGAGGGCGAGGAGGAGGGCCGCGGGAGCTCGGGGTGCTCC  
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CGCGGCTACGTGCGCGTCCAGTGGTACCCGGAGGGCGTCAAGCAGCAT  
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GCTGACAACCTCAAAGACCATCATCCTGCCCCAGCACTTGTACAACATA  
GAGTCTGAGATTGAGGAGTCAGACTACGATTCGGTAGAAGGCAGCACC  
AGCGGGGCATCCTCGGATGAATGGGAAGATGATAGTGACAGCTGGGAG  
ACGGACAATGGGCTGGTGGAGGACGAGCACCCCAAGATAGAGGAGCCC  
CCATCCCACCCCTGGAGCAGCCGGTGGCCCTGAGGACAAGGGAGTG  
GTGATCAGTGAAGAGGCAGCCACAGCTGCCGTCCAGGGGGCTGTGGCC  
ATGGCTGCCCCCATGGCCGGGCTGATGGAGAAGGCTGGCAAGGACGGG  
CCACCCAAGAGCTTCCGGGAGTTGAAAGAGGCCATCAAGATCCTGGAG  
AGCCTCAAGAACATGACTGTGGAGCAGCTGCTGACGGGCTCGCCCACC  
TCTCCGACTGTGGAGCCTGAGAAGCCAACCTCGGGAGAAGAAGTTTCTG  
GATGACATCAAGAAGCTACAGGAAAACCTCAAGAAGACCCCTGGACAAT  
GTGGCCATTGTAGAGGAGGAGAAGATGGAAGCAGTCCCCGACGTAGAG

CGCAAGGAGGACAAGCCCCGAGGGGCAGTCACCTGTGAAGGCTGAGTGG  
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GCACCCTCAAATCATTCTTTTAAGAAAATTGAGTTCCAGCCTCCAGAA  
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GACCTCTTCTCAGCTCTCATCAAGGGCCCCACTCGAACCCCTACGAG  
GATGGCCTCTACTTGTTTGACATCCAGTCCCCAACATCTACCCAGCC  
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CCCAACCTGTATGACAATGGGAAGGTGTGTGTCAGCCTCCTGGGCACC  
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CAGGTGCTCATCTCCATCCAAGGTCTGATCCTGGTAAATGAACCATAC  
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GGTGTCTGACGCAGTTCCGGGCTGCCCTGCTAGAGGCAGGCATGCCG  
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