

## *Division of Signal Transduction Therapy*

### **Standard Operation Procedure**

#### **Preparation of RNF168**

<b><u>Enzyme description:-</u></b>	GST-RNF168
<b><u>Clone number:-</u></b>	DU19748
<b><u>Source:-</u></b>	Recombinant
<b><u>Tag:-</u></b>	N-terminal GST
<b><u>Purification method:-</u></b>	GSH-Sepharose
<b><u>Expression level:-</u></b>	1mg/L
<b><u>Calculated molecular mass:-</u></b>	
Monoisotopic	93077 Da
Average Mass	93134 Da
[cysteines reduced, methionines have not been oxidised]	
<b><u>Theoretical pI:-</u></b>	7.70
<b><u>Purity:-</u></b>	60 %
<b><u>Enzyme storage buffer:-</u></b>	
50 mM HEPES pH 7.5, 10% glycerol, 150mM NaCl, 1mM DTT	
<b><u>Storage temperature:-</u></b>	-80°C
<b><u>Assay:-</u></b>	

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**Clone Data Sheet**

**Protein name RNF168**

**Protein** GST-RNF8 1 - 571 (full length)

**Synonyms**

**Clone Number** DU19748

**Species** Human

**Accession Number** Protein: Q8IYX5

**Tags** N-terminal GST

Aminoacid sequence  
of the expressed  
protein

MSPILGYWKIKGLVQPTRLLEYLEEKYEEHLYERDEGDKWRNKKFELGL  
EFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGAVL  
DIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDHVTH  
PDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSSKYIA  
WPLQGWQATFGGGDHPPKSDLEVLFOGPLGSPEFPGRLERPL**MALPKDAI**  
**PSLSECQCGICMEILVEPVTLPCNHTLCKPCFQSTVEKASLCCPFCRRRV**  
**SSWTRYHTRRNSLVNVELWTIQKHYPRECKLRASGOESEEVADDYQVVR**  
**LLSKPGELRREYEEEISKVAAERRASEEEENKASEEYIQRLLAEEEEEEK**  
**RQAEKRRRAMEEQKLSDEELARKLSIDINNFCEGSISASPLNSRKSDPVT**  
**PKSEKKSKNKQRTGDIQKYLTPKSQFGSASHSEAVQEVKDSVSKDIDS**  
**SDRKSPTGQDTEIEDMPTLSPQISLGVGEQADSSIESPMPWLCACGAEW**  
**YHEGNVKTRPSNHGKELCVLSHERPKTRVPYSKETAVMPCGRTEGSCAPT**  
**SGVTQTNGNNTGETENEESCLLSKEISKRKNQESSFEAVKDPCFSAKRR**  
**KVSPESPQDEETEINFQKLIDLEHLLFERHKQEEQDRLLALQLOKEVD**  
**KEQMVPNRQKQSPDEYHLRATSSPPDKVLNGQRKNPKDGNFKRQTHTKHP**  
**TPERGSRDKNRQVSLKMQLKQSVNRRKMPNSTRDHCKVSKSAHSLQPSIS**  
**QKSVFQMFQRCTK**

Native sequence in bold

Protease cleavage Precission site underlined

Cloning sites Not1

**DNA sequence of  
insert**

GCGGCCGCTCATGGCTCTACCCAAAGACGCCATCCCCTCGCTGTCCGAGTG  
CCAGTGCGGGATCTGCATGGAAATCCTCGTGGAGCCCCTCACCCCTCCCGTG  
TAACCACACGCTGTGTAAACCGTGCTTCCAGTCGACCCGTCGAAAAGGCGAG  
TTTATGCTGTCCCTTCTGTGCGCCGCGGGTATCGTCGTGGACTCGGTACCA  
TACCCGAAGAAATCTCTCGTCAACGTGGAACGTGGACGATAATTCAAAA  
ACACTATCCCAGGGAGTGCAAGCTTAGAGCGTCTGGCCAAGAATCAGAGGA  
AGTGGCTGATGACTATCAGCCAGTTCGTCTGCTCAGTAAACCTGGGGAAC  
GAGAAGAGAATATGAAGAGGAAATAAGCAAGGTGGCGGCAGAGCGACGGGC  
CAGCGAGGAAGAAGAAAACAAAGCCAGTGAAGAATACATACAGAGGTTGTT  
GGCAGAGGAGGAAGAAGAGGAAAAAAGACAGGCAGAAAAAAGGCCAAGAGC  
GATGGAAGAACAAC TGAAAAGTGATGAGGAAC TGGCAAGAAAGCTAAGCAT  
TGATATTAACAATTTCTGTGAGGGAAGTATCTCGGCTTCTCCCTTGAATTC  
CAGAAAATCTGATCCAGTTACACCCAAGTCTGAAAAGAAAAGTAAGAACAA  
ACAAAGAAACACTGGAGATATTCAGAAGTATTTGACACCGAAATCTCAGTT  
TGGGTCAGCCTCACACTCTGAAGCTGTACAAGAAGTCAGGAAAGACTCCGT  
ATCTAAGGACATTGACAGTAGTGATAGGAAAAGCCCAACAGGGCAAGACAC  
AGAAATAGAAGATATGCCGACACTTTCTCCACAGATATCCCTTGGAGTTGG  
AGAACAAGGTGCAGATTCTTCAATAGAGTCCCCTATGCCATGGTTATGTGC  
CTGTGGTGCCGAATGGTACCATGAAGGAAACGTCAAAAACAAGACCAAGCAA  
TCATGGGAAAGAGTTATGTGTCTTAAGTCACGAGCGACCTAAAACCAGAGT  
TCCCTACTCGAAAGAAACTGCAGTTATGCCTTGTGGCAGAACAGAAAGTGG  
GTGCGCCCCCACATCAGGGGTGACACAGACAAATGGAAACAACACAGGTGA  
GACAGAAAATGAAGAGTCGTGCCTACTGATCAGTAAGGAGATTTCCAAAAG  
AAAAAACCAAGAATCTTCCTTTGAAGCAGTCAAGGATCCATGCTTTTCTGC  
AAAAAGAAGAAAAGTGTCCCCGAATCTTCCCAGATCAAGAGGAAACAGA  
AATAAACTTTACCCAAAAACTGATAGATTTGGAGCATCTACTGTTTGAGAG  
ACATAAACAAGAAGAACAGGACAGGTTATTGGCATTACAAC TTCAGAAGGA  
GGTGGATAAAGAGCAAATGGTGCCAAACCGGCAAAAAGGATCCCCAGATGA  
GTATCACTTACGCGCTACATCCTCCCCTCCAGACAAAGTGCTAAATGGACA  
GAGGAAGAATCCCAAAGATGGGAAC TTCAAAAGGCAAACTCACACAAAGCA  
TCCAACACCAGAGAGAGGCTCAAGGGACAAAAATAGGCAAGTGTCTTTAAA  
GATGCAGTTGAAGCAGTCAGTTAATAGAAGAAAGATGCCAAATCTACTAG  
AGATCACTGTAAGGTATCCAAAAGTGCTCACTCCCTACAGCCTAGCATTTT  
ACAGAAAAGTGT TTTTTCAGATGTTTTCAGAGATGCACAAAAGTAAGCGGCCG