## *Division of Signal Tranduction Therapy*

# Standard Operating Procedure

# Preparation of HSP27 [2 - 205]

**Protein description:-** HSP27 [2 - 205]

**Clone number:-** DU 808

**Source:-** Recombinant

**Expression system:-** *E.coli*

**Tag:-** N-terminal GST

**Purification method:-** GSH Sepharose

**Expression level:-** 2 mg/L

**Calculated molecular mass:-** 49, 444 daltons

**Purity:-** 90 %

**Enzyme storage buffer:-**

50 mM Tris-HCl pH 7.5, 270 mM Sucrose, 150 mM NaCl, 0.1 mM EGTA,
0.1 % 2-mercaptoethanol, 0.02 % Brij-35, 0.2 mM PMSF, 1 mM Benzamidine.

**Storage temperature:-** –70 °C

**Assay:-** Substrate for MAPKAP-K2

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# CLONE DATA SHEET

## HSP27 [2 - 205]

| | |
|---|---|
| **Protein** | HSP27 [2 - 205] |
| **Clone number** | DU 808 |
| **Species** | Human |
| **Accession no** | NM_001540 |
| **Tags** | N-terminal GST |
| **Bacterially expressed protein** | MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKW<br>RNKKFELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNML<br>GGCPKERAEISMLEGAVLDIRYGVSRIAYSKDFETLKVDFL<br>SKLPEMLKMFEDRLCHKTYLNGDHVTHPDFMLYDALDVVLY<br>MDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSSKYIAWPLQG<br>WQATFGGGDHPPKSDLEVLFQGPLGS**TERRVPFSLLRGPSW**<br>**DPFRDWYPHSRLFDQAFGLPRLPEEWSQWLGGSSWPGYVRP**<br>**LPPAAIESPAVAAPAYSRALSRQLSSGVSEIRHTADRWRVS**<br>**LDVNHFAPDELTVKTKDGVVEITGKHEERQDEHGYISRCFT**<br>**RKYTLPPGVDPTQVSSSLSPEGTLTVEAPMPKLATQSNEIT**<br>**IPVTFESRAQLGGPEAAKSDETAAK** |
| **Native sequence** | Amino acids T2 – K205 (end) of human HSP27.<br>Residue T232 of the fusion protein is equivalent to T2 of the native protein. The GST tag is located at residues 1 - 220 |
| **Protease cleavage** | PreScission (LEVLFQGPL) residues 221 - 229 |
| **Cloning sites** | *Bam*H1 and *Eco*R1 of pGEX-6P-1 |

| | |
|---|---|
| **Nucleotide sequence of insert** | GGATCCACCGAGCGCCGCGTCCCCTTCTCGCTCCTGCGGGG<br>CCCCAGCTGGGACCCCTTCCGCGACTGGTACCCGCATAGCC<br>GCCTCTTCGACCAGGCCTTCGGGCTGCCCCGGCTGCCGGAG<br>GAGTGGTCGCAGTGGTTAGGCGGCAGCAGCTGGCCAGGCTA<br>CGTGCGCCCCCTGCCCCCCGCCGCCATCGAGAGCCCCGCAG<br>TGGCCGCGCCCGCCTACAGCCGCGCGCTCAGCCGGCAACTC<br>AGCAGCGGGGTCTCGGAGATCCGGCACACTGCGGACCGCTG<br>GCGCGTGTCCCTGGATGTCAACCACTTCGCCCCGGACGAGC<br>TGACGGTCAAGACCAAGGATGGCGTGGTGGAGATCACCGGC<br>AAGCACGAGGAGCGGCAGGACGAGCATGGCTACATCTCCCG<br>GTGCTTCACGCGGAAATACACGCTGCCCCCCGGTGTGGACC<br>CCACCCAAGTTTCCTCCTCCCTGTCCCCTGAGGGCACACTG<br>ACCGTGGAGGCCCCCATGCCCAAGCTAGCCACGCAGTCCAA<br>CGAGATCACCATCCCAGTCACCTTCGAGTCGCGGGCCCAGC<br>TTGGGGGCCCAGAAGCTGCAAAATCCGATGAGACTGCCGCC<br>AAGtaagaattc |