*Division of Signal Tranduction Therapy*

**Standard Operating Procedure**

**Preparation of Annexin A1 [1 – 346]**

**Enzyme description:-**     Annexin A1 [1 – 346]

**Clone number:-**      DU 38293

**Source:-**        Recombinant

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

**Tag:-**          N-terminal GST

**Purification  method**:-    GSH Sepharose

**Calculated molecular mass:-**
Monoisotopic   65, 496.62 daltons
Average Mass   65, 538.41 daltons
[cysteines reduced, methionines have not been oxidised

**Theoretical pI:-**      6.09

**Purity:-**        >80 %

**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, 1 mM benzamidine, 0.2 mM PMSF

**Storage temperature:-**   -70 °C

# Clone Data Sheet

## Annexin A1 [1 – 346]

**Protein**    Annexin A1 [1 – 346]

**Clone number**    DU 38293

**Species**    Human

**Accession number**    NM_000700.2

**Tags**    N-terminal GST

**Bacterially expressed protein**

```
MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELG
LEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGA
VLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDH
VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSS
KYIAWPLQGWQATFGGGDHPPKSDLEVLFQGPLGSMAMVSEFLKQAWFI
ENEEQEYVQTVKSSKGGPGSAVSPYPTFNPSSDVAALHKAIMVKGVDEA
TIIDILTKRNNAQRQQIKAAYLQETGKPLDETLKKALTGHLEEVVLALL
KTPAQFDADELRAAMKGLGTDEDTLIEILASRTNKEIRDINRVYREELK
RDLAKDITSDTSGDFRNALLSLAKGDRSEDFGVNEDLADSDARALYEAG
ERRKGTDVNVFNTILTTRSYPQLRRVFQKYTKYSKHDMNKVLDLELKGD
IEKCLTAIVKCATSKPAFFAEKLHQAMKGVGTRHKALIRIMVSRSEIDM
NDIKAFYQKMYGISLCQAILDETKGDYEKILVALCGGN
```

**Native sequence**    Amino acids M1 – N346(end) of human Annexin A1.
Residue M232 of the fusion protein is equivalent to M1 of the native enzyme.  The GST tag is located at residues 1 – 220.

**Protease cleavage**    PreScission (LEVLFQGP) residues 221 - 228

**Cloning sites**    *Bam*H1 and *Not*1 sites of pGEX6P1

# Division of Signal Tranduction Therapy

**Nucleotide Sequence of insert**

```
ggatccATGGCAATGGTATCAGAATTCCTCAAGCAGGCCTGGTTTATTG
AAAATGAAGAGCAGGAATATGTTCAAACTGTGAAGTCATCCAAAGGTGG
TCCCGGATCAGCGGTGAGCCCCTATCCTACCTTCAATCCATCCTCGGAT
GTCGCTGCCTTGCATAAGGCCATAATGGTTAAAGGTGTGGATGAAGCAA
CCATCATTGACATTCTAACTAAGCGAAACAATGCACAGCGTCAACAGAT
CAAAGCAGCATATCTCCAGGAAACAGGAAAGCCCCTGGATGAAACACTG
AAGAAAGCCCTTACAGGTCACCTTGAGGAGGTTGTTTTAGCTCTGCTAA
AAACTCCAGCGCAATTTGATGCTGATGAACTTCGTGCTGCCATGAAGGG
CCTTGGAACTGATGAAGATACTCTAATTGAGATTTTGGCATCAAGAACT
AACAAAGAAATCAGAGACATTAACAGGGTCTACAGAGAGGAACTGAAGA
GAGATCTGGCCAAAGACATAACCTCAGACACATCTGGAGATTTTCGGAA
CGCTTTGCTTTCTCTTGCTAAGGGTGACCGATCTGAGGACTTTGGTGTG
AATGAAGACTTGGCTGATTCAGATGCCAGGGCCTTGTATGAAGCAGGAG
AAAGGAGAAAGGGGACAGACGTAAACGTGTTCAATACCATCCTTACCAC
CAGAAGCTATCCACAACTTCGCAGAGTGTTTCAGAAATACACCAAGTAC
AGTAAGCATGACATGAACAAAGTTCTGGACCTGGAGTTGAAAGGTGACA
TTGAGAAATGCCTCACAGCTATCGTGAAGTGCGCCACAAGCAAACCAGC
TTTCTTTGCAGAGAAGCTTCATCAAGCCATGAAAGGTGTTGGAACTCGC
CATAAGGCATTGATCAGGATTATGGTTTCCCGTTCTGAAATTGACATGA
ATGATATCAAAGCATTCTATCAGAAGATGTATGGTATCTCCCTTTGCCA
AGCCATCCTGGATGAAACCAAAGGAGATTATGAGAAAATCCTGGTGGCT
CTTTGTGGAGGAAACtaagcggccgc
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