

Division of Signal Transduction Therapy

Standard Operating Procedure

Preparation of PALM [1 - 387]

Enzyme description:- PALM [1 - 387]

Clone number:- DU 25814

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Sepharose

Calculated molecular mass:-

Monoisotopic 69, 782.26 daltons

Average Mass 69, 826.15 daltons

[cysteines reduced, methionines have not been oxidised]

Theoretical pI:- 5.11

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

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Clone Data Sheet

PALM [1 - 387]

Protein PALM [1 – 387]

Clone number DU 25814

Species Human

Accession number NM_002579.2

Tags N-terminal GST

Bacterially expressed protein

MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELG
LEFPNLPYYIDGDVKLTSMAIIRYIADKHNMLGGCPKERAIEISMLEGA
VLDIRYGVSR IAYS KDFETLKVDFLSKLP EMLKMFEDRLCHKTYLNGDH
VTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPOIDKYLKSS
KYIAWPLQGWQATFGGGDHPPKSDLEVLFGQPLGSPEFPGRLE**MEVLAA**
ETTSQOERLQAIAEKRKRQAEIENKRRQLEDERRQLQHLKSKALRERWL
LEGTPSSASEGDEDLRRQMQDDEQKTRLLEDVSRLEKEIEVLERGDSA
PATAKENAAAPSPVRAPAPSPAKEERKTEVVMNSQOTPVGTPKDKRVSN
TPLRTVDGSPMMKAAMYSVEITVEKDKVTGETRVLSSTLLPRQPLPLG
IKVYEDETKVVHAVDGTAENGIHPLSSSEVDEL I HKADEVTLSEAGSTA
GAAETRGAVEGAARTTPSRREITGVQAQPGEATSGPPGIQPGQEPVMT
IFMGYQVEDEAE**TKKVLGLQDTITAE**LVVIEDAAEPKEPAPPNGSAAE
PPTEAASREENQAGPEATTSDPQDLDMKKHRCKCCS**IM**

Native sequence Amino acids M1 – M387 (end) of human PALM.
Residue M240 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 *Xho*1 and *Not*1 sites of pGEX6P-1

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Nucleotide Sequence of insert

ctcgagATGGAGGTCCTGGCGGCAGAGACCACGTCCCAGCAGGAGCGGCT
GCAGGCCATCGCAGAGAAGCGGAAGCGGCAGGCGGAGATCGAGAACAAGC
GCCGGCAGCTGGAGGACGAGCGGAGGCAGCTGCAGCACCTGAAGTCCAAG
GCACTGCGGGAGCGCTGGCTGCTGGAGGGGACGCCGTCCTCGGCCTCAGA
GGGGATGAGGACCTGAGGAGGCAGATGCAGGACGACGAGCAGAAGACAC
GGCTGCTGGAGGACTCGGTGTCCAGGTTGGAGAAGGAAATTGAGGTGCTG
GAGCGTGGAGACTCCGCCCCAGCCACTGCCAAGGAGAACGCGGCGGCCCC
GAGCCCAGTCCGGGCCCCAGCCCCGAGTCCAGCCAAGGAGGAGCGCAAGA
CAGAGGTGGTGATGAATTCACAGCAGACGCCGGTGGGCACGCCAAAGAC
AAGCGAGTCTCCAACACGCCCTGAGGACGGTTGACGGCTCCCCATGAT
GAAGGCAGCCATGTACTCGGTTGAGATCACTGTGGAGAAGGACAAGGTGA
CAGGGGAGACCAGGGTGCTGTCCAGCACCACGCTGCTCCCTCGGCAGCCG
CTCCCTCTGGGCATCAAAGTCTACGAGGACGAGACCAAAGTGGTCCATGC
TGTGGACGGCACCGCCGAGAACGGGATCCACCCCCTGAGCTCCTCCGAGG
TGGACGAACTCATCCACAAAGCGGACGAGGTCACGCTGAGCGAGGCAGGG
TCCACGGCCGGGGCGGCAGAGACCCGGGGGCTGTGGAGGGGGCAGCCCC
GACCACGCCCTCCCGGCGGGAGATCACCGGTGTGCAGGCACAGCCAGGCG
AGGCCACGTCCGGCCCGCCGGGGATCCAGCCCGGCCAGGAGCCCCCGGTC
ACAATGATCTTCATGGGTTACCAGAACGTGGAGGATGAGGCCGAGACCAA
GAAGGTGCTGGGCCTTCAAGATACCATCACGGCGGAGCTGGTGGTCATCG
AAGACGCGGCTGAGCCCAAGGAGCTGCACCACCCAACGGCAGTGCTGCC
GAGCCTCCCACGGAGGCCGCCTCCAGGGAAGAGAATCAGGCGGGGCCGA
GGCCACCACCAGCGACCCCCAGGACCTCGACATGAAGAAGCACCGTTGTA
AATGCTGCTCCATCATGtgagcggccgc