

MRCPPU Reagents and Services

Standard Operating Procedure

Preparation of CDC37 (1 – 378)

<u>Enzyme description:-</u>	CDC37 [1 – 378]
<u>Clone number:-</u>	DU 53034
<u>Source:-</u>	Recombinant
<u>Expression system:-</u>	Baculovirus expression vector system
<u>Tag:-</u>	N-terminal His(6)
<u>Purification method:-</u>	Cobalt Agarose
<u>Calculated molecular mass:-</u>	
Monoisotopic	47, 809.41 daltons
Average Mass	47, 839.93 daltons
[cysteines reduced, methionines have not been oxidised]	
<u>Theoretical pI:-</u>	5.27
<u>Purity:-</u>	>80 %
<u>Enzyme storage buffer:-</u>	
50 mM Tris-HCl pH 7.5, 270 mM Sucrose, 150 mM NaCl, 0.1 mM EGTA, 0.1 % 2-mercaptoethanol	
<u>Storage temperature:-</u>	-70 °C

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Clone Data Sheet

CDC37 [1 – 378]

Protein CDC37 [1 – 378]

Clone number DU 53034

Species Human

Accession number NM_007065.3

Tags N-terminal His6

Baculovirus expressed protein MSYYHHHHHHHDYDIPTTENLYFQGAMGSMVDYSVVDHIEVSDDEDETHP
NIDTASLFRWRHQARVERMEQFQKEKEELDRGCRECKRKVAECQRKLKE
LEVAEGGKAELERLQAEAQQLRKEERSWEQKLEMRKKEKSMPWNVDTL
SKDGFSSMVNTKPEKTEEDSEEVREQKHKTFFVEKYEQIKHFGMLRRW
DDSQKYLSDNVHLVCEETANYLVIWCIDLEVEEKCALMEQVAHQTIVMQ
FILELAKSLKVDPRACFRQFFTKIKTADRQYMEGFNDELEAFKERVGR
AKLRIEKAMKEYEEEEERKKRLGPGGLDPVEVYESLPEELQKCFDVKDVO
MLQDAISKMDPTDAKYHMQRCDISGLWVPNSKASEAKEGEEAGPGDPLL
EAVPKTGDEKDVSV

Native sequence Amino acids M1 – V378 (end) of human CDC37.
Residue M29 of the fusion protein is equivalent to M1 of the native enzyme. The His(6) tag is located at residues 5 – 10.

Protease cleavage TEV Protease (ENLYFQG) residues 14 - 21

Cloning sites *Bam*H1 and *Not*1 sites of pFastBac HTb

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Nucleotide
Sequence of insert

ggatccATGGTGGACTACAGCGTGTGGGACCACATTGAGGTGTCTGATG
ATGAAGACGAGACGCACCCCAACATCGACACGGCCAGTCTCTTCCGCTG
GCGGCATCAGGCCCGGGTGGAAACGCATGGAGCAGTTCCAGAAGGAGAAG
GAGGAACTGGACAGGGGCTGCCGCGAGTGCAAGCGCAAGGTGGCCGAGT
GCCAGAGGAAACTGAAGGAGCTGGAGGTGGCCGAGGGCGGCAAGGCAGA
GCTGGAGCGCCTGCAGGCCGAGGCACAGCAGCTGCGCAAGGAGGAGCGG
AGCTGGGAGCAGAAGCTGGAGGAGATGCGCAAGAAGGAGAAGAGCATGC
CCTGGAACGTGGACACGCTCAGCAAAGACGGCTTCAGCAAGAGCATGGT
AAATACCAAGCCCAGAGAAGACGGAGGAGGACTCAGAGGAGGTGAGGGAG
CAGAAACACAAGACCTTCGTGGAAAAATACGAGAAACAGATCAAGCACT
TTGGCATGCTTCGCCGCTGGGATGACAGCCAAAAGTACCTGTCAGACAA
CGTCCACCTGGTGTGCGAGGAGACAGCCAATTACCTGGTCATTTGGTGC
ATTGACCTAGAGGTGGAGGAGAAATGTGCACTCATGGAGCAGGTGGCCC
ACCAGACAATCGTCATGCAATTTATCCTGGAGCTGGCCAAGAGCCTAAA
GGTGGACCCCCGGGCCTGCTTCCGGCAGTTCTTCACTAAGATTAAGACA
GCCGATCGCCAGTACATGGAGGGCTTCAACGACGAGCTGGAAGCCTTCA
AGGAGCGTGTGCGGGGCCGTGCCAAGCTGCGCATCGAGAAGGCCATGAA
GGAGTACGAGGAGGAGGAGCGCAAGAAGCGGCTCGGCCCGGCGGCCTG
GACCCCGTCGAGGTCTACGAGTCCCTCCCTGAGGAACTCCAGAAGTGCT
TCGATGTGAAGGACGTGCAGATGCTGCAGGACGCCATCAGCAAGATGGA
CCCCACCGACGCAAAGTACCACATGCAGCGCTGCATTGACTCTGGCCTC
TGGGTCCCCAACTCTAAGGCCAGCGAGGCCAAGGAGGGAGAGGAGGCAG
GTCCTGGGGACCCATTACTGGAAGCTGTTCCCAAGACGGGCGATGAGAA
GGATGTCAGTGTGtgagcggccgc