MRC PPU REAGENTS

Standard Operating Procedure

Preparation of RAB11FIP4 [1 - 637]

Enzyme description:- RAB11FIP4 [1 – 637]

Clone number:- DU 26818

Source:- Recombinant

Expression system:- *E.coli*

Tag:- N-terminal GST

Purification method:- GSH Sepharose

Calculated molecular mass:-

Monoisotopic 99, 063.26 daltons
Average Mass 99, 125.76 daltons
[cysteines reduced, methionines have not been oxidised

Theoretical pI:- 4.91

Purity:- >75 %

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 deg C

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Clone Data Sheet

RAB11FIP4 [1 – 637]

Protein	RAB11FIP4 [1 – 637]
Clone number	DU 26818
Species	Human
Accession number	NM_032932.5
Tags	N-terminal GST
Bacterially expressed protein	MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFEL GLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLE GAVLDIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLN GDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKY LKSSKYIAWPLQGWQATFGGGDHPPKSDLEVLFQGPLGSPEF**MAGGAG WSGAPAALLRSVRRLREVFEVCGRDPDGFLRVERVAALGLRFGQGEEV EKLVKYLDPNDLGRINFKDFCRGVFAMKGCEELLKDVLSVESAGTLPC APEIPDCVEQGSEVTGPTFADGELIPREPGFFPEDEEEAMTLAPPEGP QELYTDSPMESTQSLEGSVGSPAEKDGGLGGLFLPEDKSLVHTPSMTT SDLSTHSTTSLISNEEQFEDYGEGDDVDCAPSSPCPDDETRTNVYSDL GSSVSSSAGQTPRKMRHVYNSELLDVYCSQCCKKINLLNDLEARLKNL KANSPNRKISSTAFGRQLMHSSNFSSSNGSTEDLFRDSIDSCDNDITE KVSFLEKKVTELENDSLTNGDLKSKLKQENTQLVHRVHELEEMVKDQE TTAEQALEEEARRHREAYGKLEREKATEVELLNARVQQLEEENTELRT TVTRLKSQTEKLDEERQRMSDRLEDTSLRLKDEMDLYKRMMDKLRQNR LEFQKEREATQELIEDLRKELEHLQMYKLDCERPGRGRSASSGLGEFN ARAREVELEHEVKRLKQENYKLRDQNDDLNGQILSLSLYEAKNLFAAQ TKAQSLAAEIDTASRDELMEALKEQEEINFRLRQYMDKIILAILDHNP SILEIKH**
Native sequence	Amino acids M1 – H637 (end) of human RAB11FIP4. Residue M235 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	*Eco*R1 and *Sal*1 sites of pGEX6P-1

Nucleotide Sequence Of Insert

gaattcATGGCGGGCGGCGCGGGCTGGTCGGGCGCCCCCGCGGCTCTGCTGCGCTCCGTGCGCCG
CCTGCGCGAGGTGTTCGAGGTGTGCGGCCGCGACCCCGACGGCTTCCTGCGCGTGGAGCGCGTCG
CGGCGCTCGGACTGCGCTTCGGCCAGGGCGAGGAGGTGGAAAAACTTGTGAAATATTTGGATCCC
AACGACCTGGGGAGAATCAACTTCAAGGACTTTTGCCGGGGGGTGTTCGCCATGAAAGGGTGCGA
GGAGCTGCTGAAGGATGTGCTGTCGGTGGAGAGCGCGGGGACGCTGCCGTGCGCGCCAGAGATCC
CAGACTGCGTGGAGCAGGGCAGCGAGGTCACAGGCCCCACCTTTGCTGATGGCGAGCTCATCCCC
AGGGAACCCGGCTTTTTTCCCGAGGACGAGGAGGAGGCTATGACGCTGGCGCCACCTGAGGGCCC
CCAGGAGTTGTACACAGACAGCCCCATGGAGAGCACTCAGAGCCTGGAGGGGTCTGTCGGGAGTC
CTGCCGAGAAGGACGGGGGACTTGGGGGCCTGTTTCTGCCAGAAGACAAGTCCCTGGTCCACACT
CCATCCATGACGACCTCAGACCTTTCTACACACTCCACCACCTCGCTCATCAGCAATGAGGAGCA
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CAAGCGCATGATGGACAAGCTGCGACAGAACCGCCTTGAGTTCCAGAAGGAGCGGGAGGCGACGC
AGGAGCTCATCGAGGACTTGCGGAAGGAGCTGGAGCACCTGCAGATGTACAAGCTGGACTGCGAG
CGGCCAGGCAGGGGCCGCAGTGCCTCCTCTGGCCTAGGCGAGTTCAATGCCAGGGCCCGCGAGGT
GGAGCTCGAGCACGAGGTCAAGCGGCTCAAGCAGGAGAATTATAAGCTGCGGGATCAGAACGACG
ACTTGAATGGGCAGATTTTGAGCCTCAGCCTCTACGAAGCAAAAAACCTCTTTGCTGCCCAGACT
AAAGCCCAGTCTCTGGCTGCAGAGATAGACACCGCCTCGCGCGATGAGCTAATGGAAGCCCTGAA
GGAGCAGGAGGAGATCAACTTCCGGCTGAGGCAGTACATGGACAAGATTATCCTCGCCATCCTGG
ACCACAATCCCTCCATCCTCGAGATCAAACACtaagtcgac