

Certificate of Analysis

FRAP1 (mTOR), 100 µg

FK506 binding protein 12-Rapamycin Associated Protein 1, GST-tagged

ThermoFisher
SCIENTIFIC

Part Number: PR8683B

Lot Number: 2902334A

Immediate Storage: -80°C

Shipping Conditions: dry ice

5781 Van Allen Way

Carlsbad, CA 92008

Phone: 760.603.7200

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Description:

Recombinant Human (amino acids 1360-2549), GST-tagged, expressed in insect cells. No special measures were taken to activate this kinase.

Specific Activity:

32 nmoles of phosphate transferred to PHAS-1 substrate per minute per mg of total protein at 30°C. Activity determined at a final protein concentration of 5 µg/mL.

Concentration:

0.21 mg/mL total protein as measured using the Bradford protein assay with BSA as a standard.

Calculated **1,280 nM**.

Aliases:

mTOR, FRAP, FRAP2, TOR, RAFT1

Storage and Handling:

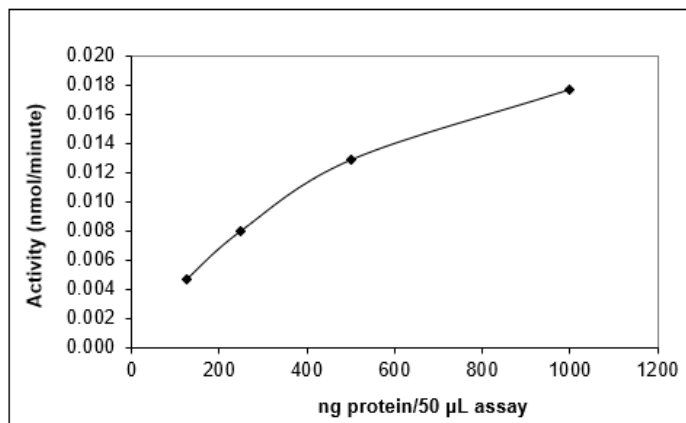
For maximum recovery please spin prior to use. Unless noted below, aliquots of the 5 µg, 10 µg and 20 µg sizes of kinase are not recommended as materials can be used in original packaging until exhausted. For larger sizes, the number of freeze/thaws may be reduced by preparing aliquots, aliquots below 20 µL are not recommended. **Please never store a kinase diluted.** If properly stored at -80°C, this product is guaranteed for 6 months from date of purchase.

Storage Buffer:

50 mM Tris (pH 7.5), 150 mM NaCl, 0.5 mM EDTA, 0.1% Polysorbate 20, 2 mM DTT and 50% Glycerol.

QUALITY ASSURANCE

FRAP1 (mTOR) Activity Graph



Dilution Buffer:

20 mM HEPES (pH 7.5), 0.02% Polysorbate 20, 0.1 mg/mL BSA, and 2 mM DTT.

Assay Conditions:

FRAP1 (mTOR) was pre-diluted in enzyme dilution buffer and assayed in 50 mM HEPES (pH 7.5), 10 mM MnCl₂, 1 mM EGTA, 2.5 mM DTT, 0.01% Polysorbate 20, 100 µM ATP, 500 µg/mL PHAS-1 substrate and trace [³²P]-γ-ATP for 10 minutes at 30°C.

Gel Information for FRAP1 (mTOR)

Page Description: The SDS-PAGE and/or Native PAGE were run on 4-20% Tris-Glycine Novex™ gels (Catalog #: EC6025BOX).

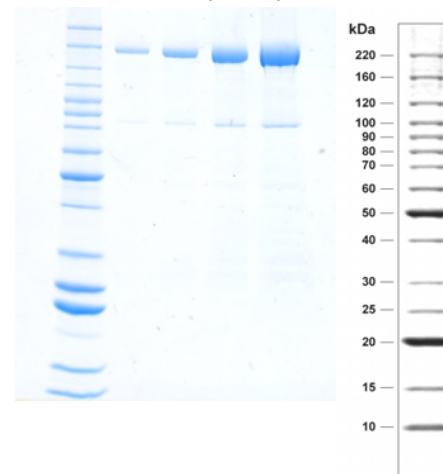
Lane 1: Invitrogen™ BenchMark™ Protein Ladder (Catalog #: 10747-012).

Lane 2: 0.4 µg FRAP1 (mTOR)

Lane 3: 0.8 µg FRAP1 (mTOR)

Lane 4: 2.0 µg FRAP1 (mTOR)

Lane 5: 4.0 µg FRAP1 (mTOR)



Purity:

80% as determined by a SDS-PAGE gel stained with SimplyBlue™ SafeStain.

Molecular Weight:

163.9 kDa. Calculated from the protein sequence(s).

Mass Spectrometry:

FRAP1 (mTOR) was subjected to proteolytic digest followed by mass spec analysis. The resulting MS/MS data verified FRAP1 (mTOR) identity by comparison against the amino acid sequence(s) of the recombinant protein.

Protein sequence alignment with reference sequence(s)

GenBank Accession Number: NP_004949.1

1	MAPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	GDVKLTQDMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	GST TAG
1	MAPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	GDVKLTQDMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	IVGN FRAP1(MTOR)
1360											NP_004949.1
101	DIRYGVSRIA	YSKDFETLKV	DFLSKLPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	KRIEAIQID	KYLKSSKYIA	
101	DIRYGVSRIA	YSKDFETLKV	DFLSKLPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	KRIEAIQID	KYLKSSKYIA	
1360											
201	WPLQGWQATF	GGGDHPPKSD	LVPR								
201	WPLQGWQATF	GGGDHPPKSD	LVPRHNTQSL	YKKAGTMLAE	FMEHSDKGPL	PLRDDNGIVL	LGERAAKRA	YAKALHYKEL	EFQKGPTPAI	LESLSINNK	
1360				-----LAE	FMEHSDKGPL	PLRDDNGIVL	LGERAAKRA	YAKALHYKEL	EFQKGPTPAI	LESLSINNK	
224											
301	LQQPEAAAGV	LEYAMKHFG	LEIQATWYEK	LHEWEDALVA	YDKKMDTNKD	DPELMLGRMR	CLEALGEWQ	LHQQCCEKWT	LVNDETQAKM	ARMAAAAWG	
1423	LQQPEAAAGV	LEYAMKHFG	LEIQATWYEK	LHEWEDALVA	YDKKMDTNKD	DPELMLGRMR	CLEALGEWQ	LHQQCCEKWT	LVNDETQAKM	ARMAAAAWG	
224											
401	LGQWDSMEEY	TCMIPRDTHD	GAFYRAVLAL	HQDLFSLAQ	CIDKARDLLD	AELTAMAGES	YSRAYGAMVS	CHMLSELEEV	IQYKLVPER	EIRQIWWER	
1523	LGQWDSMEEY	TCMIPRDTHD	GAFYRAVLAL	HQDLFSLAQ	CIDKARDLLD	AELTAMAGES	YSRAYGAMVS	CHMLSELEEV	IQYKLVPER	EIRQIWWER	
224											
501	LQGCQRIVED	WQKILMVRSL	VVSPHEDMRT	WLKYASLCGK	SGRLALAHKT	LVLLLGVDP	RQLDHPLPTV	HPQVTYAYMK	NMWSARKID	AFQHMVHFVQ	
1623	LQGCQRIVED	WQKILMVRSL	VVSPHEDMRT	WLKYASLCGK	SGRLALAHKT	LVLLLGVDP	RQLDHPLPTV	HPQVTYAYMK	NMWSARKID	AFQHMVHFVQ	
224											
601	TMQQAQAHAI	ATEDQQHKE	LHKLMARCF	KLGEWQLNLQ	GINESTIPKV	LQYSAATEH	DRSWYKAWHA	WAVMNF	HYKHQNOARD	EKKKL RHASG	
1723	TMQQAQAHAI	ATEDQQHKE	LHKLMARCF	KLGEWQLNLQ	GINESTIPKV	LQYSAATEH	DRSWYKAWHA	WAVMNF	HYKHQNOARD	EKKKL RHASG	
224											
701	ANITNATTAA	TTAATATTTA	STEGSNSSE	AESTENSPT	SPLQKKVTD	LSKTLMLYTV	PAVQGF	SLSRGNLQD	TLRVLTWFD	YGHWP	PDVNEA
1823	ANITNATTAA	TTAATATTTA	STEGSNSSE	AESTENSPT	SPLQKKVTD	LSKTLMLYTV	PAVQGF	SLSRGNLQD	TLRVLTWFD	YGHWP	PDVNEA
224											
801	LVEGVKAIQI	DTWLQVIPQL	IARIDTPRPL	VGRLIHQLLT	DIGRYHPQAL	IYPLTVASKS	TTTARHNAAN	KILKNMCEHS	NTLVQQAMMV	SEELIRVAIL	
1923	LVEGVKAIQI	DTWLQVIPQL	IARIDTPRPL	VGRLIHQLLT	DIGRYHPQAL	IYPLTVASKS	TTTARHNAAN	KILKNMCEHS	NTLVQQAMMV	SEELIRVAIL	
224											
901	WHEMWHEGLE	EASRLYFGER	NVKGMFEVLE	PLHAMMERGP	QTLKETSFNQ	AYGRDLME	EWCRKYMKS	NVKDLTQAWD	LYYHVFRRIS	KQLPQLTSLE	
2023	WHEMWHEGLE	EASRLYFGER	NVKGMFEVLE	PLHAMMERGP	QTLKETSFNQ	AYGRDLME	EWCRKYMKS	NVKDLTQAWD	LYYHVFRRIS	KQLPQLTSLE	
224											
1001	LQYVSPKLLM	CRDLELAVPG	TYDPNQPIIR	IQSIAPSLQV	ITSKQRPRL	TLMGSGHEF	VFLKKGHEDL	RQDERVMQLF	GLVNTLLAND	PTSLRKNLSI	
2123	LQYVSPKLLM	CRDLELAVPG	TYDPNQPIIR	IQSIAPSLQV	ITSKQRPRL	TLMGSGHEF	VFLKKGHEDL	RQDERVMQLF	GLVNTLLAND	PTSLRKNLSI	
224											
1101	QRYAVIPLST	NSGLIGWVPH	CDTLHALIRD	YREKKKILLN	IEHRIMLRMA	PDYDHLTMQ	KVEVFEHAVN	NTAGDDLAKL	LWLKSPSSEV	WFDRTNYTR	
2223	QRYAVIPLST	NSGLIGWVPH	CDTLHALIRD	YREKKKILLN	IEHRIMLRMA	PDYDHLTMQ	KVEVFEHAVN	NTAGDDLAKL	LWLKSPSSEV	WFDRTNYTR	
224											
1201	SLAVMSMGVY	ILGLGDRHPS	NMLDRLSGK	ILHIDFGDCF	EVAMTREKFP	EKIPFRLTRM	LTNAMEVTGL	DGNYRITCHT	VMEVLREHKD	SVM	AVLEAFV
2323	SLAVMSMGVY	ILGLGDRHPS	NMLDRLSGK	ILHIDFGDCF	EVAMTREKFP	EKIPFRLTRM	LTNAMEVTGL	DGNYRITCHT	VMEVLREHKD	SVM	AVLEAFV
224											
1301	YDPLLNRWLM	DTNTKGNKRS	RTRTDSYSAG	QSVEILDGVE	LGEPAHKKTG	TTVPESIHSF	IGDGLVKPEA	LNNKAIQIIN	RVRDKLTGRD	FSHDDTLDP	
2423	YDPLLNRWLM	DTNTKGNKRS	RTRTDSYSAG	QSVEILDGVE	LGEPAHKKTG	TTVPESIHSF	IGDGLVKPEA	LNNKAIQIIN	RVRDKLTGRD	FSHDDTLDP	
224											
1401	TQVELLIKQA	TSHENLCQCY	IGWCPFW								
2523	TQVELLIKQA	TSHENLCQCY	IGWCPFW								

* highlighted residues denote differences from the reference protein sequence(s).

Chevoyn Joseph, Director, Quality

Date: 06/Aug/2024

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