

RB-CTF

RB transcriptional corepressor 1

Recombinant Protein Kinase Substrate

HGNC Symbol: RB1

Synonyms: OSRC, P105-RB, PP110, RB

Product No.: 0040-0000-6

Lot: 019

Description: Human RB, C-terminal fragment, amino acids S773-K928 (as in [NCBI/Protein](#) entry NP_000312.1), N-terminal GST fusion protein with a factor Xa cleavage site, expressed in E.coli

Theoretical MW_{Fusion Protein}: 44,460 Da

Expression host: E.coli

Purification: GST-Affinity chromatography, ion-exchange chromatography

ATPase activity: In an ADP-Glo™ assay (Promega) with 10 µM ATP or 30 µM ATP, the ATP → ADP conversion within 30 min is approx. 1% at a concentration of 100 µg/ml substrate.

Detailed ATPase assay conditions on request

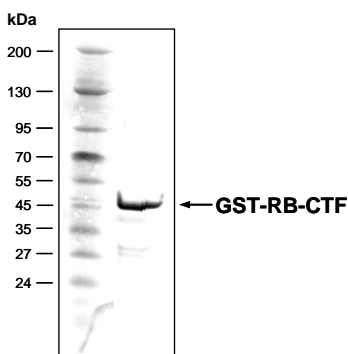
Storage buffer: 50 mM HEPES pH 7.5, 100 mM NaCl, 5 mM DTT, 20 % glycerol

Storage temperature: -80°C

For complete recovery, mix well and spin before use. Product must not be stored in diluted solutions, aliquots below 10µl are not advisable. Avoid repeated freeze-thaw cycles!

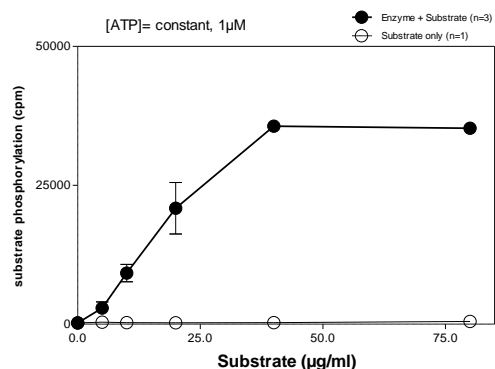
Protein concentration: 0.678 µg/µl (Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

RB-CTF Lot 019:
Coomassie stain



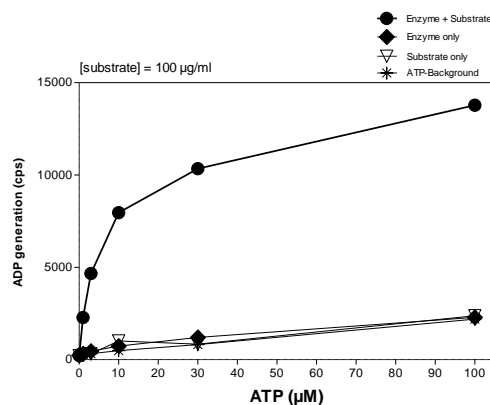
3.0 µg RB-CTF

Phosphorylation of RB-CTF Lot 019 by the kinase CDK4/CycD1 Radiometric filter binding assay:



Assay conditions:
70 mM HEPES-NaOH, pH 7.5
3 mM MgCl₂
3 mM MnCl₂
3 µM Na-orthovanadate
1.2 mM DTT
50 µg/ml PEG_{20,000}
ATP: 1 µM
Substrate: variable concentration
CDK4/CycD1: 4 µg/ml
MSFC membrane (Millipore)

Phosphorylation of RB-CTF Lot 019 by the kinase CDK4/CycD1 ADP-Glo™ assay (Promega)



Assay conditions:
70 mM HEPES-NaOH, pH 7.5
3 mM MgCl₂
3 mM MnCl₂
3 µM Na-orthovanadate
1.2 mM DTT
50 µg/ml PEG_{20,000}
ATP: variable concentration
1 % (v/v) DMSO
Substrate: 100 µg/ml
CDK4-CycD1: 2 µg/ml

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SUBSTRATE Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP EML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQG WQATF	GGGDHPPKSD	L IEGRG IRIP	RG STRPPTLS	240
241	PIPHIPRSPY	KFPSSPLRIP	GGNIYISPLK	SPYKISEGLP	TPTKMTPRSR	ILVSIGESFG	300
301	TSEKFQKINQ	MVCNSDRVLK	RSAEGSNPPK	PLKKLRFDIE	GSDEADGSKH	LPGESKFQQK	360
361	LAEMTSTRTR	MQKQKMND SM	DTSNKEEK				420

1-218: GST **Pink**: Factor Xa cleavage site **blue**: RB1 fragment

Recombinant Proteins

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