

## RB ER-CDC25tide

Generic Recombinant Protein Kinase Substrate

HGNC Symbol: n/a

Synonyms: n/a

Product No.: 0590-0000-1

Lot: 012

**Description:** Artificial fusion protein consisting of an N-terminal GST-tag separated by a Thrombin cleavage site from a fragment of the human retinoblastoma protein RB1, amino acids S<sub>773</sub>-K<sub>928</sub> (as in [NCBI/Protein](#) entry NP\_000312.2) followed by 11 Arg residues (ER) and a peptide sequence (ISDELMDATFADQEAQ; CDC25tide). Expressed in E.coli.

**Theoretical MW<sub>Fusion Protein</sub>:** 47,608 Da

**Expression host:** E.coli

**Purification:** GST-Affinity Chromatography

### Field of application:

RB ER-CDC25tide has been validated for use in radiometric in-vitro kinase activity assays.

It is not recommended for use in ATP-consumption based kinase activity assays.

**Storage buffer:** 50 mM HEPES pH 7.5, 100 mM NaCl, 5 mM DTT, 15 mM reduced glutathione, 10 % glycerol

**Storage temperature:** -80°C

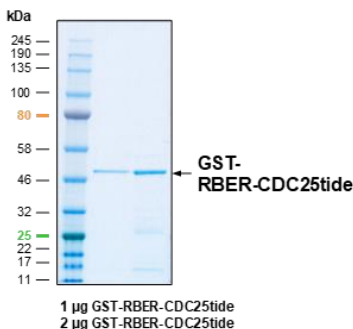
For complete recovery, mix well and spin before use. Product must not be stored in diluted solutions, aliquots below 10µl are not advisable. Avoid repeated freeze-thaw cycles!

**Protein concentration:** 1.274 µg/µl

(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

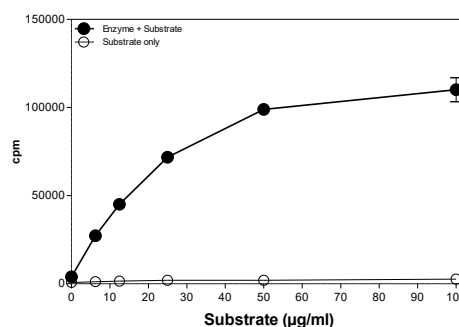
### RB ER-CDC25tide Lot 012:

#### Coomassie stain



### Phosphorylation of RB ER-CDC25tide by PLK1

#### Radiometric filter binding assay



#### Assay conditions:

70 mM HEPES-NaOH, pH 7.5  
 3 mM MgCl<sub>2</sub>  
 3 mM MnCl<sub>2</sub>  
 3 µM Na-orthovanadate  
 1.2 mM DTT  
 50 µg/ml PEG<sub>20,000</sub>  
 ATP: 1 µM  
 Substrate: variable concentration  
 Kinase: 2 µg/ml  
 MSFC membrane (Millipore)

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# RB ER-CDC25tide

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RB ER-CDC25tide Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP EML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	<b>LVPRGSP EFS</b>	<b>TRPPTLSPI P</b>	240
241	<b>HIPRSPYKFP</b>	<b>SSPLRIPGGN</b>	<b>IYISPLKSPY</b>	<b>KISEGLPTPT</b>	<b>KMTPRSRILV</b>	<b>SIGESFGTSE</b>	300
301	<b>KFQKINQ MVC</b>	<b>NSDRVLK RSA</b>	<b>EGSNPPKPLK</b>	<b>KLRFDIEGSD</b>	<b>EADGSKH LPG</b>	<b>ESKFQQLAE</b>	360
361	<b>MTSTRTRMQK</b>	<b>QKMND SMDTS</b>	<b>NKEEKRRRRR</b>	<b>RRRRRR</b>	<b>ISDELM DATFADQ EAK</b>		420

1-218: GST **Pink**: Thrombin cleavage site **blue**: RB1 fragment **Green**: R<sub>11</sub>-sequence **boxed**: CDC25tide sequence

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