## **Certificate of Analysis**



### **ProQinase™ RBER-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 (ISDELMDATFADQEAK; 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:

JNK1 K55R/K56R 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.

based killase 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

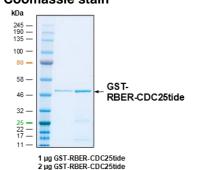
For complete recovery, mix well and spin before use. Product must not be stored in diluted solutions, aliquots below 10µl are not advisable. Avoid repeated freeze-thaw cycles!

Protein concentration: 1.274 μg/μl

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

as standard protein)

# RBER-CDC25tide Lot 012: Coomassie stain

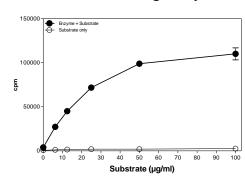


This product was manufactured at Reaction Biology in Freiburg, Germany, and is for in vitro research use only, not for use in humans or animals.

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# Phosphorylation of RBER-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)

Recombinant Proteins



## **ProQinase™ RBER-CDC25tide**

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RBER-CDC25tide Recombinant Fusion Protein Amino Acid Sequence			
1	MSPILGYWKI KGLVQPTRLL LEYLEEKYEE	HLYERDEGDK WRNKKFELGL EFPNLPYYID	60
61	GDVKLTQSMA IIRYIADKHN MLGGCPKERA	EISMLEGAVL DIRYGVSRIA YSKDFETLKV	120
121	DFLSKLPEML KMFEDRLCHK TYLNGDHVTH	PDFMLYDALD VVLYMDPMCL DAFPKLVCFK	180
181	KRIEAIPQID KYLKSSKYIA WPLQGWQATF	GGGDHPPKSD LVPRGSPEFS TRPPTLSPIP	240
241	HIPRSPYKFP SSPLRIPGGN IYISPLKSPY	KISEGLPTPT KMTPRSRILV SIGESFGTSE	300
301		KLRFDIEGSD EADGSKHLPG ESKFQQKLAE	360
361	MTSTRTRMQK QKMNDSMDTS NKEEKRRRRR	RRRRRR I SDELMDATFADQEAK	420

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

Recombinant Proteins