NOVEX[®] by *life* technologies[®]

Rabbit (polyclonal) Anti-RSK1 [pS221] / 2 [pS227] Phosphospecific Antibody, Unconjugated

Store at 2° C to 8° C (short-term), or -20° C (long-term)

Catalog Number: 44924G

Pub. No. MAN0005702 **Rev.** A.00

Clonality: Polyclonal	Quantity:	10 mini-blot size	Volume : 100 μL
Host/Class: Rabbit IgG	Reactivity:	Human RSK1 [pS ²²¹]/ 2 [pS ²²⁷]	Predicted Reactivity: Human, Mouse, Chicken

Product description

RSK1 is a member of the broadly expressed p90 Ribosomal S6 Kinase (RSK) family of serine/threonine kinases. RSK proteins possess two separate kinase domains, the C-terminal kinase domain and the N-terminal kinase domain, separated by a linker region containing a hydrophobic motif. They are substrates for, and downstream transducers of MAPK signaling proteins (e.g. ERK1 and ERK2). RSK proteins are activated by growth factors, phorbol esters, cAMP, heat shock, and irradiation. Activation is a multi-step process involving phosphorylation of multiple residues within the three domains. PDK1 is a constitutively active cytoplasmic kinase that phosphorylates Ser 221 (Ser 227 in RSK2) to activate the N-terminal kinase domain to phosphorylate various substrates that include transporters, transcription factors, and transcription co-activators.

Product specifications

Cross Reactivity:	Mouse, Human RSK2		
	Human RSK4		
Immunogen:	Chemically synthesized phosphopeptide derived from a region of human RSK1 containing serine 221		
Alternate Names:	MAPKAP-K1a (MAPK Activated Protein Kinase-1a)		
Purification:	Antibody negatively preadsorbed using a non- phosphopeptide then purified by epitope-specific affinity chromatography		
Lot:	See product label		

Product applications

The antibody has been used in Western blotting. Other applications may work but have not been tested.

Because conditions may vary, it is recommended that each investigator determine the optimal amount of antibody to be used for each application.

Storage and handling

Store the antibody at 2°C to 8°C for up to 1 week, or apportion into working aliquots and keep at –20°C for long-term storage.



Figure 1 Upregulation and Antibody-Peptide Competition.

Extracts of A431 cells unstimulated (lane 1) or stimulated with PMA (100 ng/mL for 30 minutes at 37° C) (lanes 2–5) were resolved on a 10% Tris-glycine gel and transferred to PVDF. The membrane was blocked with 3% milk-TBST for 1 hr at room temperature and incubated with the RSK1 [pS²²¹]/2 [pS²²⁷] antibody for 2 hrs at room temperature in 1% milk-TBST, following prior incubation with: no peptide (lanes 1, 2), a non-phosphorylated peptide corresponding to the immunogen (lane 3), a generic phosphoserine-containing peptide (lane 4), or the phosphopeptide immunogen (lane 5). The blots were developed using chemiluminescence (ECL) method with a goat F(ab')² anti-rabbit IgG HRP conjugate (Cat. no. ALI4404).

Only the phosphopeptide corresponding to RSK1 [pS²²¹]/2 [pS²²⁷] blocks the antibody signal (lane 5) demonstrating the specificity of the antibody. The data also show RSK1/2 is phosphorylated after PMA treatment in this cell system.

Positive controls used

A431 or NIH3T3 cells treated with 100 ng/mL of PMA for 30 min at 37°C, or HeLa +/– 100–500 ng/mL PMA for 30 min.

For Research Use Only. Not for use in diagnostic procedures

Stability

When stored as instructed, expires one year from date of receipt unless otherwise indicated on the Certificate of Analysis.

Storage buffer

Dulbecco's phosphate buffered saline (without Mg²⁺ and Ca²⁺), pH 7.3 (+/– 0.1), 50% glycerol with 1.0 mg/mL BSA (IgG, protease free) as a carrier, and 0.05% sodium azide.



CAUTION! Sodium azide is extremely toxic and may react with lead and copper plumbing to form highly explosive metal azides. Properly dispose of solutions containing sodium azide. Read the Safety Data Sheet (SDS) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. SDSs are available at www.lifetechnologies.com/support.

References

- Cavet, M.E., et al. (2003) 14-3-3[®] is a p90 Ribosomal S6 Kinase (RSK) isoform 1-binding protein that negatively regulates RSK kinase activity. *J. Biol. Chem.* 278(20):18376-18383.
- Roux, P.P., et al. (2003) Phosphorylation of p90 Ribosomal S6 Kinase (RSK) regulates Extracellular Signal-Regulated Kinase docking site and RSK activity. *Mol. Cell. Biol.* 23(14):4796-4804.
- Kim, K.-W., et al. (2001) Extracellular Signal-Regulated Kinase/90-kDa Ribosomal S6 Kinase/ Nuclear Factor B pathway mediates phorbol-12-myristate 13-acetate-induced megakaryocytic differentiation in K562 cells. *J. Biol. Chem.* 276(16):13186-13191.
- 4. Richards, S.A., et al. (2001) Characterization of regulatory events associated with membrane targeting of p90 Ribosomal S6 Kinase 1. *Mol. Cell. Biol.* 21(21):7470-7480.
- Zhang, Y., et al. (2001) UVA induces Ser³⁸¹ phosphorylation of p90^{RSK}/MAPKAP-K1 via ERK and JNK pathways. *J. Biol. Chem.* 276(18):14572-14580.
- Jensen, C.J., et al. (1999) 90-kDa Ribosomal S6 Kinase is phosphorylated and activated by 3-Phosphoinositide-Dependent Protein Kinase-1. *J. Biol. Chem.* 274(38):27168-27176.
- 7. Smith, J.A., et al. (1999) Identification of an Extracellular Signal-Regulated Kinase (ERK) docking site in Ribosomal S6

Related products

Product Name	Quantity	Cat. No.
RSK1 [pS ³⁸⁰] Rabbit Polyclonal Antibody	10 blots	44928G
ERK1/2 [pTpY ^{185/187}] Polyclonal Antibody, Rabbit	10 blots	44680G
PDK-1 Rabbit anti-Human Polyclonal Antibody	50 µg	AHZ0512
JNK (SAPK) [pTpY ^{183/185}] Polyclonal Antibody, Rabbit	10 blots	44682G
p70-S6K [pT ²²⁹] Rabbit Polyclonal Antibody	10 blots	44918G
I-kappa-B-alpha [pSpS ^{32/36}] Rabbit Polyclonal Antibody	10 blots	44726G

Product Documentation

To obtain a Certificate of Analysis or Safety Data Sheets (SDSs), visit **www.lifetechnologies.com/support.**

Important Licensing Information

This product may be covered by one or more Limited Use Label Licenses. By use of this product, you accept the terms and conditions of all applicable Limited Use Label Licenses.

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale found on Life Technologies' website at **www.lifetechnologies.com/termsandconditions**. If you have any questions, please contact Life Technologies at **www.lifetechnologies.com/support**.

-	•				
Symbol	Description	Symbol	Description	Symbol	Description
	Manufacturer	REF	Catalog number	LOT	Batch code
	Use by	X	Temperature limitation		
i	Consult instructions for use	\triangle	Caution, consult accompanying documents		

DISCLAIMER: LIFE TECHNOLOGIES AND/OR ITS AFFILIATE(S) DISCLAIM ALL WARRANTIES WITH RESPECT TO THIS DOCUMENT, EXPRESSED OR IMPLIED, INCLUDING BUT NOT LIMITED TO THOSE OF MERCHANTABILITY, FITNESS FOR A PARTICULAR PURPOSE, OR NON-INFRINGEMENT. TO THE EXTENT ALLOWED BY LAW, IN NO EVENT SHALL LIFE TECHNOLOGIES AND/OR ITS AFFILIATE(S) BE LIABLE, WHETHER IN CONTRACT, TORT, WARRANTY, OR UNDER ANY STATUTE OR ON ANY OTHER BASIS FOR SPECIAL, INCIDENTAL, INDIRECT, PUNITIVE, MULTIPLE OR CONSEQUENTIAL DAMAGES IN CONNECTION WITH OR ARISING FROM THIS DOCUMENT, INCLUDING BUT NOT LIMITED TO THE USE THEREOF.

©2014 Life Technologies Corporation. All rights reserved. The trademarks mentioned herein are the property of Life Technologies Corporation and/or its affiliates or their respective owners.

For support visit www.lifetechnologies.com/support or email techsupport@lifetech.com

Explanation of symbols