Revision 6

Store at -20C

#8753

Phospho-p90RSK (Thr359) (D1E9) Rabbit mAb

Cell Signaling TECHNOLOGY®

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Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
W, IP, IHC-P, IF-IC	H R Mk	Endogenous	90	Rabbit IgG	#Q15418	6195

Product Usage Information

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:100
Immunohistochemistry (Paraffin)	1:50 - 1:200
Immunofluorescence (Immunocytochemistry)	1:50 - 1:100

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. Do not aliquot the antibody.

For a carrier-free (BSA and azide free) version of this product see product #55773.

Specificity/Sensitivity

Phospho-p90RSK (Thr359) (D1E9) Rabbit mAb recognizes endogenous levels of p90RSK1 protein when phosphorylated at Thr359. This antibody also detects p90RSK2 phosphorylated at Thr365 and p90RSK3 phosphorylated at Thr356. Phosphorylation of p90RSK isoforms at a proximal serine residue (Ser363 of p90RSK1, Ser369 of p90RSK2, and Ser360 of p90RSK3) does not affect the ability of this antibody to detect the phospho-Thr residue.

Species predicted to react based on 100% sequence homology

Chicken, Xenopus, Dog, Horse

Source / Purification

Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Thr356 of human p90RSK3 protein.

Background

The 90 kDa ribosomal S6 kinases (RSK1-4) are a family of widely expressed Ser/Thr kinases characterized by two nonidentical, functional kinase domains (1) and a carboxy-terminal docking site for extracellular signal-regulated kinases (ERKs) (2). Several sites both within and outside of the RSK kinase domain, including Ser380, Thr359, Ser363, and Thr573, are important for kinase activation (3). RSK1-3 are activated via coordinated phosphorylation by MAPKs, autophosphorylation, and phosphoinositide-3-OH kinase (PI3K) in response to many growth factors, polypeptide hormones, and neurotransmitters (3).

Upon mitogenic stimulation, p44/42 Erk1/2 and Erk5 MAP kinases cooperatively phosphorylate p90RSK at Thr573 (p90RSK1 numbering) located within the C-terminal kinase domain and at Thr359/Ser363 in the linker region between the two kinase domains (3). Phosphorylation at Thr573 within the activation loop of the p90RSK C-terminal kinase domain promotes activation and directs phosphorylation at Ser380 within the hydrophobic stretch of the linker region (4,5). When phosphorylated, Ser380 acts as a docking site for the constitutively active Ser/Thr kinase PDK1, which in turn phosphorylates p90RSK at Ser221 within the N-terminal kinase domain activation loop, resulting in full enzymatic activation of p90RSK (6). Antibodies against these phosphorylation sites are useful for understanding the kinetics and regulation of p90RSK activation.

For more information regarding the phospho-regulatory sites within each RSK isoform, including more information regarding the seminal studies demonstrating the complex phosphorylation cascades involved, please see the references herein and PhosphoSitePlus® (www.phosphosite.org).

Background References

1. Fisher, T.L. and Blenis, J. (1996) *Mol Cell Biol* 16, 1212-9.
2. Smith, J.A. et al. (1999) *J Biol Chem* 274, 2893-8.
3. Dalby, K.N. et al. (1998) *J Biol Chem* 273, 1496-505.
4. Roux, P.P. et al. (2003) *Mol Cell Biol* 23, 4796-804.
5. Cargnello, M. and Roux, P.P. (2011) *Microbiol Mol Biol Rev* 75, 50-83.
6. Romeo, Y. et al. (2012) *Biochem J* 441, 553-69.

Species Reactivity

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

Western Blot Buffer

IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

Applications Key

W: Western Blotting **IP:** Immunoprecipitation **IHC-P:** Immunohistochemistry (Paraffin) **IF-IC:** Immunofluorescence (Immunocytochemistry)

Cross-Reactivity Key

H: Human **R:** Rat **Mk:** Monkey

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