Revision 1

Store at -20C

#4951

# Phospho-KSR1 (Ser392) Antibody

Cell Signaling TECHNOLOGY®

**Orders:** 877-616-CELL (2355)
orders@cellsignal.com

**Support:** 877-678-TECH (8324)

**Web:** info@cellsignal.com
cellsignal.com

3 Trask Lane | Danvers | Massachusetts | 01923 | USA

**For Research Use Only. Not for Use in Diagnostic Procedures.**

| Applications: | Reactivity: | Sensitivity: | MW (kDa): | Source/Isotype: | UniProt ID: | Entrez-Gene Id: |
|---|---|---|---|---|---|---|
| W | H M R Mk | Endogenous | 115 | Rabbit | #Q8IVT5 | 8844 |

## Product Usage Information

| Application | Dilution |
|---|---|
| Western Blotting | 1:1000 |

## Storage

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

## Specificity/Sensitivity

Phospho-KSR1 (Ser392) Antibody detects endogenous levels of KSR1 only when phosphorylated at serine 392. This antibody may also react with phosphorylated KSR2 at the equivalent site.

## Source / Purification

Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser392 of mouse KSR1. Antibodies are purified by protein A and peptide affinity chromatography.

## Background

KSR1 (kinase supressor of Ras) was identified from a genetic screen in *Drosophila* and *C. elegans* as a component of the Ras signaling pathway (1). KSR1 has a putative carboxy-terminal kinase domain that lacks a key Lys residue for phospho-group transfer. Although reports indicate that ceramide and EGF activate KSR1 (2,3), other evidence demonstrates that KSR1 regulates Raf in a kinase-independent manner (4,5). It is now widely accepted that KSR1 functions as a scaffold that binds MEK1/2 and 14-3-3 protein constitutively and binds ERK1/2 in a Ras activation-dependent manner (1,5,6). HSP70/HSP90 and p50 Cdc37 associate with the KSR1 complex to ensure its stability (5). Multiple phosphorylation sites have been identified: Ser297 and Ser392 mediate 14-3-3 binding, and putative MAPK phosphorylation sites include Thr260, Thr274 and Ser443 (6). C-TAK1 (Cdc25C-associated kinase 1) binds and phosphorylates KSR1 at Ser392 in quiescent cells (7). In response to stimuli, Ser392 is dephosphorylated by PP2A, which leads to ERK1/2 association and allows the KSR1 complex to translocate from cytosol to membrane, where the MAPK pathway is activated (8). IMP, a Ras-responsive E3 ubiquitin ligase, is also involved in interaction with KSR1 and may regulate its localization and stability (9). Very high expression levels of KSR1 inhibit MAPK signaling, whereas physiological levels promote MAPK signaling, indicating that the scaffold protein can turn signaling "on" or "off" depending on the scaffold concentration (10).

## Background References

1. Morrison, D.K. (2001) *J. Cell Sci.* 114, 1609-1612.
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3. Xing, H.R. and Kolesnick, R. (2001) *J. Biol. Chem.* 276, 9733-9741.
4. Michaud, N. R. et al. (1997) *Proc. Natl. Acad. Sci. USA* 94, 12792-12796.
5. Stewart, S. et al. (1999) *Mol. Cell. Biol.* 19, 5523-5534.
6. Muller, J. et al. (2001) *Mol. Cell* 8, 983-993.
7. Cacace, A. M. et al. (1999) *Mol. Cell. Biol.* 19, 229-240.
8. Ory, S. et al. (2003) *Curr. Biol.* 13, 1356-1364.
9. Matheny, S. A. et al. (2004) *Nature* 427, 256-260.
10. Kortum, R.L. and Lewis, R.E. (2004) *Mol Cell Biol* 24, 4407-16.

## 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

## Cross-Reactivity Key

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

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