

# Phospho-p70 S6 Kinase (Thr389) Antibody

- Small 100 µl (10 Western mini-blots)
- Large 300 µl (30 Western mini-blots)

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This product is for *in vitro* research use only and is not intended for use in humans or animals. This product is not intended for use as a therapeutic or in diagnostic procedures.

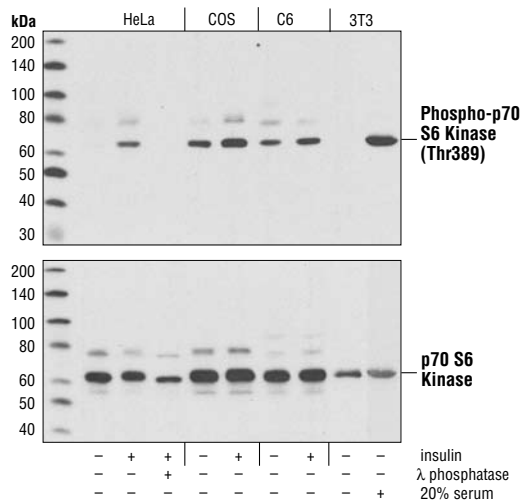
Applications	Species Cross-Reactivity*	Molecular Wt.	Source
W Endogenous	H, M, R, Mk	70, 85 kDa	Rabbit**

**Background:** p70 S6 kinase is a mitogen activated Ser/Thr protein kinase that is required for cell growth and G1 cell cycle progression (1,2). p70 S6 kinase phosphorylates the S6 protein of the 40S ribosomal subunit and is involved in translational control of 5' oligopyrimidine tract mRNAs (1). A second isoform, p85 S6 kinase, is derived from the same gene and is identical to p70 S6 kinase except for 23 extra residues at the amino-terminus, which encode a nuclear localizing signal (1). Both isoforms lie on a mitogen activated signaling pathway downstream of phosphoinositide-3 kinase (PI-3K) and the target of rapamycin, FRAP/mTOR, a pathway distinct from the Ras/MAP kinase cascade (1). The activity of p70 S6 kinase is controlled by multiple phosphorylation events located within the catalytic, linker and pseudosubstrate domains (1). Phosphorylation of Thr229 in the catalytic domain and Thr389 in the linker domain are most critical for kinase function (1). Phosphorylation of Thr389, however, most closely correlates with p70 kinase activity *in vivo* (3). Prior phosphorylation of Thr389 is required for the action of phosphoinositide 3-dependent protein kinase 1 (PDK1) on Thr229 (4,5). Phosphorylation of this site is stimulated by growth factors such as insulin, EGF and FGF, as well as by serum and some G-protein-coupled receptor ligands, and is blocked by wortmannin, LY294002 (PI-3K inhibitor) and rapamycin (FRAP/mTOR inhibitor) (1,6,7).

Ser411, Thr421 and Ser424 lie within a Ser-Pro-rich region located in the pseudosubstrate region (1). Phosphorylation at these sites is thought to activate p70 S6 kinase via relief of pseudosubstrate suppression (1,2). Another LY294002 and rapamycin sensitive phosphorylation site, Ser371, is an *in vitro* substrate for mTOR and correlates well with the activity of a partially rapamycin resistant mutant p70 S6 kinase (8).

**Specificity/Sensitivity:** Phospho-p70 S6 Kinase (Thr389) Antibody detects endogenous levels of p70 S6 kinase only when phosphorylated at threonine 389. This antibody also detects p85 S6 kinase when phosphorylated at the analogous site (Thr412), and possibly S6KII phosphorylated at Thr401.

**Source/Purification:** Polyclonal antibodies are produced by immunizing rabbits with a synthetic phospho-peptide (KLH-coupled) corresponding to residues around Thr389 of human p70 S6 kinase. Antibodies are purified by protein A and peptide affinity chromatography.



◀ Western blot analysis of HeLa, COS, C6 and 3T3 cells, serum-starved overnight, then treated with insulin, λ-phosphatase or 20% serum as indicated. Upper panel probed with Phospho-p70 S6 Kinase (Thr389) Antibody #9205; lower panel probed with p70 S6 Kinase Antibody #9202.

Entrez-Gene ID # 6198  
Swiss-Prot Acc. # P23443

**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.

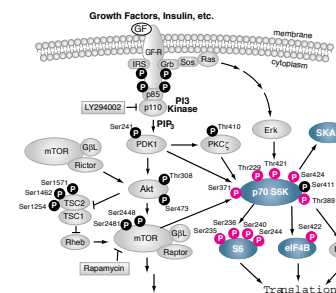
\*Species cross-reactivity is determined by Western blot.  
\*\*Anti-rabbit secondary antibodies must be used to detect this antibody.

**Recommended Antibody Dilutions:**  
Western Blotting 1:1000

For application specific protocols please see the web page for this product at [www.cellsignal.com](http://www.cellsignal.com).

- Companion Products:**
- Phospho-S6 Ribosomal Protein (Ser235/236) Antibody #2211
  - Phospho-S6 Ribosomal Protein (Ser240/244) Antibody #2215
  - p70 S6 Kinase Antibody #9202
  - p70 S6 Kinase Control Cell Extracts #9203
  - Phospho-p70 S6 Kinase (Thr421/Ser424) Antibody #9204
  - PhosphoPlus® p70 S6 Kinase (Thr389, Thr421/Ser424) Antibody Kit #9430
  - Anti-rabbit IgG, HRP-linked Antibody #7074
  - Prestained Protein Marker, Broad Range (Premixed Format) #7720
  - Biotinylated Protein Ladder Detection Pack #7727
  - 20X LumiGLO® Reagent and 20X Peroxide #7003
  - Phototope®-HRP Western Blot Detection System, Anti-rabbit IgG, HRP-linked Antibody #7071
  - Phospho-p70 S6 Kinase (Thr389) (1A5) Mouse mAb #9206
  - Phospho-p70 S6 Kinase (Ser371) Antibody #9208

Please visit [www.cellsignal.com](http://www.cellsignal.com) for a complete listing of recommended companion products.



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

**Selected Application References:**

Aoki, M. et al. (2001) A role of the kinase mTOR in cellular transformation induced by the oncoproteins P3k and Akt. *Proc. Natl. Acad. Sci. USA* 98, 136–141. Application: W.

Giallourakis, C. et al. (2000) Positive regulation of interleukin-4-mediated proliferation by the SH2-containing inositol-5'-phosphatase. *J. Biol. Chem.* 275, 29275–29282. Application: W.

Valentinis, B. et al. (2000) Insulin receptor substrate-1, p70S6K, and cell size in transformation and differentiation of hemopoietic cells. *J. Biol. Chem.* 275, 25451–25459. Application: W.

Shioi, T. et al. (2000) The conserved phosphoinositide 3-kinase pathway determines heart size in mice. *EMBO J.* 19, 2537–2548. Application: W.

Feng, L.X. et al. (2000) Stem cell factor/c-kit up-regulates cyclin D3 and promotes cell cycle progression via the phosphoinositide 3-kinase/p70S6 kinase pathway in spermatogonia. *J. Biol. Chem.* 275, 25572–25576. Application: W.

Prieto, A.L. et al. (2007) Localization and signaling of the receptor protein tyrosine kinase Tyro3 in cortical and hippocampal neurons. *Neuroscience* 150, 319–34. Application: W.

**Background References:**

- (1) Pullen, N. and Thomas, G. (1997) *FEBS Lett.* 410, 78–82.
- (2) Dunfer, A. et al. (1999) *Exp. Cell Res.* 253, 100–109.
- (3) Weng, Q.P. et al. (1998) *J. Biol. Chem.* 273, 16621–16629.
- (4) Pullen, N. et al. (1998) *Science* 279, 707–710.
- (5) Alessi, D. et al. (1998) *Curr. Biol.* 8, 69–81.
- (6) Polakiewicz, R.D. et al. (1998) *J. Biol. Chem.* 273, 23534–23541.
- (7) Finger, D.C. et al. (2002) *Genes Dev.* 16, 1472–1487.
- (8) Saitoh, M. et al. (2002) *J. Biol. Chem.* 277, 20104–20112.