Human/Mouse/Rat Phospho-p70 S6 Kinase (T421/S424) Antibody

Antigen Affinity-purified Polyclonal Rabbit IgG
Catalog Number: AF8965

DESCRIPTION

Species Reactivity	Human/Mouse/Rat
Specificity	Detects human, mouse and rat p70 S6 Kinase when phosphorylated at T421 and S424 and p85 S6K when phosphorylated at T444 and S447.
Source	Polyclonal Rabbit IgG
Purification	Antigen and protein A Affinity-purified
Immunogen	Phosphopeptide containing human, rat, and mouse p70 S6 Kinase T421/S424 sites
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

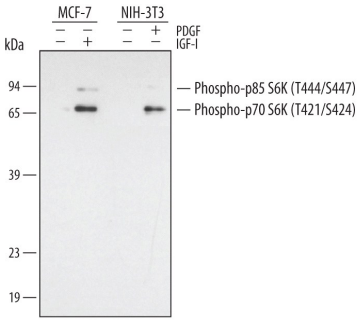
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	0.25 µg/mL	See Below
Simple Western	5 µg/mL	See Below

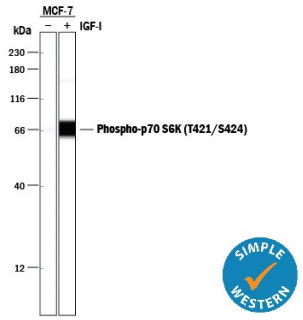
DATA

Western Blot



Detection of Human and Mouse Phospho-p70 S6 Kinase (T421/S424) by Western Blot. Western blot shows lysates of MCF-7 human breast cancer cell line untreated (-) or treated (+) with 100 ng/mL of Recombinant Human IGF-1 (Catalog # 291-G1) for 20 minutes and NIH-3T3 mouse embryonic fibroblast cell line untreated or treated with 100 ng/mL of Human PDGF (Catalog # 120-HD) for 20 minutes. PVDF membrane was probed with 0.25 µg/mL of Rabbit Anti-Human/Mouse/Rat Phospho-p70 S6 Kinase (T421/S424) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF8965), followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). Specific bands were detected for Phospho-p70 S6 Kinase (T421/S424) at approximately 70 and 85 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Simple Western



Detection of Human Phospho-p70 S6 Kinase (T421/S424) by Simple Western™. Simple Western lane view shows lysates of MCF-7 human breast cancer cell line untreated (-) or treated (+) with 100 ng/mL Recombinant Human IGF-I (Catalog # 291-G1) for 20 minutes, loaded at 0.2 mg/mL. A specific band was detected for Phospho-p70 S6 Kinase (T421/S424) at approximately 66 kDa (as indicated) using 5 µg/mL of Rabbit Anti-Human/Mouse/Rat Phospho-p70 S6 Kinase (T421/S424) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF8965). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

p70 S6 Kinase (p70S6K) is responsible for the phosphorylation of 40S ribosomal protein S6 and is ubiquitously expressed in human adult tissues (1). p70S6K is activated by serum stimulation and this activation is inhibited by wortmannin and rapamycin. p70S6K activity undergoes changes in the cell cycle and increases 20-fold in G1 cells released from G0 (2). p70S6K activation requires sequential phosphorylations at proline-directed residues in the putative autoinhibitory pseudosubstrate domain, as well as T389, a site phosphorylated by phosphoinositide-dependent kinase 1 (PDK1).

References:

1. Ferrari, S. *et al.* (1994) Crit. Rev. Biochem. Mol. Biol. **29**:385.
2. Edelmann, H.M. *et al.* (1996) J. Biol. Chem. **271**:963.

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