

DESCRIPTION

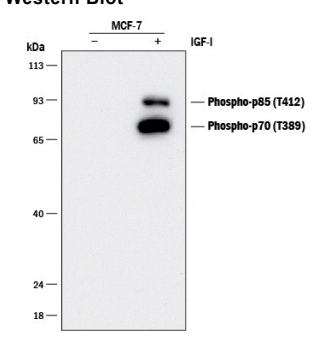
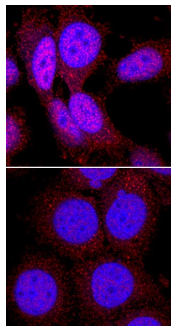
Species Reactivity	Human
Specificity	Detects human p70 S6 Kinase/p85 S6 Kinase when phosphorylated at T389/T412, respectively.
Source	Recombinant Monoclonal Rabbit IgG Clone # 1045C
Purification	Protein A or G purified from cell culture supernatant
Immunogen	Phosphopeptide containing the human p70 S6 Kinase T389 site
Formulation	Supplied as a solution in PBS containing BSA, Glycerol and Sodium Azide. 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	1:1000 dilution	See Below
Immunocytochemistry	1:50 dilution	See Below

DATA

<p>Western Blot</p>  <p>Detection of Human Phospho-p70 S6 Kinase (T389)/p85 S6 Kinase (T412) by Western Blot. Western blot 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 60 minutes. PVDF membrane was probed with 1:1000 dilution of Rabbit Anti-Human Phospho-p70 S6 Kinase (T389) Monoclonal Antibody (Catalog # MAB8963) followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). Specific bands were detected for Phospho-p70 S6 Kinase (T389) and Phospho-p85 S6 Kinase (T412) at approximately 70 and 85 kDa, respectively (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.</p>	<p>Immunocytochemistry</p>  <p>Phospho-p70 S6 Kinase (T389) in MCF-7 Human Cell Line. p70 S6 Kinase phosphorylated at T389 was detected in immersion fixed serum starved MCF-7 human breast cancer cell line untreated (lower panel) or treated with Recombinant Human IGF-I (Catalog # 291-G1) using Rabbit Anti-Human Phospho-p70 S6 Kinase (T389) Monoclonal Antibody (Catalog # MAB8963) at a 1:50 dilution for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Rabbit IgG Secondary Antibody (red; Catalog # NL004) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm and nuclei. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.</p>
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PREPARATION AND STORAGE

Shipping	The product is shipped with polar packs. 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	<ul style="list-style-type: none"> ● 12 months from date of receipt, -20 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after opening. ● 6 months, -20 °C under sterile conditions after opening.

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 (PKD1).

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.

PRODUCT SPECIFIC NOTICES

* Contains <0.1% Sodium Azide, which is not hazardous at this concentration according to GHS classifications. Refer to SDS for additional information and handling instructions.