

DESCRIPTION

Species Reactivity	Human/Mouse/Rat
Specificity	Detects human, mouse, and rat ribosomal protein S6 when dually phosphorylated at S235 and S236.
Source	Polyclonal Rabbit IgG
Purification	Antigen Affinity-purified
Immunogen	Phosphopeptide containing human, mouse and rat Ribosomal Protein S6 S235/S236 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 either lyophilized or as a 0.2 µm filtered solution in PBS.

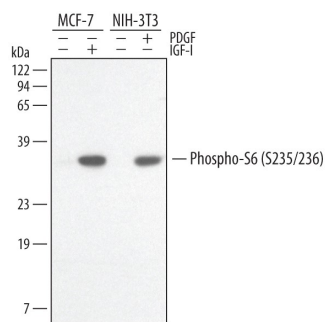
APPLICATIONS

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	0.5 µg/mL	See Below
Immunohistochemistry	5-15 µg/mL	See Below
Simple Western	5 µg/mL	See Below

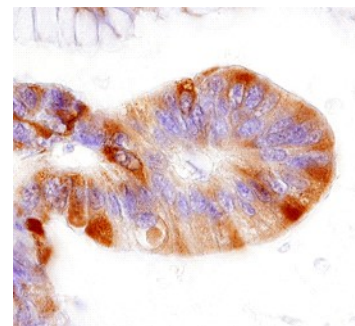
DATA

Western Blot



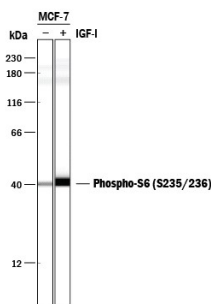
Detection of Human and Mouse Phospho-Ribosomal Protein S6 (S235/S236) by Western Blot. Western blot shows lysates of MCF-7 human breast cancer cell line untreated (-) or treated (+) with 100 ng/mL Recombinant Human IGF-1 (Catalog # 291-G1) for 20 minutes and NIH-3T3 mouse embryonic fibroblast cell line untreated or treated with 10 ng/mL Human PDGF (Catalog # 120-HD) for 20 minutes. PVDF membrane was probed with 0.5 µg/mL of Rabbit Anti-Human/Mouse/Rat Phospho-Ribosomal Protein S6 (S235/S236) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3918), followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). A specific band was detected for Phospho-Ribosomal Protein S6 (S235/S236) at approximately 32 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunohistochemistry



Ribosomal Protein S6 in Human Colon Cancer Tissue. Ribosomal Protein S6 was detected in immersion fixed paraffin-embedded sections of human colon cancer tissue using 5 µg/mL Rabbit Anti-Human/Mouse/Rat Phospho-Ribosomal Protein S6 (S235/S236) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3918) overnight at 4 °C. Tissue was stained with the Anti-Rabbit HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS005) and counterstained with hematoxylin (blue). Specific labeling was localized to vascular endothelial cell within glomeruli. View our protocol for [Chromogenic IHC Staining of Paraffin-embedded Tissue Sections](#).

Simple Western



Detection of Human Phospho-Ribosomal Protein S6/RPS6 (S235/S236) 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-1 (Catalog # 291-G1) for 20 minutes, loaded at 0.5 mg/mL. A specific band was detected for Phospho-Ribosomal Protein S6/RPS6 (S235/S236) at approximately 40 kDa (as indicated) using 5 µg/mL of Rabbit Anti-Human/Mouse/Rat Phospho-Ribosomal Protein S6/RPS6 (S235/S236) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3918). 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

40S ribosomal protein S6 is the major substrate of protein kinases, particularly p70 S6 kinase, in eukaryotic ribosomes. S6 phosphorylation at S235, S236, S240, and S244 upregulates the translation of mRNAs containing an oligopyrimidine tract at their transcriptional start sites. This phosphorylation is stimulated by growth factors, tumor promoting agents, and other mitogens.