Monoclonal Antibody to Phosphothreonine (incl. pos. control) - Purified

Catalog No.: AM00121PU-N
Quantity: 0.1 mg

Background: Phosphorylation and dephosphorylation of cellular proteins are central steps in transducing extracellular signals to the cell nucleus. Phosphorylated epitopes may serve as docking sites for the assembly of protein complexes or may alter the 3-dimensional protein structure thus modulating enzymatic activity or the ability to undergo protein-protein-interactions. Modification of proteins on serine residues is mediated by serine/threonine kinases.

Host / Isotype: Mouse / IgG1
Clone: 14B3
Immunogen: Synthetic phosphopeptide conjugated to KLH

Format: State: Lyophilized purified Ig fraction
Purification: Subsequent Thiophilic Adsorption and Size Exclusion Chromatography.
Buffer System: 1 ml 2 x PBS containing 0.09% Sodium Azide, PEG and Sucrose.
Reconstitution: Restore with 1 ml H2O (15 min, RT).

Applications: ELISA: 0.05 µg/ml.
Immunoprecipitation: 1-10 µg per 10e6 pervanadate-treated A431 cells.
Western Blot (Immunoblotting): 1 µg/ml for HRPO/ECL detection.
Recommended blocking buffer: BSA/Tween 20 based blocking buffer. DO NOT USE MILK OR CASEIN FOR BLOCKING!

Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.

Specificity: This antibody recognizes a broad range of threonine-phosphorylated proteins in crude cell extracts, providing a valuable tool for non-radioactive phosphothreonine detection.
Species Reactivity: Tested: Human, Mouse, Rat and Dog.

Add. Information: This product contains a Positive Control (for details see "Protocols").

Storage: Store lyophilized (preferably in a desiccator) at -20°C and reconstituted (aliquote and freeze in liquid nitrogen) at -80°C. Avoid repeated freezing and thawing. Thaw aliquots at 37°C. Thawed aliquots may be stored at 2-8°C up to 1 week. Shelf life: one year from despatch.

Protocols: Positive Control: pSer / pThr Molecular Weight Marker

Formulation:
The pSer/pThr molecular weight marker contains rabbit muscle phosphoproteins isolated by Fe3+/IDA - affinity chromatography. Proteins are lyophilized from
PBS/NaF/PEG/Sucrose/ Bromophenolblue and Na - azide. After reconstitution the solution contains 0.09% Na-azide.

**Stability:**
Reconstitute by addition of 200 µl H2O. After complete solubilization add 200 µl 2x SDS-PAGE sample buffer, mix and incubate at 90°C for 5 min.

**Application:**
The pSer/pThr molecular weight marker is recommended for immunoblot applications. Use 20µl molecular weight marker per lane. Note: Use BSA based blot incubation buffers. Milk, Casein and Blotto might interfere with antibody - antigen interaction.

**Storage:**
Aliquote and store frozen.
Avoid repeated freeze/thaw cycles.
Shelf life: one year from despatch.

**Pictures:**

Figure 1. Phosphothreonine Detection: A431 cell Lysates probed with Lane 1: mab 14B3 (IgG), 1 µg/ml. Lane 2: mab 4D11 (IgM), 1 µg/ml. Lane 3: mab 1E11 (IgG), 1 µg/ml.