Phosphotyrosine 7x Sampler kit (incl. pos. control) - Purified

Catalog No.: AM00129PU-N
Quantity: 7 x 25 µg
Background: Phosphorylation and dephosphorylation of cellular proteins are central steps in transducing extracellular signals to the 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 tyrosine residues is mediated by protein tyrosin kinases. Tyrosine phosphorylation may alter the biological activity or mediate the assembly of protein complexes via the interaction of phosphotyrosine residues with SH2 or PTB domains. Antibodies direct against phosphorylated epitopes recognize the phosphorylated amino acid in the context of the surrounding amino acid sequence. Recognition is therefore dependent on 2 criteria: 1) phosphorylation and 2) the surrounding amino acid motif. If one of the two criteria is not fulfilled, the antibody will not detect the phosphorylation site. Since the amino acid sequence varies between different phosphorylation sites, certain proteins - though phosphorylated - may not be detected by the antibody. Phosphorylation patterns in a given cell extract may differ when probed with different antibodies due to sequence specificity.

Host: Mouse
Specificity: The different monoclonal antibodies recognize phosphorylated tyrosine. Species: Human, Mouse, Rat, Dog. Other species not tested.
Add. Information: The Phosphotyrosine Detection Kit contains 7 different phosphotyrosine specific monoclonal antibodies.

AM03182PU-25 (Clone 1F9)
AM00123PU-25 (Clone 2A5)
AM00124PU-25 (Clone 2C8)
AM00125PU-25 (Clone 3B12)
AM00126PU-25 (Clone 9F1)
AM00127PU-25 (Clone 9H8)
AM00128PU-25 (Clone 16F4)

This product contains a positive control for immunoblot applications (for details see "Protocols").

Protocols: Positive Control for Immunoblot Applications. Phosphotyrosine Molecular Weight Marker:
**Formulation:**
Phosphotyrosine modified standard proteins lyophilized from PBS/0.1% SDS/PEG/Sucrose/Malachitgreen and Na-azide. After reconstitution the solution contains 0.09% Na-azide.
The following standard proteins were modified with phosphotyrosine: galactosidase (116kD), phosphorylase A (98kD), BSA (67kD), ovalbumin (46kD), carbonic anhydrase (32kD), and soybean trypsin inhibitor (24kD).

**Reconstitution:**
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 phosphotyrosine molecular weight marker is recommended for immunoblot applications. Use 20 µl of the phosphotyrosine molecular weight marker per lane (mini gel).

The individual proteins of the marker are recognized by the following commercially available monoclonal antibodies:
- 2C8, 1F9, 3B12, 9F1, 9H8, 16F4
- 4G10 (Upstate/Millipore)
- PY20 (BD)
- PT66 (Sigma)

**Pictures:**
Phosphotyrosine Detection Lysates of pervanadate-treated A431 cells were probed with: