Anti-Phosphotyrosine

Mouse Monoclonal

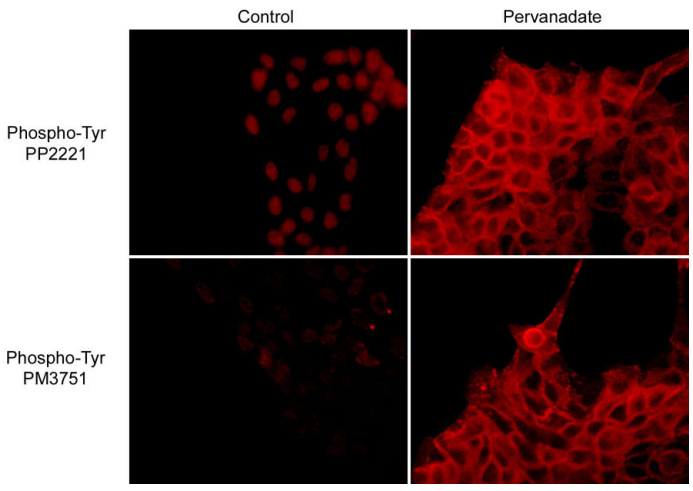
Cat. # PM3751
Size 100 µl

Background

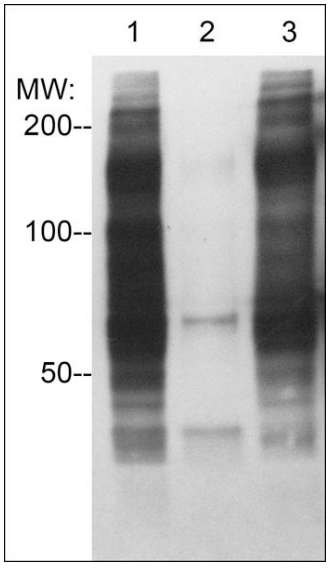
Phosphorylation of specific tyrosine residues is an important post-translational modification for regulating the activity of most proteins. Stimulation of a variety of cell signaling pathways activates the receptor and non-receptor tyrosine kinases that mediate these protein modifications. Antibodies that can detect phosphotyrosine residues are excellent tools for characterizing changes in the post-translational state of a broad range of phosphotyrosine-containing proteins. Immunoprecipitation of proteins of interest, followed by detection of phosphotyrosine using anti-phosphotyrosine antibody is commonly used to correlate changes in tyrosine phosphorylation state with alterations in protein activity.

Background References

Ross, A.H. et al. (1981) Nature 294:654.
Hunter T.(1987) Cell. 50(6):823.
Wang, J.Y.J. (1988) Anal. Biochem 172:1.



Immunocytochemical labeling of phosphotyrosine in control and pervanadate-treated A431 cells. The cells were labeled with rabbit polyclonal anti-Phosphotyrosine (PP2221) and mouse monoclonal anti-Phosphotyrosine (PM3751), then the antibodies were detected using appropriate secondary antibodies conjugated to Cy3.



Western blot of HeLa cells treated with pervanadate (1 mM) for 30 min. Phosphotyrosine containing proteins were immunoprecipitated with rabbit polyclonal anti-Phosphotyrosine:Agarose (Lane 1) or with no antibody agarose beads (Lane 2), and blots were made that included the whole lysate (Lane 3). The blots were probed with mouse monoclonal anti-Phosphotyrosine (PM3751) to detect phosphotyrosine containing proteins.

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Rev10/14/2019

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Immunogen

Clone M375 was generated from tyrosine phosphorylated proteins purified from pervanadate treated A431 cells.

Buffer and Storage

Mouse monoclonal antibody purified with protein A chromatography is supplied in 100µl phosphate-buffered saline, 50% glycerol, 1 mg/ml BSA, and 0.05% sodium azide. Store at –20°C. Stable for 1 year.

Applications

WB	1:1000
ELISA	1:2000
ICC	1:100
IP	1:100

Species Reactivity

Hu, Rt, Ms, Ck

Isotype: IgG2b

End user should determine optimal dilution for their particular applications and experiments.
Western blot membranes were incubated with diluted antibody in 5% non-fat milk, Tris buffer, 0.04% Tween20 for 1 hour at room temperature.
Abbreviations: E = ELISA, ICC = immunocytochemistry, IHC = immunohistochemistry, IP = immunoprecipitation, MS = mass spectrometry, WB = western blot
Hu = Human, Ms = Mouse, Rt = Rat, Ck = Chicken, F = Frog, B = Bovine

Specificity

This antibody detects phosphotyrosine-containing proteins on SDS-PAGE immunoblots of EGF treated A431 cells, as well as pervanadate treated A431, HeLa, Jurkat, and endothelial cells.

*All molecular weights (MW) are confirmed by comparison to MW standards and to western blot mobilities of known proteins with similar MW.
"Native" western blot utilizes non-reducing sample buffer (no mercaptoethanol or SDS), normal SDS-PAGE gel electrophoresis, and no methanol in transfer buffers.

Related Products

PK6330 Phospho-Tyrosine, Serine, Threonine Antibody Sampler Kit

PM3801 Anti-Phosphoserine/threonine Mouse Monoclonal

PP2221 Anti-Phosphotyrosine Rabbit Polyclonal

PP2551 Anti-Phosphoserine/threonine Rabbit Polyclonal

PP4651 Anti-Phosphotyrosine:Agarose Rabbit Polyclonal

PP4641 Anti-Phosphothreonine Rabbit Polyclonal

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