sigma-aldrich.com

3050 Spruce Street, St. Louis, MO 63103 USA Tel: (800) 521-8956 (314) 771-5765 Fax: (800) 325-5052 (314) 771-5757 email: techservice@sial.com sigma-aldrich.com

Product Information

Anti-Phosphotyrosine antibody, Mouse monoclonal clone PT-66, purified from hybridoma cell culture

Catalog Number P5872

Product Description

Monoclonal Anti-Phosphotyrosine (mouse IgG1 isotype) is derived from the PT-66 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with phosphotyrosine-BSA conjugate. The isotype is determined by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Catalog Number ISO2).

Monoclonal Anti-Phosphotyrosine is specific for phosphorylated tyrosine both as the free amino acid or when conjugated to carriers such as BSA or KLH. It does not react with non-phosphorylated tyrosine nor other phosphorylated amino acids, including serine and threonine, nor does it react with phosphorylated molecules such as AMP or ATP. Monoclonal Anti-Phosphotyrosine may be used for the identification of proteins containing phosphorylated tyrosine in immunohistochemisty, immunocytochemistry,^{8,12} flow cytometry, immunoprecipitation,⁵ immunoblotting,^{5,8-10,13} ELISA,¹¹ RIA. or for immunoaffinity isolation.¹⁻⁵

Protein phosphorylation is a basic signaling mechanism that modifies protein function in eukaryotic cells. Post-translational modifications of proteins are important for their activity and stability. Several distinct modifications of cellular proteins have been identified, among them protein phosphorylation is the most abundant. Serine, threonine, and tyrosine are the major phosphorylated amino acids in proteins. Tyrosine phosphorylation is a rare post-translational event in normal tissues, accounting for only 0.03% of phosphorylated amino acids. However, this phosphorylation increases several fold by various activation signals and the process is mediated by protein tyrosine kinases.^{6,7} Protein-tyrosine kinases (PTKs) are enzymes that catalyze the transfer of γ -phosphate of ATP to tyrosine residues of protein substrates.

These enzymes are divided into two major families, receptor tyrosine kinases (RTKs) and non-receptor tyrosine kinases (NRTKs).^{6,7} The RTK family includes many growth factor receptors such as the insulin-R, EGF-R, PDGFR,FGF-R, and NGF receptor. These receptors possess an extracellular domain that is responsible for ligand binding, a transmembrane domain, and an intracellular domain that contains the catalytic activity and a regulatory sequence. The NRTK family consists of modular domains responsible for subcellular targeting and regulation of catalytic activity. This family includes Src, Abl, Fak, Jak, and many others. The PTKs are responsible for many biological processes like cell cycle, proliferation, oncogenesis, and development.

Reagent

The antibody is supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

Antibody concentration: ~2 mg/mL

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

For continuous use, store at 2–8 °C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing is not recommended. Storage in "frost-free" freezers is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discarded if not used within 12 hours.

Product Profile

A working concentration of 0.25–0.5 μ g/mL is determined by immunoblotting, using total cell extract of human platelets.

A working concentration of $0.5-1 \ \mu g/mL$ is determined by indirect ELISA using microwell plates coated with phosphotyrosine conjugated to BSA (Catalog Number P3967, 10 $\mu g/mL$).

<u>Note</u>: In order to obtain best results in different techniques and preparations we recommend determining optimal working dilution by titration test.

References

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