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Product Information

Anti-Phosphotyrosine antibody, Mouse monoclonal clone pT-154, purified from hybridoma cell culture

Product Number P1869

Product Description

Anti-Phosphotyrosine antibody, Mouse monoclonal (mouse IgG2b isotype) is derived from the pT-154 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with a tyrosine phosphorylated peptide. The isotype is determined by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents, Product Number ISO2.

Anti-Phosphotyrosine antibody, Mouse monoclonal reacts specifically with phosphorylated tyrosine and does not recognize non-phosphorylated tyrosine, phosphorylated serine or threonine, ATP or AMP, as determined by competitive ELISA. The product is used in immunoblotting immunohistochemistry, ELISA and competitive ELISA applications.

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.^{1,2}

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). ^{1,2} The RTK family includes many growth factor receptors such as the insulin-R, EGF-R, PDGF-R, FGF-R and NGF receptor. These receptors possess an extra-cellular domain that is responsible for ligand binding, a trans-membrane 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. They are tightly regulated by other kinases and by autophosphorylation activity. Monoclonal antibodies specific for phosphotyrosine are essential tools for the study of tyrosine phosphorylation in post-translational modification in many biological processes.

Reagent

Supplied at ~2 mg/mL in a solution in 0.01 M phosphate buffered saline pH 7.4, containing 15 mM sodium azide.

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. Upon initial thawing, freeze the solution in working aliquots for extended storage. Avoid repeated freezing and thawing to prevent denaturing the antibody. Storage in "frost-free" freezers is also not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilutions should be discarded if not used within 12 hours.

Product Profile

 $\frac{Immunoblotting}{Immunoblotting}: a working dilution of 2-4 \ \mu g/ml is determined using total extract of A431 cells stimulated by EGF.$

Note: In order to obtain best results in different techniques and preparations we recommend determining optimal working dilutions by titration test.

References

- Hubbard, S.R., et al., J. Biol. Chem., 273, 11987-11990 (1998).
- 2. Zou, X., et al., J. Biol. Chem., **274**, 18141-18144 (1999).

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