MONOCLONAL ANTI-PHOSPHO-RETINOBLASTOMA (RB) (pS^{795}) CLONE RB-10
Purified Mouse Immunoglobulin

Product Number **R 6878**

ProductInformation

Product Description

Monoclonal Anti-phospho-Retinoblastoma (Rb) (pS^{795}) (mouse IgG1 isotype) is derived from the RB-10 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from a BALB/c mouse immunized with a synthetic phospho peptide corresponding to the C-terminal amino acids 791-804 (pS^{795}) of human retinoblastoma protein, conjugated to KLH. The isotype is determined using Sigma ImmunoType™ Kit (Sigma ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Sigma ISO-2).

Monoclonal Anti-phospho-Retinoblastoma (Rb) (pS^{795}) reacts specifically with Rb phosphorylated at serine 795, and does not react with nonphosphorylated Rb, nor with retinoblastoma-related proteins p107 and p130. The product is useful in ELISA, immunoblotting (110 kDa) and immunocytochemistry (3% paraformaldehyde fixation, 0.15% Triton X-100 permeabilization). Reactivity has been observed with human, bovine, hamster, rat and mouse Rb.

Before any gene can be expressed by translation into amino acids, it must first be transcribed in the nucleus into messenger RNA (mRNA), which stores a complementary copy of the DNA code. The initial event in this process is the binding of specific proteins to the enhancer and the promoter. The binding of these proteins depends on their recognition of specific nucleotide sequences. Whereas some DNA-binding proteins are "positive regulators" that stimulate transcription, others are "negative regulators" that block transcription. Retinoblastoma proteins p107, p110 (also designated Rb, or pRb) and p130, referred to collectively as pocket proteins, constitute a nuclear protein family that share a common structural unit (the pocket) dedicated to binding certain transcription factors and thereby regulating cellular proliferation.[1-5] All known pocket proteins bind certain viral oncoproteins, i.e., papovirus T antigen, adenovirus E1A, and Human Papilloma virus E7.[2,3] Rb exerts its function by repressing the transcription of cellular genes required for DNA replication and cell division.[6] Rb has three functionally distinct binding domains, and interacts with critical regulatory proteins including the E2F family of transcription factors, c-Abl tyrosine kinase and proteins with conserved LXCXE motif.[7,8] Members of the E2F transcription factor family are targets of Rb during G_1 phase,[3,6,7,9] where Rb protein is responsible for effecting reversible G1 growth arrest.[3,9] Rb function is modulated by the binding of MDM2, viral oncoproteins, cyclin D overexpression, Cdk4 amplification, and loss of p16^{INK4a} arrest.[3,9] The binding to and inactivation of E2F proteins by Rb is regulated to a certain extent by cyclin dependent kinase-mediated phosphorylation. Unphosphorylated Rb, p107, and p130 inhibit the activity of the E2F family of transcription factors thereby halting cell cycle progression. Subsequent serine/threonine phosphorylation by G_1 cyclin dependent kinases results in inactivation and release of E2F.[10] Rb can be phosphorylated at multiplicity of sites *in vitro* by Cdc2, Cdk2 and Cdk4/Cdk6 kinase complexes.[11,12] Differential phosphorylation has been shown to modulate Rb function, by affecting the ability to interact with its various partner proteins.[12,13] Both serine 608, 780, 795, 807 and 811, and threonine 821 and/or 826, have been identified among the sites that are phosphorylated. Phosphorylation of pS^{795} is required to inactivate Rb-imposed growth suppression.[12] Induction of the Cdk inhibitors p16^{INK4a} and p21^{WAF1}, effectively prevents phosphorylation of Rb family proteins as do the Rb-specific phosphatases.[9,14] Antibodies reacting specifically with phosphorylated retinoblastoma protein are useful tools in the study of the detailed mechanisms of the control of transcription in intracellular pathways, and its essential roles during developmental and pathological processes.

Reagent

Monoclonal Anti-phospho-Retinoblastoma (Rb) (pS^{795}) is supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide.

Antibody Concentration: Approx. 2 mg/ml.

Precautions and Disclaimer
Due to the sodium azide content a material safety sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution.
Consult the MSDS for information regarding hazardous and safe handling practices.

Storage/Stability
For continuous use, store at 2 °C to 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 2 to 4 µg/ml is determined by immunoblotting using a whole cell extract of human melanoma cell line G-361.

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

References

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