Technical Data Sheet

Purified Mouse Anti-Human Retinoblastoma Protein

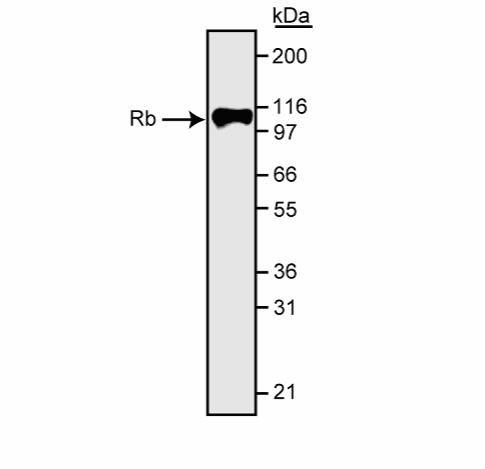
Product Information

Material Number:	**554144**
Alternate Name:	RB
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	XZ55
Immunogen:	Recombinant carboxy-terminal human RB protein aa. 387-928
Isotype:	Mouse IgG1
Reactivity:	QC Testing: Human Reported: Mouse, Chicken, Xenopus, Quail
Target MW:	105-116 kDa
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The retinoblastoma gene encodes a nuclear phosphoprotein (Rb or p110[Rb]) which is expressed in most normal cells of vertebrates and acts as a tumor suppressor gene product. An underphosphorylated form of RB is mainly found in resting or fully differentiated cells, whereas the hyperphosphorylated form is present in proliferating cells. Only the underphosphorylated form of RB binds specifically to viral oncogenes such as SV40 large T, adenviral EIA and HPV-E7. This interaction may partially contribute to the transforming activity of these viral oncoproteins. Rb also interacts with several cyclins including A, D, and E as well as the transcriptional activator E2F. The importance of these interactions for the biological function of Rb is still being elucidated.

XZ55 recognizes an epitope located between amino acids 443-622 of human Rb. It cross-reacts with chicken, mouse and Xenopus Rb, and a putative quail Rb.A recombinant carboxy-terminal (amino acids 387-928) human Rb protein was used as immunogen.



Western blot analysis of Rb in MOLT-4 human leukemia cell lysate. *The XZ55 antibody (Cat. No. 554144) recognizes phosphorylated and underphosphorylated forms of Rb (~110-116 kDa).*

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at 4°C.

Application Notes

Application

Western blot	Routinely Tested
Immunoprecipitation	Reported
Gel shift	Reported

Recommended Assay Procedure:

Application includes western blot analysis (1-2 µg/ml). Rb migrates as multiple closely-spaced bands between approximately 110-116 kDa when sized on denaturing polyacrylamide gels (i.e. by SDS-PAGE). The different bands represent different Rb phosphorylation states, the higher molecular weight bands are more highly phosphorylated than the lower molecular weight bands. The level of phosphorylation is cell cycle

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dependent, and may also be cell type dependent (not all forms are seen in all cell types that express Rb). Gel conditions influence the actual number of bands observed. In cases where optimal band separation is desired, use a 7 to 10% non-gradient long (≥ 12 inches) gel. MOLT-4 human leukemia cells (ATCC CRL-1582) are suggested as a positive control. Additional applications not routinely tested at BD Biosciences Pharmingen include immunoprecipitation (1-2 µg/ml) and electrophoretic mobility gel shift assays (EMSA).

Suggested Companion Products

Catalog Number	Name	Size	Clone
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

References

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