Datasheet for ABIN675294

anti-Retinoblastoma 1 antibody (pSer780)

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Overview

Quantity:	100 µL
Target:	Retinoblastoma 1 (RB1)
Binding Specificity:	pSer780
Reactivity:	Human, Rat
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	This Retinoblastoma 1 antibody is un-conjugated
Application:	Western Blotting (WB), ELISA, Immunohistochemistry (Paraffin-embedded Sections) (IHC (p)), Immunofluorescence (Cultured Cells) (IF (cc)), Immunofluorescence (Paraffin-embedded Sections) (IF (p)), Flow Cytometry (FACS), Immunohistochemistry (Frozen Sections) (IHC (fro))

Product Details

Immunogen:	KLH conjugated synthetic phosphopeptide derived from human Rb around the phosphorylation site of (Ser780)
Isotype:	IgG
Cross-Reactivity:	Human, Rat
Predicted Reactivity:	Mouse,Dog,Cow,Chicken
Purification:	Purified by Protein A.

Target Details

Target:	Retinoblastoma 1 (RB1)

Target Details

Alternative Name:	Rb/p105-Rb (RB1 Products)
Background:	Synonyms: RbSer780, OSRC, P105 RB, P105RB, PP105, PP110, pRb, RB 1, RB1, RB1 protein, Retinoblastoma 1 including osteosarcoma, Retinoblastoma 1, Retinoblastoma associated protein, Including osteosarcoma, Osteosarcoma, p105-Rb, Rb, RB_HUMAN, Retinoblastoma suspectibility protein, Retinoblastoma-associated protein, Retinoblastoma related osteosarcoma, Retinoblastoma susceptibility gene. Background: Nuclear Marker.The protein encoded by this gene is a negative regulator of the cell cycle and was the first tumor suppressor gene found. The encoded protein also stabilizes constitutive heterochromatin to maintain the overall chromatin structure. The active, hypophosphorylated form of the protein binds transcription factor E2F1. Defects in this gene are a cause of childhood cancer retinoblastoma (RB), bladder cancer, and osteogenic sarcoma.
Gene ID:	5925
Pathways:	Cell Division Cycle, Intracellular Steroid Hormone Receptor Signaling Pathway, Mitotic G1-G1/S Phases, DNA Replication, Maintenance of Protein Location, Synthesis of DNA, Autophagy

Application Details

Application Notes:	WB 1:300-5000
	ELISA 1:500-1000
	FCM 1:20-100
	IHC-P 1:200-400
	IHC-F 1:100-500
	IF(IHC-P) 1:50-200
	IF(IHC-F) 1:50-200
	IF(ICC) 1:50-200

Restrictions:	For Research Use only

Handling

Format:	Liquid
Concentration:	1 μg/μL
Buffer:	0.01M TBS(pH 7.4) with 1 % BSA, 0.02 % Proclin300 and 50 % Glycerol.
Preservative:	ProClin
Precaution of Use:	This product contains ProClin: a POISONOUS AND HAZARDOUS SUBSTANCE, which should be

Handling

	handled by trained staff only.
Storage:	4 °C,-20 °C
Storage Comment:	Shipped at 4°C. Store at -20°C for one year. Avoid repeated freeze/thaw cycles.
Expiry Date:	12 months

Publications

Product cited in:	Béguelin, Rivas, Calvo Fernández, Teater, Purwada, Redmond, Shen, Challman, Elemento, Singh, Melnick: "EZH2 enables germinal centre formation through epigenetic silencing of CDKN1A and an Rb-E2F1 feedback loop." in: **Nature communications**, Vol. 8, Issue 1, pp. 877, (2018) (PubMed).

Validation report #029795 for Western Blotting (WB)

Successfully validated (Western Blotting (WB))

by Alamo Laboratories Inc

Report Number: 029795

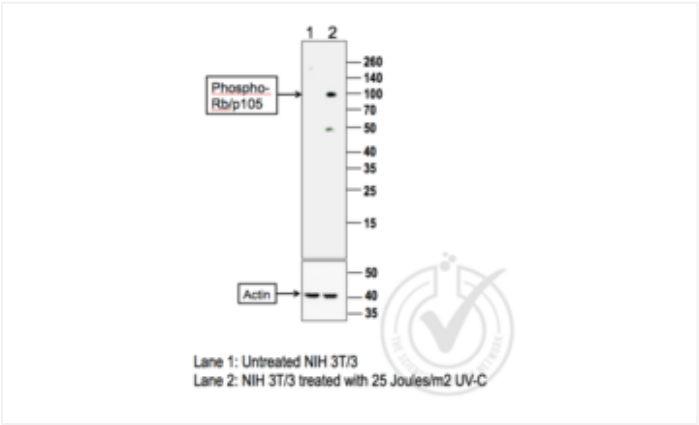
Date: Aug 21 2014

Target:	Retinoblastoma 1 (RB1) (pSer780)
Lot Number:	120508
Method validated:	Western Blotting (WB)
Positive Control:	NIH/3T3 cells irradiated with 25 Joules/m2 UV-C
Negative Control:	Non-irradiated NIH/3T3 cells
Notes:	A strong band was observed in the positive control sample at the correct molecular weight. An additional band was also observed in the positive sample at a lower molecular weight which may represent non-specific binding. No bands were observed in the negative control sample.
Primary Antibody:	- Antigen: Retinoblastoma 1 (RB1) (pSer780) - Catalog number: ABIN675294 - Supplier: Bioss - Supplier catalog number: bs-1347R - Lot number: 120508 - Antibody Dilution: 1:100
Secondary Antibody:	- Antigen: Goat Anti-Rabbit IgG (H + L)-HRP Conjugate - Supplier: Bio-Rad - Catalog number: #170-6515 - Lot number: L170-6515 - Antibody Dilution: 1:10,000
Controls:	• Positive control: NIH/3T3 cells irradiated with 25 Joules/m2 UV-C* • Negative control: Non-irradiated NIH/3T3 Cells • *UV-C Treatment: Sub-confluent cells (~50% confluent) in T150 flask were trypsinized and transferred to 12 cell-culture dishes (100mmx10mm) and returned to incubator for 24 hours. Thereafter, the growth medium was siphoned and cells were irradiated with UV-C @ 25 Joules/m2 using Hoefer UVC 500 Ultraviolet crosslinker. After irradiation, the complete growth medium was added to dishes and cells were incubated for another 16 hours. Thereafter, cells were collected by adding 0.5ml of Modified RIPA (with protease and phosphatase inhibitor cocktails) to each dish and protein extract prepared.
Protocol:	• 1. The cell extracts were heated at 95°C for 5 minutes in 1X SDS Sample Buffer containing 1% SDS and 1.25% β-mercaptoethanol. • 2. 20 µL of heated culture-media were loaded and resolved on 8-16% SDS-polyacrylamide gel. • 3. The Thermo Scientific - Spectra Multicolor Broad Range (Cat # 26634) were used as molecular mass markers. • 4. Proteins were then transferred onto PVDF membrane by wet transfer and protein transfer was confirmed with Ponceau-S staining.

- 5. The PVDF membrane was incubated with 25 mL of blocking buffer [Tris Buffered Saline, pH 7.4 plus 0.1% TW20 (TBST)] containing 5% (W/V) BSA at room temperature for 1 hour.
- 6. The membrane was rinsed with TBST once.
- 7. The membrane was immersed with the protein side up in the primary antibody solution in TBST containing 5% (W/V) BSA and incubated for 20 hours at 4°C.
- 8. The membrane was rinsed in TBST thrice for 5 minutes each.
- 9. The membrane was incubated in the HRP-conjugated secondary antibody solution in TBST containing 5% (W/V) BSA and incubated for 1 hour at room temperature (~26°C) with gentle agitation.
- 10. The membrane was rinsed thrice TBST thrice for 5 minutes each.
- 11. The membrane was rinsed in TBS twice for 30 seconds each.
- 12. Signals were detected with ECL-2 Substrate. The blot was scanned for 50 minutes.
- 13. The membrane was rinsed three times TBST.
- 14. Incubated in Acidic Glycine Stripping Buffer at room temperature with gentle agitation for 3 times, 10 minutes each.
- 15. The membrane was washed in TBST 2 times for 10 minutes each.
- 16. Repeated Steps 5-12 with the loading control antibody (for Anti-actin) and its matching secondary antibody.

Experimental Notes:	- No experimental challenges noted.

Image for Validation report #029795



Validation image no. 1 for anti-Retinoblastoma 1 (RB1) (pSer780) antibody (ABIN675294)

Figure 1: Western Blot for RB1 pSer780. Grey arrowhead indicates the expected molecular weight of ~105 kDa.