

## PRDM1/Blimp-1

CONTACT INFORMATION:	Monoclonal Antibodies Unit. Centro Nacional de Investigaciones Oncológicas
STATUS:	Validated
TYPE:	mouse anti human
CLONE NAME:	ROS195
PROTEIN:	Human full length PRDM1
PROTEIN WEB:	<a href="http://www.ncbi.nlm.nih.gov/omim/603423">http://www.ncbi.nlm.nih.gov/omim/603423</a>
ANTIGEN USED:	GST-PRDM1 recombinant protein
FUSION PARTNER:	NS1/Ag4-1 (NS1) cells
ISOTYPE:	IgG1
SPECIES REACTIVITY:	human and mouse
PREPARATION AND STORAGE:	Aliquot and store at 4C. Do not freeze
APP RECOMMENDED:	IHQ-paraffin, IF, WB, IP, Flow cytometry
APP NO TESTED:	IHQ-frozen

### DESCRIPTION

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B lymphocyte-induced maturation protein-1 (Blimp-1) is a 98-kDa protein containing five Kruppel-type zinc fingers that confer sequence specific DNA binding. Based on studies in B cell lines, Blimp-1 has been postulated to be a master regulator of terminal B cell differentiation. In the BCL-1 lymphoma model of differentiation from a mature B cell to a plasma cell, ectopic expression of Blimp-1 is sufficient to cause terminal differentiation evidenced by loss of surface Ig, IgM secretion, expression of syndecan-1 on the cell surface, and cessation of cell division.

### PUBLICATION DESCRIBING ANTIBODY CHARACTERIZATION/VALIDATION

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Garcia JF, Roncador G, Garcia JF, Sanz AI, Maestre L, Lucas E, Montes-Moreno S, Fernandez Victoria R, Martinez-Torrecladrara JL, Marafioti T, Mason DY, Piris MA. PRDM1/BLIMP-1 expression in multiple B and T-cell lymphoma. Haematologica. 2006. Apr; 91(4):467-74.

### REFERENCES

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Liu YY, Leboeuf C, Shi JY, Li JM, Wang L, Shen Y, Garcia JF, Shen ZX, Chen Z, Janin A, Chen SJ, Zhao WL. Rituximab plus CHOP (R-CHOP) overcomes PRDM1-associated resistance to chemotherapy in patients with diffuse large B-cell lymphoma. Blood 2007. Jul1; 110(1): 339-44.

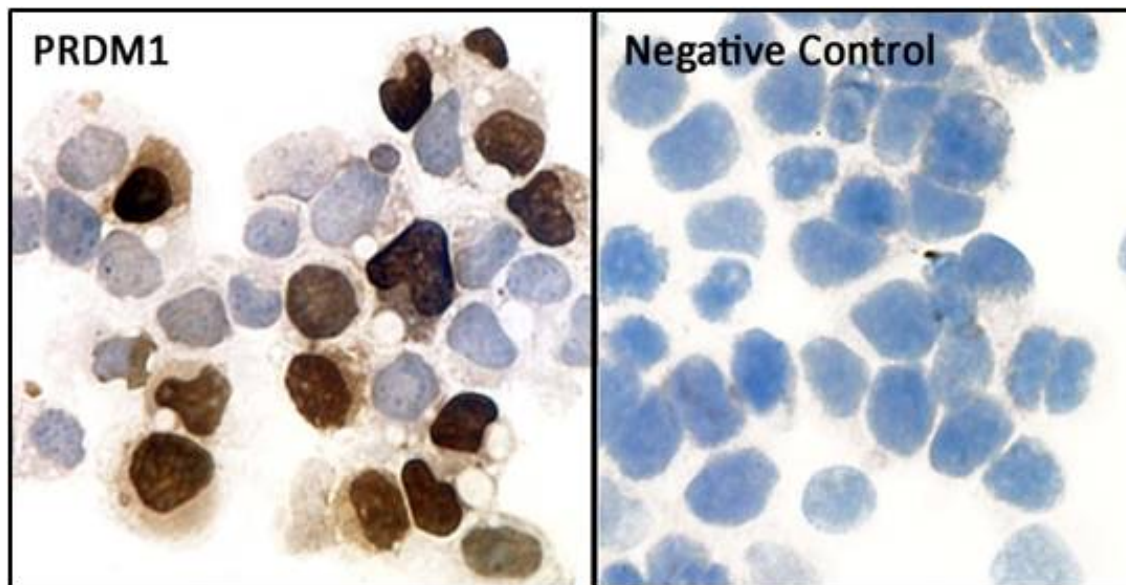
Zhao WL, Liu YY, Zhang QL, Wang L, Leboeuf C, Zhang YW, Ma J, Garcia JF, Song YP, Li JM, Shen ZX, Chen Z, Janin A, Chen SJ. PRDM1 is involved in chemoresistance of T-cell lymphoma and down-regulated by the proteasome inhibitor. Blood 2008. Apr 1;111(7): 3867-71.

Nie K, Gomez M, Landgraf P, Garcia JF, Liu Y, Tan LH, Chadburn A, Tuschl T, Knowles DM, Tam W. MicroRNA-mediated down-regulation of PRDM1/Blimp-1 in Hodgkin/Reed-Sternberg cells: a potential pathogenetic lesion in Hodgkin lymphomas. Am J Pathol. 2008. Jul; 173(1): 242-52.

Garcia JF, Roncador G, Garcia JF, Sanz AI, Maestre L, Lucas E, Montes-Moreno S, Fernandez Victoria R, Martinez-Torrecedrara JL, Marafioti T, Mason DY, Piris MA. PRDM1/BLIMP-1 expression in multiple B and T-cell lymphoma. Haematologica. 2006. Apr; 91(4):467-74.

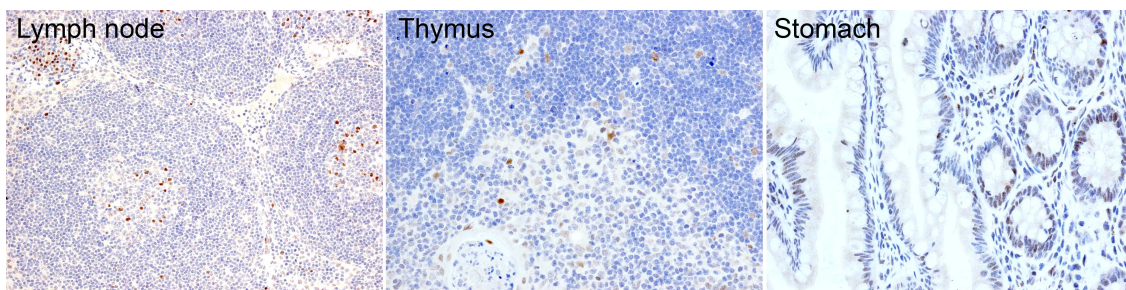
## APPLICATIONS

IHC Techniques	Clone	Dilution	Antibody concentration	Antigen retrieval method	Visualization kit	Positive control	Negative control	Protein localization	Positivity in other species
<b>Frozen tissue and cytopins</b>									
Recommended	ROS	1:2	supernatant	none	DAKO goat&mouse HRP	tonsil	muscle	nuclear	mouse
<b>Paraffin tissue</b>									
Recommended	ROS	1:10	supernatant	Tris-EDTA	Novolink Kit	tonsil	muscle	nuclear	mouse
<b>Immunofluorescence</b>									
Recommended	ROS	1:4	supernatant	tris-EDTA	Alexa goat&mouse 488	tonsil	muscle	nuclear	mouse



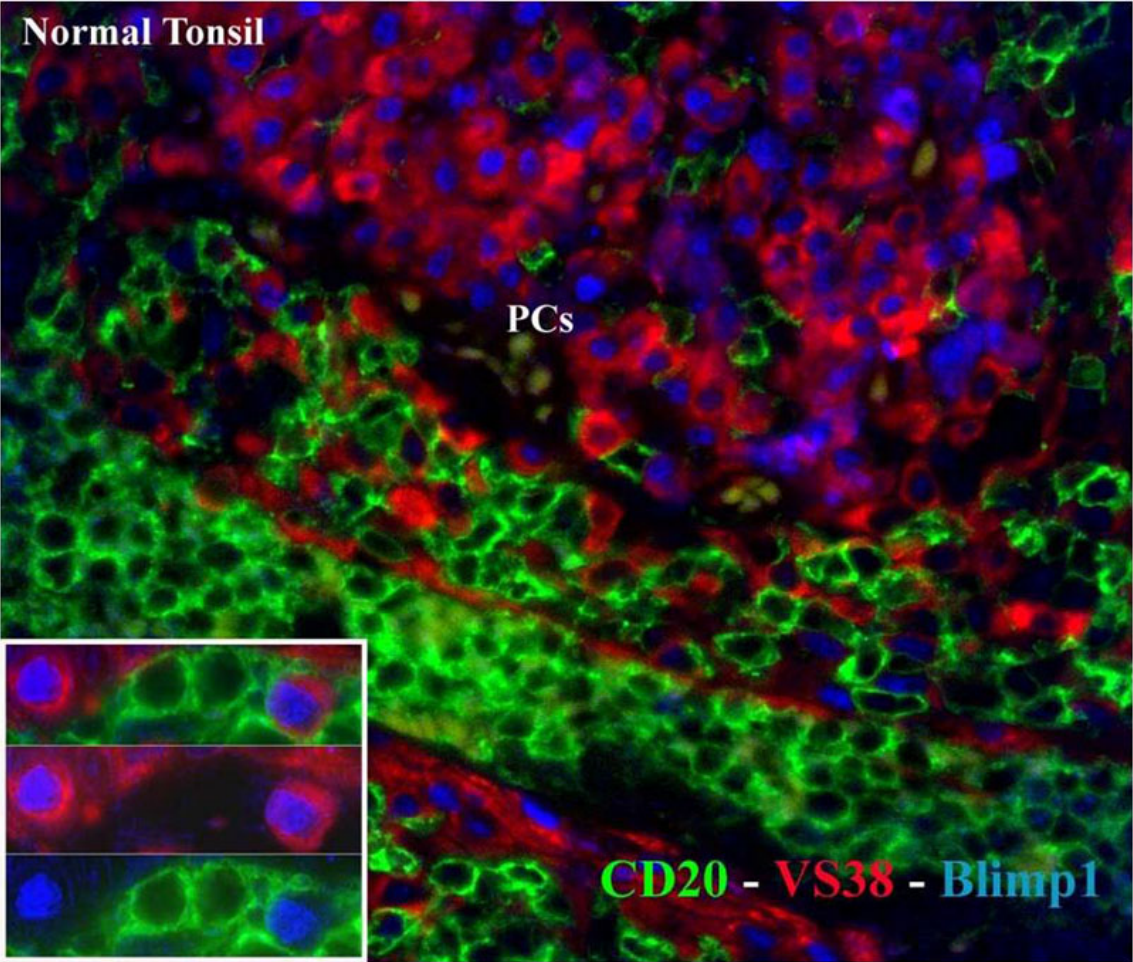
**ROS195 monoclonal antibody in transfected cells**

Nuclear staining on frozen cytospin preparations of transfected HEK293T/PRDM1 cells using antibody ROS195.



**PRDM-1/Blimp-1 (ROS195) IHC in human paraffin sections.**

In tonsil PRDM1 protein is strongly expressed by plasma cells present in germinal centres, and in subepithelial areas.



**Triple immunostaining of PRDM1/Blimp-1 (ROS195) in reactive tonsil.**

PRDM1/Blimp-1 nuclear protein (blue) is expressed in plasma cells (VS38c, plasma cell marker, red) but absent in CD20 positive (B lymphocytes, green).

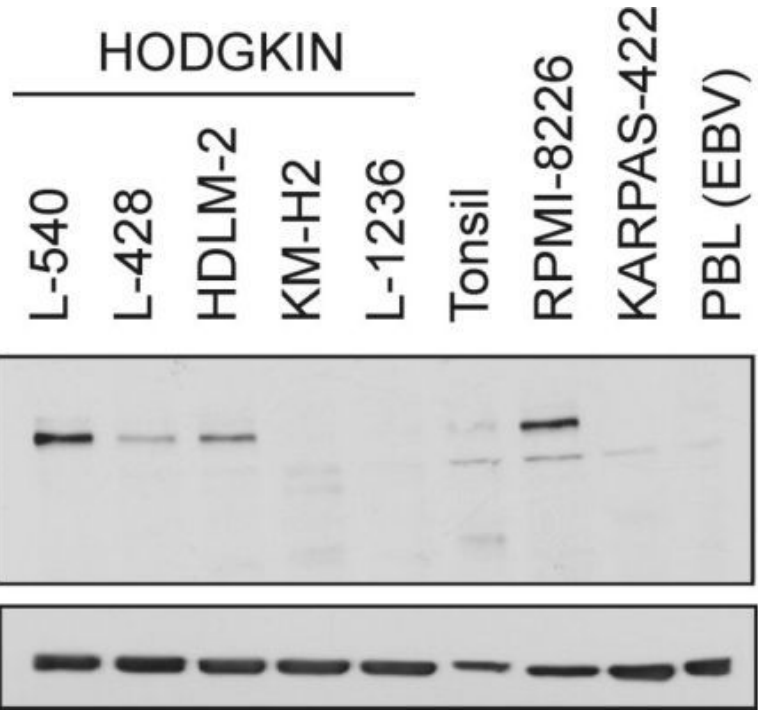
WB Techniques	Clone	Dilution	Antibody concentration	Positive control	Negative control	Expected MW	Observed Mw	Positivity in other species
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Western Blotting

Recommended	ROS	neat	supernatant	tonsil	muscle	98kDa	97kDa	not done
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Immunoprecipitation

Recommended	ROS	neat	supernatant	tonsil	muscle	98kDa	97kDa	not done
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Biochemical characterization of ROS195 monoclonal antibody.

Western blot analysis of PRDM1 expression in total protein extracts from lymphoma cell lines and human tonsil. Tubulin was used as loading control.

Lane 1 L-540 cell line (100ug) (+)

Lane 2 L-428 cell line (100ug) (+)

Lane 3 HDLM-2 cell line (100ug) (+)

Lane 4 KM-H2 cell line (100ug) (-)

Lane 5 L-1236 cell line (100ug) (-)

Lane 6 Human tonsil (100ug) (+)

Lane 7 RPMI-8226 cell line (100ug) (+)

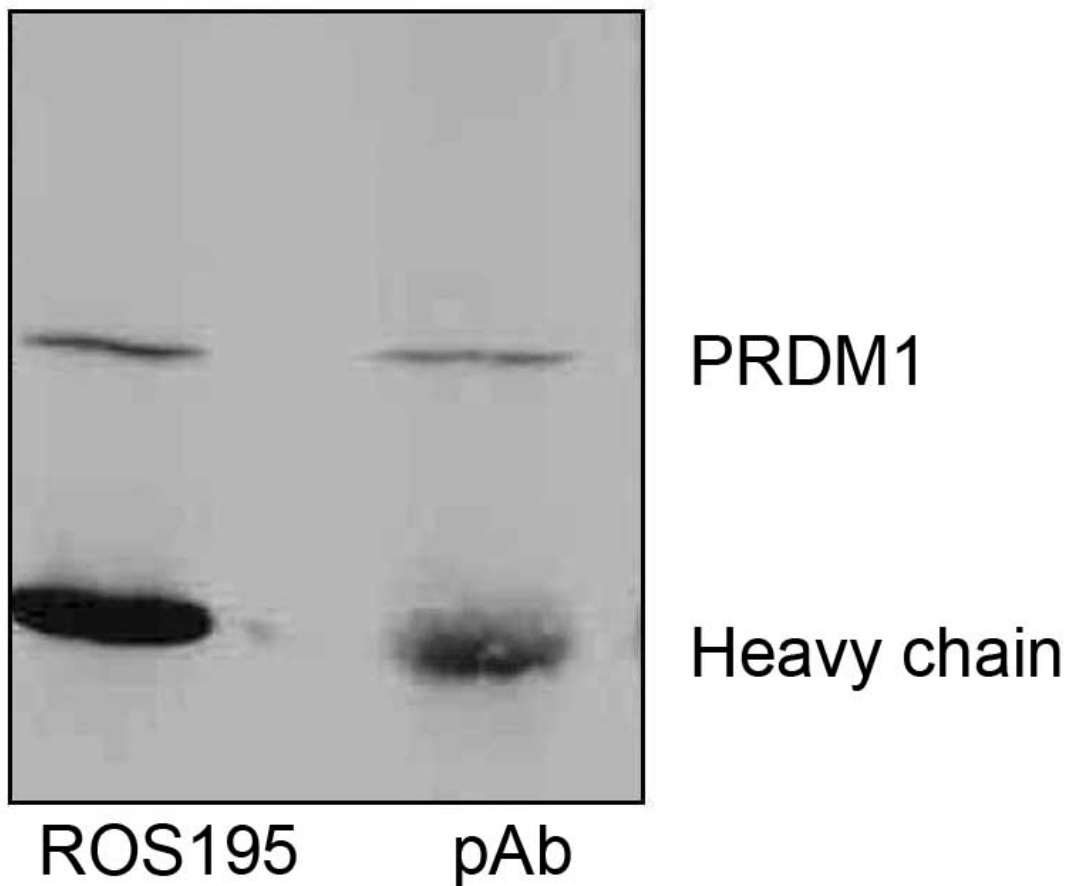
Lane 8 Karpas-422 cell line (100ug) (+)

Lane 9 PBL (EBV) cell line (100ug) (-)

Lane 10 LP-1 cell line (100ug) (+)

Tubulin was used as loading control.

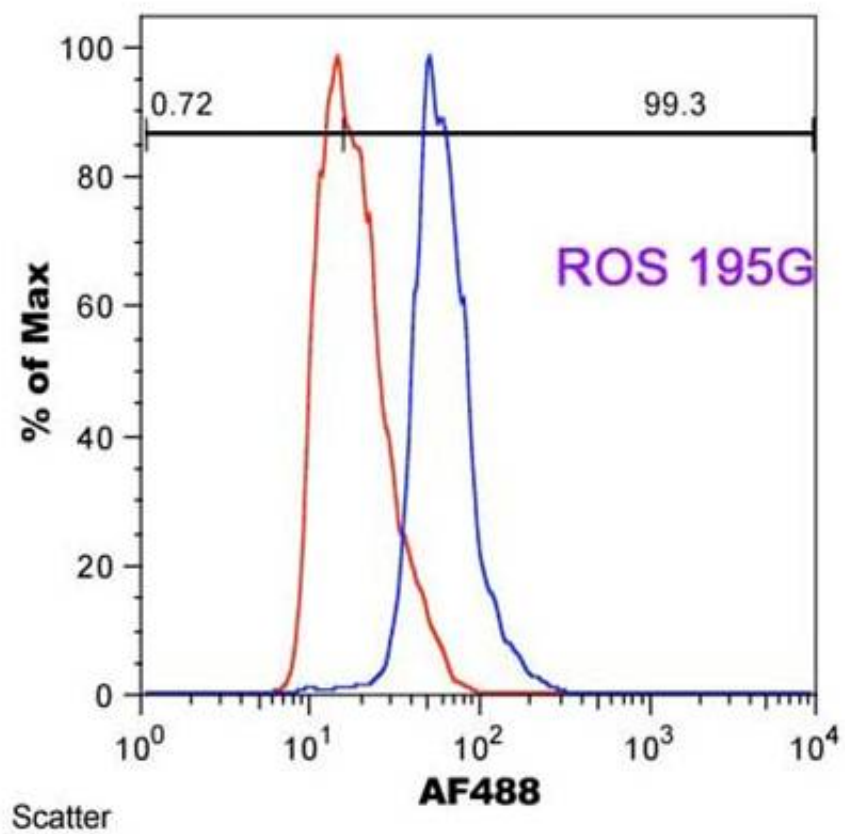


**PRDM1/Blimp-1 (ROS195) immunoprecipitation.**

Immunoprecipitation of protein extracts from normal tonsil with the anti-PRDM1 monoclonal antibody (ROS195 lane) and the specific polyclonal antibody (pAb lane) followed by a Western blotting with ROS195 antibody, shows a ~97 kDa band.

FLOW CYTOMETRY	Clone	Dilution	Positive control	Negative control	Type of fluorocrom
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Recommended	ROS	50ul supernatant/one million cells/tube	OPM2 cell line	MOLT4 cell line	
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ROS mAb by Flow cytometry

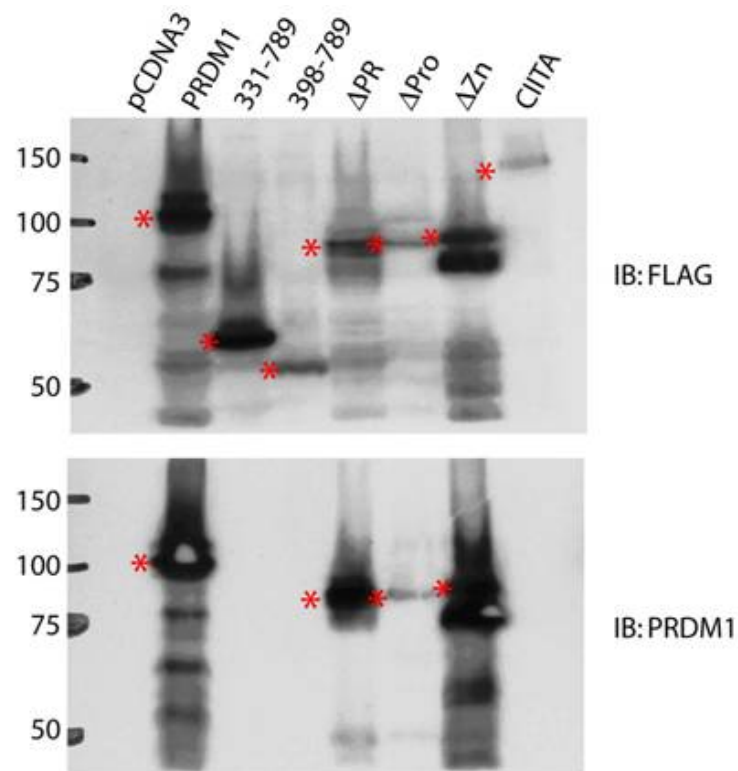


RED: Negative cell line MOLT4 (T cell leukemia)

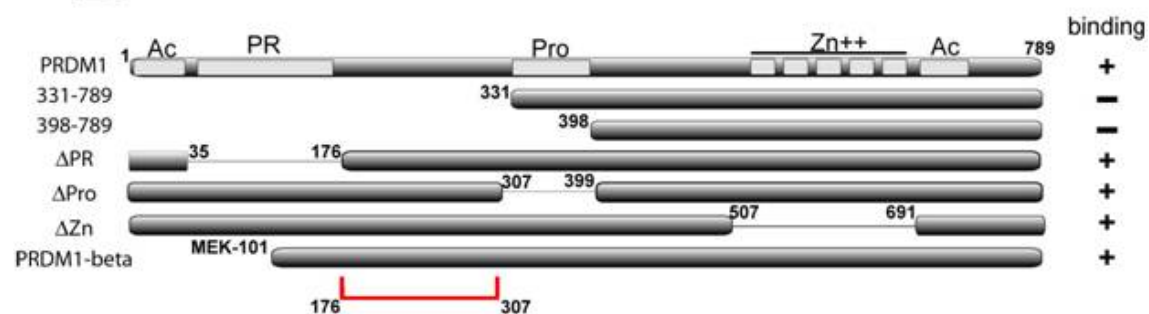
BLUE: Positive cell line OPM2 (multiple myeloma)

OTHERS	Title	Description
Recommended	Epitope recognise by ROS mAb	ROS) monoclonal antibody recognize site between amino acids 176 and 307 and is able to recognise the two blimp-1 isoforms.

**A**



**B**



### Epitope recognised by ROS mAb

ROS monoclonal antibody recognize site between amino acids 176 and 307 and is recognising the two blimp-1 isoforms.

Work done by Dr K. Wright (Lee Moffitt Cancer Center)