

## Brief report

## PRDM1 is involved in chemoresistance of T-cell lymphoma and down-regulated by the proteasome inhibitor

Wei-Li Zhao,<sup>1,2</sup> Yan-Yan Liu,<sup>1,3</sup> Qun-Ling Zhang,<sup>1,2</sup> Li Wang,<sup>1,2</sup> Christophe Leboeuf,<sup>2,4,5</sup> Yi-Wen Zhang,<sup>1,2</sup> Jie Ma,<sup>3</sup> José-Francisco García,<sup>6</sup> Yong-Ping Song,<sup>3</sup> Jun-Min Li,<sup>1</sup> Zhi-Xiang Shen,<sup>1</sup> Zhu Chen,<sup>1,2</sup> Anne Janin,<sup>2,4,5</sup> and Sai-Juan Chen<sup>1,2</sup>

<sup>1</sup>State Key Laboratory of Medical Genomics, Shanghai Institute of Hematology, Shanghai Rui Jin Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai, China; <sup>2</sup>Pôle de Recherches Franco-Chinois en Science du Vivant et Génomique, Laboratory of Molecular Pathology, Shanghai, China; <sup>3</sup>Henan Institute of Hematology, Henan Tumor Hospital, Zhengzhou, China; <sup>4</sup>Inserm, U728, Institut d'Hématologie, Hôpital Saint Louis, Paris, France; <sup>5</sup>Université Paris 7-Denis Diderot, Paris, France; and <sup>6</sup>The Monoclonal Antibodies Unit, Biotechnology Program, Spanish National Cancer Center (CNIO), Madrid, Spain

**The positive regulatory domain I (PRDM1) is a master regulator of terminal B-cell differentiation. However, PRDM1 is not B-cell specific. To determine its role in T-cell lymphoma, PRDM1 expression was investigated in 60 patients. *PRDM1α* and *PRDM1β* transcripts were detected in laser-microdissected T-lymphoma cells in 27 and 14 patients, respectively, mostly in cases with IRF4 expression. *PRDM1β* was associated with increased c-MYC ex-**

**pression. *PRDM1β*-positive patients displayed advanced Ann Arbor stage and high-risk International Prognostic Index and were linked to short survival times. In vitro, *PRDM1β* was related to resistance to chemotherapeutic agents and could be down-regulated by the proteasome inhibitor bortezomib. Kinetic studies showed that bortezomib down-regulation of *PRDM1β* preceded decreased IRF4 and c-MYC expression. An earlier retaining of**

**cytoplasmic IκBα in bortezomib-treated cells was revealed, concomitant with blockade of NF-κB nuclear translocation. These results demonstrate the involvement of *PRDM1β* in T-cell lymphoma, with possible therapeutic interference by the proteasome inhibitor. (Blood. 2008;111:3867-3871)**

© 2008 by The American Society of Hematology

## Introduction

The positive regulatory domain I (PRDM1) belongs to the *PRDM* gene family of transcriptional repressors and is required for terminal B-cell differentiation into plasma cells.<sup>1</sup> However, the involvement of PRDM1 in hematopoietic development is not restricted to the B-cell lineage. Recent studies have implicated PRDM1 in the regulation of T-cell activation and homeostasis.<sup>2,3</sup> PRDM1 was also detected in a subset of T-cell lymphomas,<sup>4</sup> while the significance of PRDM1 expression in this disease remains to be investigated.

PRDM1 has 2 isoforms: PRDM1α and PRDM1β. Generated from the same gene by alternative transcription initiation, PRDM1β differs from PRDM1α by the lack of 101 NH2-terminal amino acids, resulting in a disrupted PR domain and loss of repressive function on multiple target genes.<sup>5</sup> Recently, we have shown that PRDM1β is associated with chemoresistance in B-cell lymphoma, which could be overcome by nonchemotherapeutic agent rituximab through NF-κB inactivation.<sup>6</sup> Here we identify the role of PRDM1β in T-cell lymphoma and its modulation in vitro by the proteasome inhibitor bortezomib.

summarized in Table 1. This study was approved by the Shanghai Institute of Hematology-Rui Jin Hospital Institutional Review Board. Informed consent was obtained from all patients in accordance with the Declaration of Helsinki.

## Tissue samples

Lymphoma tissue samples were immediately cut into 2 parts: 1 part fixed in formaldehyde and processed for paraffin embedding, and the other snap frozen and stored at  $-80^{\circ}\text{C}$ . Systematic microscopic control of the lymphoma lesion was performed for each block.

## Cell culture

T-lymphoma cell lines HUT78, HUT102, and MOLT4 (American Type Culture Collection, Manassas, VA) were treated with chemotherapeutic agents, bortezomib (25 nM; Millennium Pharmaceuticals, Cambridge, MA) or pyrrolidine dithiocarbamate (PDT, 20 μM; Sigma-Aldrich, St Louis, MO), which specifically inhibits NF-κB activation.<sup>7</sup> Methotrexate-resistant subclone was selected from the original MOLT4 by exposure to increasing concentrations of methotrexate, as described by Fotoohi et al.<sup>8</sup>

## Immunohistochemistry and immunofluorescence assay

Immunohistochemical analysis was performed on 5-μm paraffin sections with an indirect immunoperoxidase method, using antibodies against PRDM1,<sup>6</sup> IRF4 (1:40; Dako, Glostrup, Denmark), and c-MYC (1:100; Santa Cruz Biotechnology, Santa Cruz, CA). Immunofluorescence assay was performed as previously reported.<sup>9</sup>

## Methods

## Patients

Sixty de novo T-cell lymphoma patients (32 male and 28 female, ages 18 to 70 [median, 43] years) were included. Their main characteristics are

Submitted August 21, 2007; accepted January 19, 2008. Prepublished online as Blood First Edition paper, January 30, 2008; DOI 10.1182/blood-2007-08-108654.

W.-L.Z., Y.-Y.L., Q.-L.Z., and L.W. contributed equally to this work.

The publication costs of this article were defrayed in part by page charge payment. Therefore, and solely to indicate this fact, this article is hereby marked "advertisement" in accordance with 18 USC section 1734.

© 2008 by The American Society of Hematology

Table 1. PRDM1 expression in T-cell lymphoma

	No.	PRDM1 (%)	IRF4 (%)	Laser-microdissected cells		
				<i>PRDM1</i> α (%)	<i>PRDM1</i> β (%)	<i>IRF4</i> (%)
Histologic typing (World Health Organization classification)						
Peripheral T-cell lymphoma	23	10 (43.5)	15 (65.2)	14 (60.9)	7 (30.4)	15 (65.2)
Anaplastic large-cell lymphoma	9	5 (55.6)	8 (88.9)	7 (77.8)	4 (44.4)	8 (88.9)
T/NK nasal-type lymphoma	12	3 (25.0)	8 (66.7)	4 (33.3)	2 (16.7)	8 (66.7)
T-angioimmunoblastic lymphoma	5	1 (20.0)	2 (40.0)	2 (40.0)	1 (20.0)	2 (40.0)
T-lymphoblastic lymphoma	11	0	0	0	0	0
Ann Arbor stage						
I to II	12	2 (16.7)	6 (50.0)	4 (33.3)	0	6 (50.0)
III to IV	48	17 (35.4)	27 (56.3)	23 (47.9)	14 (29.2)	27 (56.3)
No. of sites of extranodal involvement						
0 to 1	29	10 (34.5)	18 (62.2)	13 (44.8)	6 (20.7)	18 (62.2)
2 to 3	31	9 (29.0)	15 (48.4)	14 (45.2)	8 (25.8)	15 (48.4)
Serum LDH level						
Normal or lower	23	9 (39.1)	14 (60.9)	10 (43.5)	6 (26.1)	14 (60.9)
Higher than normal	37	10 (27.0)	19 (51.4)	17 (45.9)	8 (21.6)	19 (51.4)
IPI score						
Low risk to intermediate low risk	23	5 (21.7)	13 (56.5)	9 (39.1)	2 (8.7)	13 (56.5)
Intermediate high risk to high risk	37	14 (37.8)	20 (54.1)	18 (48.6)	12 (32.4)	20 (54.1)

### Laser microdissection

Frozen sections (7  $\mu$ m each) were incubated in RNase-free conditions with anti-CD3 antibody (1:50; Dako) and fluorescein-conjugated goat anti-mouse IgG antibody (1:50; Jackson ImmunoResearch Laboratories, West Grove, PA) each for 5 minutes. Laser microdissection of fluorescent cells was immediately performed (Leica Microsystems, Wetzlar, Germany) for RNA extraction.

### Semiquantitative reverse-transcription polymerase chain reaction (PCR), sequence analysis, and Western blot

As previously reported,<sup>6</sup> the NF- $\kappa$ B (P65) and I $\kappa$ B $\alpha$  antibodies were obtained from Santa Cruz Biotechnology. The phosphorylated form of I $\kappa$ B $\alpha$  (p-I $\kappa$ B $\alpha$ ) antibody was purchased from Cell Signaling (Beverly, MA).

### Statistical analysis

Patient characteristics were compared using  $\chi^2$  and Fisher exact test. Kaplan-Meier and log-rank test were performed to estimate the survival functions. Overall survival was measured from the time of diagnosis to the date of death or last contact. Event-free survival was calculated from the time of diagnosis to the date of progression, death, or last contact. *P* values less than .05 were considered significant. All statistical analyses were performed on SAS 8.2 software (SAS Institute, Cary, NC).

## Results and discussion

### T-lymphoma cells expressed PRDM1

By immunohistochemistry, nuclear PRDM1 expression (Figure 1A) was observed in 19 (31.7%) of 60 patients. According to histologic type, PRDM1 was found in peripheral T-cell lymphoma (10/23, 43.5%), anaplastic large-cell lymphoma (5/9, 55.6%), T/natural killer (NK) nasal-type lymphoma (3/12, 25.0%), and T-angioimmunoblastic lymphoma (1/5, 20.0%) (Table 1). T-lymphoblastic lymphoma did not express PRDM1, as shown by Garcia et al.<sup>4</sup>

Using semiquantitative PCR, PRDM1 $\alpha$  and PRDM1 $\beta$  transcripts were identified in laser-microdissected lymphoma cells

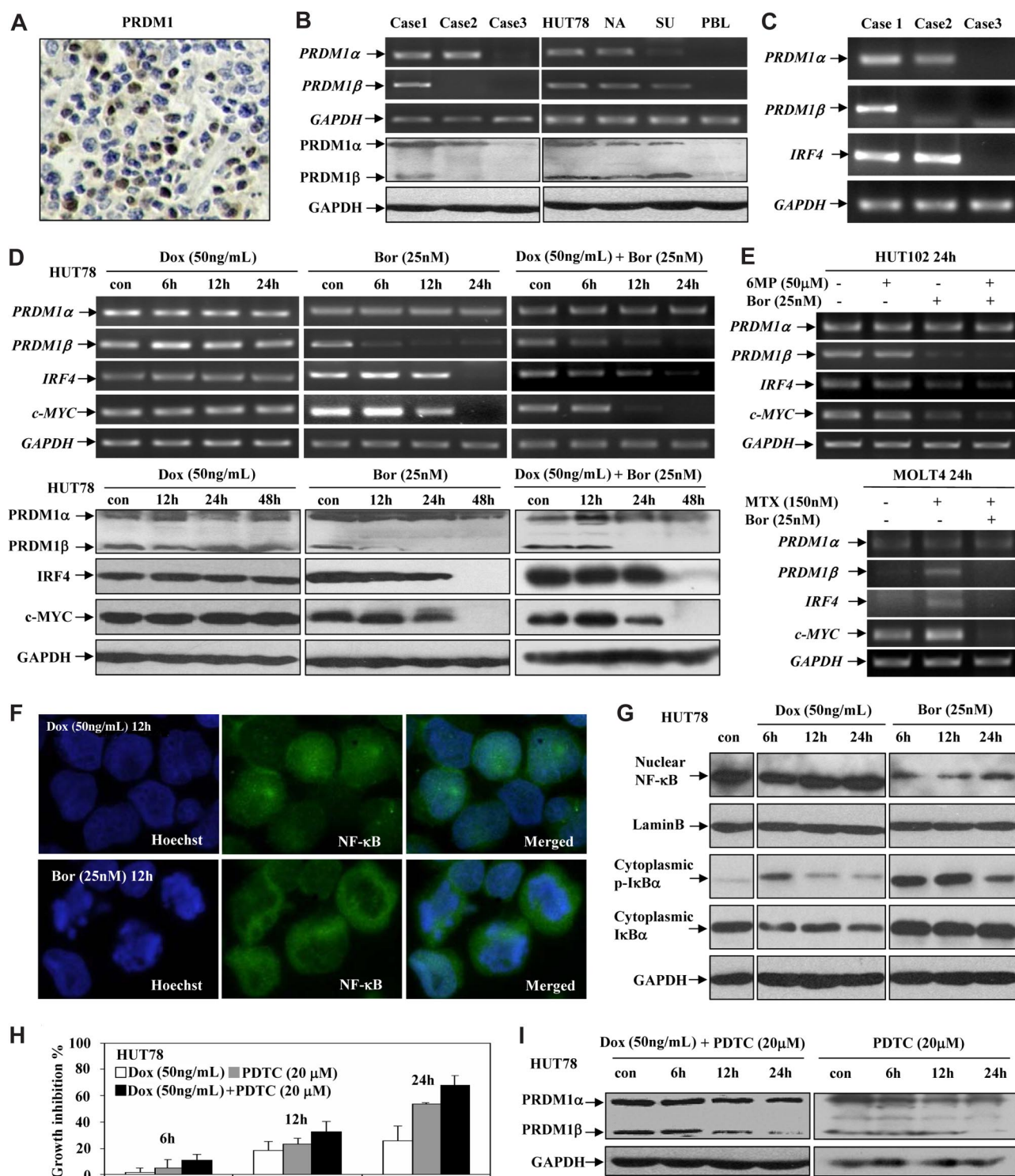
(Figure 1B) of 27 and 14 patients, respectively. PRDM1 $\alpha$  mRNA was detected in a higher number of patients (27/60) than PRDM1 protein (19/60). A similar discrepancy was also reported in B-cell lymphoma.<sup>10</sup> Although systematic sequencing of PRDM1 in 30 patients with adequate frozen tissue samples revealed no mutation, other genetic/epigenetic inactivation as well as defects in protein translation or stability might contribute to the lack of PRDM1 protein expression. Instead, PRDM1 $\beta$  mRNA was detected in 14 of 19 cases positive for PRDM1 protein. The constant association between PRDM1 $\beta$  mRNA and PRDM1 protein led us to further consider the role of this altered isoform in T-cell lymphoma.

IRF4 was significantly related to PRDM1 transcripts: it was higher in PRDM1 $\alpha$ <sup>+</sup>PRDM1 $\beta$ <sup>+</sup> (14/14, 100%) and PRDM1 $\alpha$ <sup>+</sup>PRDM1 $\beta$ <sup>-</sup> cases (11/13, 84.6%), than in PRDM1 $\alpha$ <sup>-</sup>PRDM1 $\beta$ <sup>-</sup> cases (8/33, 24.2%; *P* = .008 and *P* = .024, respectively). Laser-microdissected lymphoma cells revealed IRF4 coexpressing with PRDM1 $\alpha$  and PRDM1 $\beta$  (Figure 1C). Responsible for B-cell differentiation, IRF4 is expressed in neoplastic activated T cells and regulates T-cell function and transformation.<sup>11-14</sup> This IRF4-PRDM1 coexpression confirmed that PRDM1 happens with IRF4 in T-cell lymphoma.

c-MYC is a PRDM1-targeted gene and critical for cell proliferation.<sup>15,16</sup> c-MYC was documented more frequently in PRDM1 $\alpha$ <sup>+</sup>PRDM1 $\beta$ <sup>+</sup> cases (13/14, 92.9%), compared with PRDM1 $\alpha$ <sup>+</sup>PRDM1 $\beta$ <sup>-</sup> cases (2/13, 15.4%; *P* = .024). Therefore, PRDM1 $\beta$  was unable to repress c-MYC, which may cause T-cell proliferation.

### PRDM1 $\beta$ reflected poor disease outcome

PRDM1 $\beta$ , but not PRDM1 $\alpha$ , was significantly related to advanced Ann Arbor stage (*P* = .033), high-risk International Prognostic Index (*P* = .035, Table 1), and shorter survival times. The 3-year event-free survival and overall survival rates ( $\pm$  SE percentage) were significantly shorter for PRDM1 $\alpha$ <sup>+</sup>PRDM1 $\beta$ <sup>+</sup> patients (23.1% [ $\pm$  11.7%] and 55.6%



**Figure 1. PRDM1 was expressed in T-cell lymphoma and could be down-regulated by bortezomib through NF-κB inactivation.** (A) Nuclear PRDM1 protein expression was revealed by immunohistochemistry on lymphoma tissue. Leica AS LMD (Leica Microsystems), 40×/0.6 CORR XT, air/0.6, hematoxylin-eosin stain, Video camera 3CCD Hitachi HV-Dz0P, Leica AS LMD software, version 4.4. (B) PRDM1 mRNA and protein expression were detected by semiquantitative PCR on lymphoma cells and Western blot on lymphoma tissue, respectively. PRDM1 isoforms were expressed in laser-microdissected lymphoma cells and T- and B-lymphoma cell lines (NA indicates namalwa; SU, SU-DHL-4), but not in peripheral blood lymphocytes (PBLs). (C) Laser-microdissected lymphoma cells showed coexpression of *PRDM1* and *IRF4*. (D) In HUT78 cells resistant to doxorubicin (Dox), *PRDM1β*, *IRF4*, and *c-MYC* expression was reduced by bortezomib (Bor) both at transcriptional (top panel) and protein (bottom panel) level. (E) In HUT102 cells resistant to 6-mercaptopurine (6MP, 50 μM), *PRDM1β*, *IRF4*, and *c-MYC* expression was also reduced by bortezomib (Bor, 25 nM) (top panel). In MOLT4 cells, when methotrexate resistance was developed (MTX, 150 nM), *PRDM1β* was expressed, and subsequently reduced by bortezomib (Bor, 25 nM) (bottom panel). (F) Immunofluorescence study revealed that NF-κB (P65) was located mainly in nucleus in doxorubicin (Dox)-treated HUT78 cells, while in cytoplasm in those treated with bortezomib (Bor). Leica AS LMD, 100×/1.3 oil, oil/1.3, Immunofluorescent stain, Video camera 3CCD Hitachi HV-Dz0P (Hitachi, Tokyo, Japan), Leica AS LMD software, version 4.4. (G) Bortezomib (Bor) blocked NF-κB nuclear translocation and retained the phosphorylated form of IκBα (p-IκBα) and IκBα in the cytoplasm of HUT78 cells. Lamin B was used as a nuclear protein control. (H and I) Addition of NF-κB inhibitor PDTC increased the antiproliferative effect of doxorubicin (Dox) on HUT78 cells (H) and induced *PRDM1β* down-regulation (I).



[ $\pm 14.9\%$ ]) than for  $PRDM1\alpha^+PRDM1\beta^-$  cases (48.5% [ $\pm 16.6\%$ ] and 87.5% [ $\pm 11.7\%$ ] ( $P = .040$  and  $P = 0.018$ , respectively). Therefore, like its analog  $PRDM2$  (RIZ) and  $PRDM3$  (MDS1-EV11) genes, each expressing a truncated protein missing the PR domain and critical for oncogenesis,<sup>17,18</sup>  $PRDM1\beta$  indicated poor disease outcome in T-cell lymphoma.

### Proteasome inhibitor bortezomib down-regulated $PRDM1\beta$ through NF- $\kappa$ B inactivation

HUT78 cells were resistant to doxorubicin, but sensitive to bortezomib. In a 6-hour incubation with bortezomib, either alone or with doxorubicin,  $PRDM1\beta$ , but not  $PRDM1\alpha$ , was down-regulated.  $IRF4$  and  $c-MYC$  were also decreased after 12-hour treatment. Their protein levels followed a similar reduction, observed from 12 to 24 hours after treatment (Figure 1D). In addition, such alternation was also revealed in 6-mercaptopurine-resistant HUT102 cells and methotrexate-resistant MOLT4 subclone (Figure 1E). Bortezomib, the first proteasome inhibitor used in the clinic, has recently been reported as effective in T-cell lymphoma.<sup>19-21</sup>  $PRDM1\beta$  down-regulation, with the subsequent inhibition of  $c-MYC$  and  $IRF4$ , could be one of the possible molecular mechanisms of its action.

$PRDM1$  is a downstream target of NF- $\kappa$ B,<sup>22</sup> whose activation induces T-lymphoma cell resistance to chemotherapy.<sup>23,24</sup> Nuclear localization of NF- $\kappa$ B, required for target gene transactivation, was studied by immunofluorescence assay and showed an inhibition after 12-hour treatment of bortezomib (Figure 1F). This was confirmed on Western blot, with decrease of nuclear NF- $\kappa$ B protein (Figure 1G). Further analysis of cytoplasmic I $\kappa$ B $\alpha$  revealed striking differences: it decreased under doxorubicin, while increasing under bortezomib treatment. Together, increasing cytoplasmic I $\kappa$ B $\alpha$ , concomitant with blockade of NF- $\kappa$ B nuclear translocation, suggested NF- $\kappa$ B inactivation in T-lymphoma cells treated with bortezomib.<sup>25</sup>

To further test this hypothesis, we associated PDTC, a NF- $\kappa$ B inhibitor, with doxorubicin in T-lymphoma cell cultures. Growth inhibition (Figure 1H) and the parallel decrease of  $PRDM1\beta$  (Figure 1I) could be reproduced when treated with bortezomib. We thus confirmed that NF- $\kappa$ B activity was responsible for  $PRDM1\beta$  down-regulation by bortezomib.

In conclusion, abnormal  $PRDM1\beta$  expression was correlated with poor disease outcome in T-cell lymphoma patients. Bortezomib could down-regulate  $PRDM1\beta$  through NF- $\kappa$ B inactivation.

The value of proteasome inhibitor as a therapeutic strategy should be further assessed in T-cell lymphoma, especially those cases refractory to conventional chemotherapy.

### Acknowledgments

This work was supported in part by the Chinese National Key Program for Basic Research (973:2004CB518600), the Chinese National High Tech Program (863:2006AA02A301 and 863:2006AA02A405), the National Natural Science Foundation of China (30750335), the Key Discipline Program of Shanghai Municipal Education Commission (Y0201), the Shanghai Commission of Science and Technology (44107025), the Shanghai Rising Star Program (05QMX1429), the Program for New Century Excellent Talents in University, the Scientific Research Foundation for the Returned Overseas Chinese Scholars, the Program de Recherches Avancées (PRA B 06-01), and the Samuel Waxman Cancer Research Foundation Laboratory, the Co-PI Program of Shanghai Rui Jin Hospital/Medical School of Shanghai Jiao Tong University.

### Authorship

Contribution: W.-L.Z., Z.C., A.J., and S.-J.C. designed the research and wrote the paper; Y.-Y.L., Q.-L.Z., L.W., C.L., and Y.-W.Z. performed research and statistical analysis; J.M. and J.-F.G. contributed vital new reagents; J.-M.L., Y.-P.S., and Z.-X.S. collected, analyzed, and interpreted data.

Conflict-of-interest disclosure: The authors declare no competing financial interests.

Correspondence: Sai-Juan Chen, State Key Laboratory of Medical Genomics, Shanghai Institute of Hematology, Rui Jin Hospital, Shanghai Jiao Tong University School of Medicine, 197 Rui Jin Er Road, Shanghai 200025, China; e-mail: sjchen@stn.sh.cn; or Anne Janin, Inserm U728, Laboratoire de Pathologie, Hôpital Saint Louis, 1 Avenue Claude Vellefaux, Paris 75010, France; e-mail: anne\_janin@yahoo.com.

### References

- Shapiro-Shelef M, Calame K. Regulation of plasma-cell development. *Nat Rev Immunol*. 2005;5:230-242.
- Martins GA, Cimmino L, Shapiro-Shelef M, et al. Transcriptional repressor Blimp-1 regulates T cell homeostasis and function. *Nat Immunol*. 2006;7:457-465.
- Kallies A, Hawkins ED, Belz GT, et al. Transcriptional repressor Blimp-1 is essential for T cell homeostasis and self-tolerance. *Nat Immunol*. 2006;7:466-474.
- Garcia JF, Roncador G, Sanz AI, et al.  $PRDM1/BLIMP-1$  expression in multiple B and T-cell lymphoma. *Haematologica*. 2006;91:467-474.
- Gyory I, Fejer G, Ghosh N, Seto E, Wright KL. Identification of a functionally impaired positive regulatory domain 1 binding factor 1 transcription repressor in myeloma cell lines. *J Immunol*. 2003;170:3125-3133.
- Liu YY, Leboeuf C, Shi JY, et al. Rituximab plus CHOP (R-CHOP) overcomes  $PRDM1$ -associated resistance to chemotherapy in patients with diffuse large B-cell lymphoma. *Blood*. 2007;110:339-344.
- Schreck R, Meier B, Mannel DN, Droge W, Baeuerle PA. Dithiocarbamates as potent inhibitors of nuclear factor kappa B activation in intact cells. *J Exp Med*. 1992;175:1181-1194.
- Fotoohi K, Jansen G, Assaraf YG, et al. Disparate mechanisms of antifolate resistance provoked by methotrexate and its metabolite 7-hydroxymethotrexate in leukemia cells: implications for efficacy of methotrexate therapy. *Blood*. 2004;104:4194-4201.
- Wang L, Zhao WL, Yan JS, et al. Eriocalyxin B induces apoptosis of t(8;21) leukemia cells through NF-kappaB and MAPK signaling pathways and triggers degradation of AML1-ETO oncoprotein in a caspase-3-dependent manner. *Cell Death Differ*. 2007;14:306-317.
- Pasqualucci L, Compagno M, Houldsworth J, et al. Inactivation of the  $PRDM1/BLIMP1$  gene in diffuse large B cell lymphoma. *J Exp Med*. 2006;203:311-317.
- Sciammas R, Shaffer AL, Schatz JH, Zhao H, Staudt LM, Singh H. Graded expression of interferon regulatory factor-4 coordinates isotype switching with plasma cell differentiation. *Immunity*. 2006;25:225-236.
- Falini B, Fizzotti M, Pucciarini A, et al. A monoclonal antibody (MUM1p) detects expression of the MUM1/IRF4 protein in a subset of germinal center B cells, plasma cells, and activated T cells. *Blood*. 2000;95:2084-2092.
- Mittrucker HW, Matsuyama T, Grossman A, et al. Requirement for the transcription factor LSIRF/IRF4 for mature B and T lymphocyte function. *Science*. 1997;275:540-543.
- Ramos JC, Ruiz P Jr, Ratner L, et al. IRF-4 and c-Rel expression in antiviral-resistant adult T-cell leukemia/lymphoma. *Blood*. 2007;109:3060-3068.
- Lin Y, Wong K, Calame K. Repression of c-myc transcription by Blimp-1, an inducer of terminal B cell differentiation. *Science*. 1997;276:596-599.

16. Hoffman B, Amanullah A, Shafarenko M, Liebermann DA. The proto-oncogene c-myc in hematopoietic development and leukemogenesis. *Oncogene*. 2002;21:3414-3421.
17. Sasaki O, Meguro K, Tohmiya Y, Funato T, Shibahara S, Sasaki T. Altered expression of retinoblastoma protein-interacting zinc finger gene, RIZ, in human leukaemia. *Br J Haematol*. 2002;119:940-948.
18. Cuenco GM, Nucifora G, Ren R. Human AML1/MDS1/EVI1 fusion protein induces an acute myelogenous leukemia (AML) in mice: a model for human AML. *Proc Natl Acad Sci U S A*. 2000;97:1760-1765.
19. Orlowski RZ, Voorhees PM, Garcia RA, et al. Phase 1 trial of the proteasome inhibitor bortezomib and pegylated liposomal doxorubicin in patients with advanced hematologic malignancies. *Blood*. 2005;105:3058-3065.
20. Mai W, Meng H, Jin J, Wang L. Treatment with bortezomib in a patient with heavily pretreated refractory T-cell lymphoblastic lymphoma. *Eur J Haematol*. 2006;77:445-447.
21. Zinzani PL, Musuraca G, Tani M, et al. Phase II trial of proteasome inhibitor bortezomib in patients with relapsed or refractory cutaneous T-cell lymphoma. *J Clin Oncol*. 2007;25:4293-4297.
22. Johnson K, Shapiro-Shelef M, Tunyaplin C, Calame K. Regulatory events in early and late B-cell differentiation. *Mol Immunol*. 2005;42:749-761.
23. Karin M, Cao Y, Greten FR, Li ZW. NF-kappaB in cancer: from innocent bystander to major culprit. *Nat Rev Cancer*. 2002;2:301-310.
24. Sors A, Jean-Louis F, Pellet C, et al. Down-regulating constitutive activation of the NF-kappaB canonical pathway overcomes the resistance of cutaneous T-cell lymphoma to apoptosis. *Blood*. 2006;107:2354-2363.
25. Sun SC, Ganchi PA, Ballard DW, Greene WC. NF-kappa B controls expression of inhibitor I kappa B alpha: evidence for an inducible autoregulatory pathway. *Science*. 1993;259:1912-1915.