SUPPLEMENTAL INFORMATION

SUPPLEMENTAL METHODS

TCL1-tg mice and tumor analysis.

Transgenic and wild type mice (C57BL/6 x C3H background) were housed and mated in a pathogen-free facility. Normal spleens were harvested from wild type or *Tcl1*-tg mice at six weeks of age, prior to the development of lymphoma or pre-malignant lymphocyte expansions. B cell tumors in this study occurred in *Tcl1*-tg mice between 8 and 16 months of age with a median age of 11 months (Hoyer et al., 2002). Lymphomas were diagnosed and classified by histology, flow cytometry and immunohistochemistry using criteria outlined by the Hematopathology Subcomittee of the Mouse Models of Human Cancer Consortium (MMHCC, Morse et al., 2002). Flow analysis of dispersed splenocytes from the majority of malignant spleens demonstrated at least 74% IgM^{+}, $B220^{lo}$,$CD5^{lo}$ B cells. See supplemental Table 1 for details regarding individual mice. All animal work was performed in compliance with institutional guidelines for animal use.

Drug treatment

For each treatment condition, a total of 1 x 10^6 cells were plated at equal density in RPMI media supplemented with 10% FBS. Freshly prepared 5-aza-2'-deoxycytidine (Aza, Sigma) in DMSO was added to the media every 24 hours for a total of 72 hours. For mock treatment, cells were treated with an equal volume of media + DMSO alone. For trichostatin A (TSA, Sigma) treatment, cells were first treated for 72 hours with Aza or DMSO (mock) and then treated for 24 hours with 100nM TSA.

Lymphocyte Isolations

Mouse B and T cells were isolated by negative selection (anti-CD4-PE/anti-CD8-PE/anti-Gr1-PE or anti-B220-PE/anti-Gr1-PE , respectively) of non-malignant splenocytes from eight 8-week old TCL1-tg mice as previously described (Shen et al., 2006). Subsequent FACS analysis with anti-B220 or anti-CD3 confirmed greater than 97% purity of isolated murine B or T cell populations. Human B and T cell were isolated to greater than 95% purity by separating lymphocytes from hyperplastic tonsils through a Ficoll-Paque Plus cushion and subjecting them to negative selection (anti-CD3 or anti-CD19, respectively) as previously described (Said et al., 2001).

FL-EphA7 expression in Nalm-6 cells

The full-length coding sequence for human *EPHA7* (FL-EphA7) was PCR-cloned using 293T cDNA and primers introducing *Age*I and *Not*I restriction sites at the 5' and 3' ends, respectively. The PCR product was directly cloned into the MSCV-GFP-IRES-PURO retroviral vector using above restriction sites, replacing the GFP insert. Retroviral supernatants were produced by transient transfection of the 293T Phoenix packaging cell line, after which Nalm-6 cells were bulk infected and selected in puromycin.

Chemotaxis assay

Chemotaxis assays were performed in Transwell plates (5μM diameter membrane pores, Corning) in serum free RPMI medium + 0.5% BSA. Nalm-6 cells (2x105/well) were pre-incubated for 20 minutes in the presence or absence of soluble EphA7-Fc recombinant protein and then migrated for 2 hours at 37°C to 100ng/ml SDF1α (R&D Systems) placed in the bottom well. The numbers of cells passing into the lower chamber were measured by MTT assay (ATCC). For each experiment, between 15-20% of total cells in top chamber migrated through the membrane to SDF1α, while less than 1% migration was seen in the absence of SDF1α.

Bioinformatics and statistical analysis

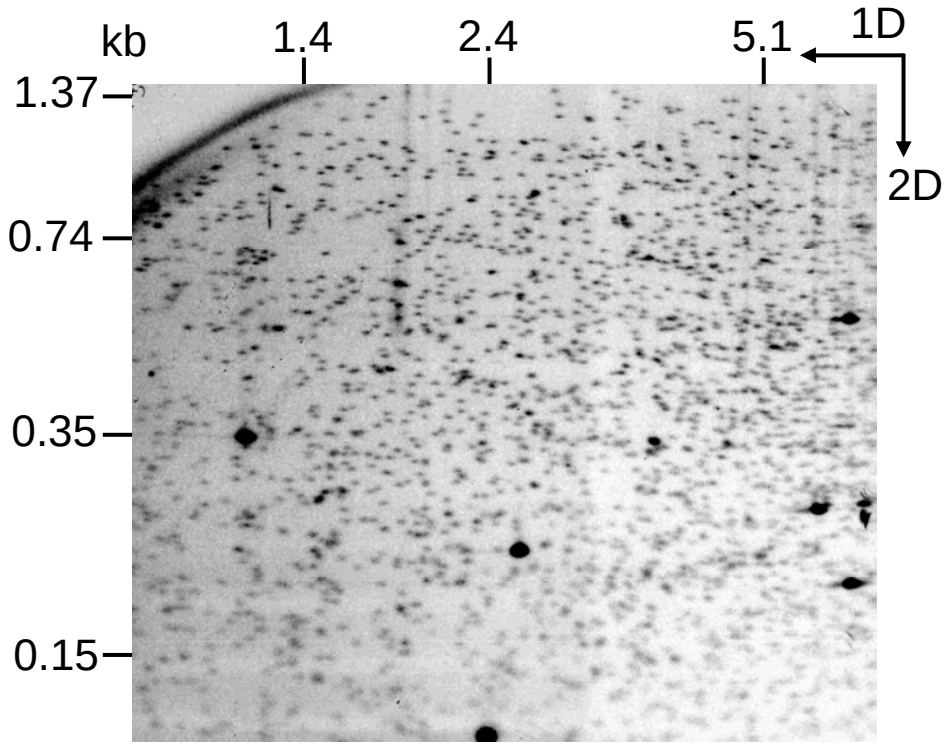
A CpG island was defined as a contiguous region of greater than 200 bp with >50% G+C and CpG content greater than 60% of that expected based on the G+C for that region. Preferential methylation was assessed as previously described using a standard goodness-of-fit test that assumes all spots are lost at equal true frequency (Yu et al., 2005).

SUPPLEMENTAL TABLE 1. Necropsy and tumor diagnoses for *TCL1-tg* mice.

TUMOR[1]	AGE (Months)	SPLEEN (Grams)	IgM^+, $B220^{lo}$,$CD5^{lo}$ (% of cells)	PAX5 Status (by IHC)
LBL1	11	2.6	81	+
LBL2	9	4.1	75	+
LBL3	8	2.6	84	ND
LBL4	10	2.5	ND	ND
LBL5	12	1.1	78	+
LBL6	11	1.8	ND	+
DLBCL1	13	1.2	ND	+
DLBCL2	16	2.1	85	+
DLBCL3	12	1.7	65	ND
MZL1	11	3.9	74	+
MZL2	12	2.2	80	+

[1]Lymphomas were diagnosed and classified by histology, flow cytometry and immunohistochemistry using criteria outlined by the Hematopathology Subcomittee of the Mouse Models of Human Cancer Consortium (MMHCC, Morse et al., 2002). Age of animal at time of sacrifice is noted, as is the weight of the spleen. Flow analysis reflects the percentage of dispersed splenocytes that gated positive for IgM^+, $B220^{lo}$,$CD5^{lo}$ expression. Positive nuclear staining for the germinal center marker PAX5 as determined by immunohistochemistry is also indicated. LBL (lymphoblastic lymphoma), DLBCL (diffuse large B cell lymphoma), MZL (marginal zone lymphoma). ND-not determined.

Supplementary Figure 1



Supplementary Figure 1. Representative RLGS profile. *NotI-EcoRV-HinfI* RLGS profile for B cells sorted from pooled non-malignant splenocytes of six *TCL1-tg* mice. Approximate molecular weights for the first and second dimensions are indicated.