Figure S1. HVEM mutations and deletions in human lymphomas, Related to Figure 1

A, Chr. 1p36 deletions in a second series of FL (UNMC, n=198); inset: GISTIC analysis of DNA copy number indicates frequent homozygous loss; **B,** Frequency of deletions by zygosity in transformed FLs; **C,** Distribution of the percentages of HVEM-positive tumor cells in FL tissue specimens arranged on a TMA. Colors represent staining intensity; **D,** Expression of HVEM in Human FLs samples in HVEM wild type (WT: left) and HVEM mutated or deleted samples (mut/del: right); **E,** The number of cases presenting with the respective staining intensities for CD272 (BTLA) in the follicular lymphoma cells are shown; **F,** BTLA staining intensity in Human FLs in cases that are HVEM + or HVEM -; **G,** Numbers indicate breakdown of how individual TMA sections scored.

Figure S2. HVEM knockdown promotes FL development in vivo, Related to Figure 2

A, Kaplan–Meier analysis of tumor onset using a second shRNA against HVEM (shHVEM-2) compared to empty vector (blue: vector, n=11; red: shHVEM-2, n=12; p<0.01); **B,** Representative FACS analysis for surface HVEM in B lymphocytes isolated from normal spleen, control lymphomas (vavPBcl2-vector), and two independent lymphomas expressing the shRNA against HVEM (vavPBcl2-shHVEM); **C,** FACS analysis of MSCV-IRES-GFP expressing cells from control lymphomas before HPC transplantation and after tumor harvest; **D,** qRT-PCR expression of HVEM in FACS sorted CD4+ T-cells; **E,** qRT-PCR

measurement of expression of HVEM in FACS sorted T_{FH} cells; **F,** FACS analysis for the indicated surface markers on HVEM deficient (shHVEM) and control (Vector) lymphomas; **G,** quantification of Ki67 in vavPBcl2-vector and vavPBcl2-HVEM tumors (n=6; mean ± SD; * $p<0.01$); **H,** FACS analysis for the indicated surface markers on HVEM (shHVEM) deficient and control (Vector) lymphomas; **I,** Quantification of CD3+ cells in shHVEM or control lymphomas.

Figure S3. Analysis of variants in the VDJ region of mouse tumors, Related to Figure 3

A, Analysis of μ heavy chain transcripts from three samples of shHVEM mice to evaluate clonality and monitor clonotypes within the samples. Table represents clones amplified above 1% (control samples had none above 0.66%). Clones with the same VDJ junction and minimal differences within the V and JH segments are represented as variants in the last column; **B,** Evolution tree shows ongoing clonal evolution of the dominant clone by connecting variants observed in the CDR3 region with (VH8.12/D2.4/JH1) in shHVEM sample #2; **C,** Pie charts represent VH family usage of the three samples (and control) analyzed to globally assess the B cell repertoire in each sample. Abundant clonal proliferation in samples 2 and 3 accordingly show clear repertoire biases; **D,** Quantification of CD3+ cells in shBTLA and control (vector) lymphomas; **E,** Immunoblots on lysates from control (vector) and BTLA deficient (shBTLA) lymphomas probed as indicated.

Figure S4. Effect of solHVEM on murine and human FL B cells, Related to Figure 4.

A, Percentage of pSyk inhibition in primary human FL B cell samples ($n = 10$; all FL grade 1 and 2) upon solHVEM treatment was calculated by comparing the ratio of MFI of pSyk +/- solHVEM and correlated to BTLA MFI ($r = 0.697$, $p = 0.03$); **B and C,** FACS measurement of phosphorylated BTK (pBTK) in DoHH2 lymphoma cells that were stimulated with anti-IgG in the presence of absence of solHVEM (10µg/ml) or the BTK inhibitor ibrutinib (10nM); quantified in (C) (*indicates $p < 0.01$); **D,** immunoblot on Myc/Bcl2 murine lymphoma cells after treatment with solHVEM (10µg/ml) probed as indicated; **E and F,** Quantification of phosphorylated SYK (pSyk) levels in DoHH2 lymphoma cells stimulated with anti-IgG in the presence or absence of solHVEM (10µg/ml) (*indicates $p < 0.01$); representative FACS measurement in (F).

Figure S5. Analysis of the lymphoid stroma in B cell lymphomas, Related to Figure 5.

A, Immunohistofluorescence staining of $CD20^{pos}$ B cells (red), $Transglutaminase^{pos}$ FRCs (green), and $CD21L^{pos}$ FDCs (blue) in reactive lymph nodes and two separate human follicular lymphoma tissue specimens; **B-D,** qRT-PCR measurement of TNFα (B), LTα (C), and LTb (D) in mouse Myc/Bcl2 lymphoma cells; **E,** Flowchart of the image processing for FRC density (Collagen I, red); briefly, images were thresholded and transformed to binaries images, then a watershed algorithm was applied and number of polygons evaluated and

analyzed by ImageJ software; **F,** Number of polygons indicates FRC density in control lymphomas (vector) and HVEM knockdown lymphomas showing no difference in FRC contribution. 40 areas were selected in the T cell zone and analyzed per mice (n=3 per each group).

Figure S6. Analysis of T_{FH} cell function in HVEM deficient lymphomas, Related to Figure 6. A and B, qRT-PCR measurement of the expression of receptors for IL-21 (IL-21ra: A), and IL-4 (IL-4ra: B) in B cells purified from control (vector) or HVEM deficient mouse lymphomas; **C,** FACS identification and sorting of human GC derived T_{FH} cells based on the markers $CD3^{pos}$, $CD4^{pos}$, $CD25^{neg}$, $PD1^{hi}$, $CXCR5^{hi}$, red: isotypic control; blue: staining with anti-BLTA antibody; **D,** Viability of purified murine T_{FH} cells (samples: n=4) that were cultured for 3 days with or without (UN) stimulation by anti-CD3/anti-CD28 in the presence or absence of the soluble HVEM ectodomain (solHVEM: 10µg/ml); **E and F,** Cell-Sorted GC-T_{FH} cultured with anti-CD3/anti-CD28 Mabs in presence or not of solHVEM, production of CXCL13(E) and IL-21(F) evaluated by ELISA.

Figure S7. SolHVEM activity against lymphomas, Related to Figure 7. A, FACS analysis of BTLA and HVEM expression in the indicated lymphoma cell lines; **B,** Analysis of cell proliferation across a panel of $BTLA^{hi}$ and $BTLA^{lo}$ lymphoma cell lines treated with solHVEM or a control protein (solTNFRSF18) (10µg/ml); **C,** Pictures of lymphomas treated with vehicle or solHVEM by local injection; **D,** Weight of tumors in (C) (n=3, mean ± SD, *p < 0.01); **E,** Systemic

(intravenous, 100μg) treatment with solHVEM or vehicle in animals engrafted with Myc/Bcl2 murine lymphomas and treated at the indicated time points (arrows); **F**, Diagram representing the concept of CAR-T cells that produce solHVEM locally and continuously at the lymphoma site in vivo; **G** and **H**, Viability (G) and activation state (H) of T cells (OT-1) treated with anti-CD3/anti-CD28 antibodies in the presence or absence of solHVEM (10 μg/ml); **I**, In vitro viability of DOHH2 human lymphoma cells exposed for 24h to the indicated ratio of control CAR-T cells (CAR-T/CD19) and solHVEM producing CAR-T cells (CAR-T/CD19/solHVEM), viability was assessed by FACS.

Supplemental Tables.

Table S1. Mutational analysis of FL samples, Related to Figure 1.

Table S2. Mutation and aCGH data sets and their sources, Related to Figure 1.

Table S3. GISTIC calls on FL and tFL samples, Related to Figure 1.

Table S4. HVEM sequencing primers, Related to Figure 1.

Table S5. Minimal deleted regions of Chr. 1p36, Related to Figure 1.

Table S6. Hairpin sequences for shHVEM and shBTLA, Related to Figure 2 and Figure 3.

Methods and Resources

Contact for Reagent and Resource Sharing

Further information and requests for reagents may be directed to, and will be fulfilled by the Lead Contact Hans-Guido Wendel (wendelh@mskcc.org)

Experimental Model and Subject Details

Generation of Mice

All mice were housed in the specific-pathogen-free animal facility at the Memorial Sloan Kettering Cancer center and all animal studies were approved by the MSKCC IACUC committee (protocol-07-01-002). Unless indicated all mice studies used female mice on the C57Bl/6J background that were between 6 and 10 weeks of age. Wildtype mice were obtained from Jackson Laboratory (Bar Harbor, ME).

The vavPBcl2 mouse model is described (Egle et al., 2004) and adapted to adoptive transfer to retrovirally transduced HPCs (Wendel et al., 2004). In