SUPPLEMENTAL METHODS

Cell lines and culture conditions

KSHV+ primary effusion lymphoma (PEL) cell lines (BC1, BC2, BC3, BC5, BCBL1, BCP1), EBV+ lymphoblastoid cell line LCL9001, EBV+ diffuse large B-cell lymphomas (DLBCL) with immunoblastic features (IBL1, IBL4), and EBV+ effusion DLBCL BCKN1 were cultured in RPMI-1640 medium (Invitrogen) supplemented with 50μg/mL Gentamicin (Sigma-Aldrich) and 10% or 20% heat-inactivated fetal bovine serum, respectively. Uninfected DLBCL cell lines (Ly2, Ly7) were cultured in IMDM (Invitrogen) supplemented with 50μg/mL Gentamicin and 10% FBS. Multiple myeloma (MM) cell lines U266 and MM1S were cultured in RPMI supplemented with 15%FBS and 50μg/mL Gentamicin, while JJN3 was cultured in 40%IMDM /40% DMEM plus 20% FBS supplemented with 50μg/mL Gentamicin. Hodgkin's lymphoma (L428, L1236) and Burkitt's lymphoma cell lines (BJAB, Namalwa) were maintained in RPMI-1640 supplemented with Gentamicin and 10% FBS. All of the PEL cell lines and IBL1 were established in our laboratory from lymphomatous effusions except BCBL1,which was obtained from the AIDS and Cancer Specimen Bank. Burkitt lymphoma cell lines Namalwa and BJAB as well as the multiple myeloma cell line U266 were obtained from the American Type Culture Collection (ATCC). JJN3 cell line and Hodgkin cell lines (L428, L1236) were obtained from DSMZ. DLBCL cell lines LY2 and LY7 were provided by Riccardo Dalla-Favera (Columbia University). MM1S cell line was provided by Dr. Giorgio Inghirami lab at Weill Cornell Medicine. Human CAG MM cell line stably expressing the HSV-TK-eGFPluciferase fusion protein was kindly provided by Malcolm Moore (Memorial Sloan-Kettering Cancer Institute) and was cultured in RPMI +20%

FBS. All cell lines were maintained in a humidified incubator at 37°C and 5% CO_2.

The xenograft cell line BC3NFκB-luc#6 was generated as previously described(1). The double reporter cell line BC3NFRen-luc#3 was generated by transduction of this cell line using a lentiviral construct expressing renilla luciferase controlled by a constitutive promoter (retroviral LTR). These cells were maintained in RPMI-1640 supplemented with 15% FBS and 50μg/mL Gentamicin, as well as 1.2mg/mL Geneticin (Life Technologies) to maintain clonal selection.

Bone marrow specimens were obtained from multiple myeloma patients at the New York–Presbyterian Hospital under informed consent as part of an Institutional Review Board-approved study in accordance with the Declaration of Helsinki. Primary $CD138^+$ human myeloma cells were isolated by Ficoll-Hypaque density gradient centrifugation. The $CD138^+$ bone marrow myeloma cells were then enriched from this fraction to >95% purity using an automated MACS CD138 MicroBeads system (Miltenyi Biotechnology, Inc., Auburn, CA). Coculturing $CD138^+$ cells with HS5 stromal cells did not alter their sensitivity to 6-ETI treatment for short treatment course. PBLs were harvested from normal blood using Ficoll-Paque (Sigma-Aldrich) as described previously. Cells were plated in RPMI-1640 supplemented with 20% FBS.

Pancreatic adenocarcinoma PA-Tu-8988T cell line was kindly provided by Lewis Cantley's lab (Weill Cornell Medicine). Knock out of ADK in this cell line was achieved by transducing these cells with ADK shRNA Lentiviral Particles (Santa cruz) and selecting the stable cell line using 2μg/ml Puromycin.

Cell cycle assays

BC3 cells in the exponential phase were seeded at a cell density of $2*10^5$ cells/mL. Cells were treated with 0.1% DMSO or with 6-ETI at a final concentration of 1μM 6-ETI for 24hrs for cell cycle. Post-treatment, cells were harvested, fixed and stained in Propidium Iodide (Roche). Subsequently, stained cells were analyzed by flow cytometry using BD FACS Aria II SORP cell sorter, and phases of the cell cycle modeled using FlowJo v7.6.5.

Apoptosis assays

BC3 and IBL1 cells in the exponential phase were seeded at a cell density of 0.2 million cells/ml. Cells were treated with 0.1%DMSO or with 6-ETI at a final concentration of 500nM or 5μM for 24, 48 or 72hrs. Post-treatment, cells were harvested and stained with 2.5μL of Alexa Fluor 647 Annexin V conjugate (Life technologies) and 2.5 μL of 7-AAD (BD biosciences) and incubated for 15 minutes in the dark at room temperature. Subsequently, 200μL of Annexin V binding buffer (BD Biosciences) was added to the cell suspension and cells analyzed by flow cytometry using BD biosciences LSRII machine.

Toxicity on normal primary tonsillar lymphocytes

Primary human tonsil specimens were obtained from the Weill Cornell/New York Presbyterian Institutional Biobank. Lymphocytes were isolated from human tonsil tissue by dissection and maceration in RPMI media. Lymphocyte-containing media was passed through a 70μm filter and pelleted at 400g for 7 minutes. RBC were lysed for 5 minutes in RBC lysing solution (0.15M ammonium chloride, 10mM potassium

bicarbonate, 0.1M EDTA). After dilution to 50ml with RPMI, lymphocytes were filtered through a 0.4µm filter, counted and pelleted a second time. Total B cells were isolated using magnetic cell sorting according to manufacturer instructions (Miltenyi Cat#130-091-151). Cells were plated at 2 million cells/ml in RPMI with 20% FBS and 100µg/ml Primocin (Invivogen) and laid over gamma-irradiated CDW32 feeder cells. Cells were then treated with DMSO, 1µM or 10µM 6-ETI for 24hrs. For the multi-color flow cytometry immunophenotyping, cells were pelleted and resuspended in FACS Wash buffer (PBS+0.5%FBS+0.1%Sodium Azide) containing B cell phenotype panel as follows for 15 minutes on ice: CD19-PE (16µl), CD38-PECy7 (6µl, BD Cat#560667), IgD-PerCP Cy5.5 (5µl, BD Cat#561315), CD138-APC (5µl, BD Cat#347207), CD27-APC H7 (5µl BD Cat#560222). Volumes indicated were based on titrating the individual antibodies on primary tonsil lymphocyte specimens. After incubation, 100µl FACS Wash was added and pelleted lymphocytes were washed with a further 200µl of FACS Wash prior to being resuspended in 200µl FACS Wash for analysis. Data was acquired on a BD LSR2 Flow Cytometer and analyzed using FlowJo software .The frequency of plasma cells was assessed based on singlet-gated cells expressing CD138.

6-ETI effects on DNA synthesis using EdU click-IT assay

BC3 and U266 cells were seeded at a density of 0.5 million cells /ml and treated with DMSO only, 1µM or 10µM 6-ETI for BC3 cells and 5µM 6-ETI for U266 cells for 24hrs. At 24hrs post 6-ETI treatment, 0.5 million cells of each of the treated conditions were labeled with 10µM of EdU for 2 hours then fixed for 30min using 2% paraformaldehyde supplemented with 0.3M sucrose. Cells were then washed in PBS and permeabilized with 0.2% saponin, PBS with 1%BSA and 0.3M sucrose for 20 min at

room temperature. During this incubation time, a click-iT cocktail mix containing click iT reaction buffer, $CuSO_4$, alexafluor488 and click iT reaction buffer additive was prepared according to the manufacturer's instructions. Cells were washed in PBS after permeabilization and the click-iT cocktail mix was added for 30 min at room temperature in the absence of light. To visualize DNA, cells were counterstained with Hoechst (1:1000) for another 30 min then washed and resuspended in PBS with1% BSA. 20µl of the resuspended cells were combined with 20µl of mounting media Fluoromount-G (Southern biotech) and cells were mounted on Ibidi µ-slides. Cells were imaged using DeltaVision image restoration microscope (Rockefeller center).

Immunofluorescence staining, image acquisition and analysis

$1*10^5$ treated cells were pelleted into a 96-well round bottom plate. Cells were washed once with PBS, and fixed in PBS+2%paraformaldehyde+0.03M sucrose for 15min at room temperature. Cells were then washed twice with PBS+1% BSA+0.03M sucrose, permeabilized with PBS+0.2% saponin+1% BSA+0.03M sucrose (IFA Wash) for 15min at room temperature, and blocked for 1 hour in PBS+5% normal goat serum. Cells were then stained in IFA Wash containing phospho-histone-H2A.X (ser139) antibody (γ-H2AX) at 1:100 dilution overnight at 4˚C. Following two washes with PBS+1% BSA+0.03M sucrose, cells were re-permeabilized and re-blocked as above prior to incubation for 1 hour at room temperature with anti-rabbit-AlexaFluor594+Hoechst diluted 1:1000 in IFA Wash. Cells were then washed twice with PBS+1% BSA+0.03M sucrose, resuspended in 30µl Fluoromount-G (Southern Biotech) and transferred to 16-well Ibidi µ-slides for imaging. Images were taken on an Applied Precision Deltavision Image Restoration Microscope using Softworx analysis software.

Z-stacks with a 0.2μm step size were taken at 100x magnification. Stacks were subjected to deconvolution analysis and projections were made superimposing 5 representative z-planes to generate the final image.

6-ETI synthesis

A mixture of ethanethiol (26g, 419mmol), methanol (16ml), and sodium hydroxide (6.7ml, 20% aqueous solution) was cooled to 0°C, using an ice water bath. The mixture was stirred for 30 minutes, at which point 6-chloropurine riboside (2g, 7mmol) was added. The resulting solution was slowly warmed to ambient temperature and stirred for 12 hours, then was neutralized with glacial acetic acid (2.4ml). Solvent was then removed using rotary evaporator; a small amount of bleach was placed in the solvent trap to quench any remaining ethanethiol. The resulting crude product was dissolved in acetone (100mL) and then passed through a short Celite plug to remove minor impurities. Concentration *in vacuo* provided 6-ETI as a white powder (2.11g, 6.7mmol, 96%). ^{1}H NMR (500 MHz, CD_3OD) δ 8.69 (s, 1H), 8.59 (s, 1H), 6.09 (d, *J* = 5.8 Hz, 1H), 4.75 (dd, *J* = 5.5, 5.4 Hz, 1H), 4.37 (dd, *J* = 5.1, 3.3 Hz, 1H), 4.18 (dd, *J* = 6.2, 3.1 Hz, 1H), 3.91 (dd, *J* = 12.4, 2.9 Hz, 1H), 3.78 (dd, *J* = 12.4, 3.1 Hz, 1H), 3.42 (q, *J* = 7.4 Hz, 2H), 1.45 (t, *J* = 7.4 Hz, 3H).

Microplate-based NF-κB inhibition assay

Exponentially growing BC3-derived reporter cell lines (BC3NFκB-luc#6 or BC3NFRen-luc#3), and unrelated luciferase expressing U251-pGL3 cells were resuspended in respective RPMI-1640 complete media, and plated in a 96-well tissue culture microplate at $7.5*10^5$ cells/mL and $3*10^5$ cells/mL, respectively, in the presence or absence of

varying concentrations of test compounds. After 24 hours of incubation, the luciferase activity was measured using Steady-Glo Luciferase Assay System or Dual-Glo Luciferase assay system (Promega, Madison, WI), according to the manufacturer's instructions.

Lytic reactivation qRT-PCR and flow

BC3 cells in the exponential phase were plated at $2*10^5$ cells/mL, and treated with 100nM, 1μM, or 10μM 6-ETI for 6, 24, or 48 hours. Expression of viral latent and lytic genes was determined using qRT-PCR as previously described (2).

Lytic reactivation flow was performed as previously described (2). JSC1 B3.1 cells in the exponential phase were seeded at $2*10^5$ cells/mL and treated with 1 or 10μM 6-ETI for 24 or 48 hours, or 20ng/mL lytic inducer TPA for 48 hours.

***In vivo* xenograft model of EBV-associated immunoblastic lymphoma**

The *in vivo* xenograft model of EBV-associated immunoblastic lymphoma was generated by injecting NOD-SCID mice (obtained from Jackson Laboratory Stock# 001303) subcutaneously on the flank region with $5*10^6$ IBL-1 cells in 200μl of 50:50 matrigel/ cell suspension per mouse. After forming palpable or measurable tumors, mice were randomly grouped (n=3 per group) and treated intraperitoneally with vehicle PBS-Tween80 or 6-ETI (300mg/kg/day) for 5 treatments over the course of 10 days where drug was administered three times a week. Tumor size was assessed by caliper measurement three times a week, and tumor volume (V) was calculated using the formula V= (length × Width^2)/2 where L= length or largest tumor diameter and

W=width or the perpendicular tumor diameter. Relative tumor volume was determined according to the formula Vd/V0 where Vd is tumor volume on the day of measurement, and V0 represents the tumor volume on day 0 when the treatment started. At the end of the treatment course, two of the control mice were treated with vehicle PBS-Tween80 or 6-ETI (300mg/kg) for 24hr after which the tumors were harvested, fixed in 10% formalin and embedded in paraffin for histological examination and immunohistochemistry.

Immunohistochemistry

Cases were collected from New York Presbyterian Hospital–Weill Cornell, and obtained with the approval and according to guidelines of the institutional review board. Primary cases used for immunohistochemistry included two primary effusion lymphomas, one extracavitary primary effusion lymphoma, two multiple myelomas, and two plasmablastic lymphomas. Immunohistochemical staining of ADK (Polyclonal Rabbit antibody, dilution 1:25, Sigma) was accomplished using the Bond III Autostainer (Leica Microsystems, Illinois, USA). Formalin fixed and paraffin-embedded tissue sections were first baked and deparaffinized. Antigen retrieval was then followed by heating the slides in Bond Epitope Retrieval Solution 1(ER1) (Leica Microsystems) at 99-100°C for 30 minutes. Sections were then incubated sequentially with the primary antibody, post-primary (equivalent to secondary antibody), polymer (equivalent to tertiary antibody), endogenous peroxidase block, diaminobenzidine (DAB) and hematoxylin for 15, 8, 8, 5, 10 and 5 minutes (Bond Polymer Refine Detection; Leica Microsystems), respectively. Finally the sections were dehydrated in 100% ethanol, and mounted in Cytoseal^{TM} XYL (Richard-Allan Scientific, Kalamazoo, MI). Immunohistochemical staining of Cleaved Caspase 3 (Polyclonal Rabbit antibody, dilution 1:300, Cell Signaling) was

accomplished as above, and used to assess apoptosis *in vivo* in mice with IBL1 xenografts treated with 6-ETI.

RNA sequencing data analysis

To search for single nucleotide variants (SNVs) and small indels in the human genome, raw data was mapped to human reference GRCh37 using Bowtie2 via Tophat2 v.2.0.11 (3) and each tumor sample was compared against the two normal BC3 samples using Strelka v.1.0.10 (4). Putative variants were annotated using snpEff v.3.3. Annotated variants were examined with a combination of custom shell scripts and manual visualization in Integrative Genomics Viewer (IGV) (5) to identify candidates that were 1) present in tumor samples but in neither of the normal samples; 2) of a type that was likely to affect the protein (non-synonymous, start codon loss, stop codon gain, frame shift); and 3) recurrent in the tumor samples. Once candidates were identified, expression (or lack thereof) across each of the exons and introns of the candidate was checked with HTSeq-counts v.0.6.1 (6) and visualization in IGV. To search for variants in the KSHV genome, Bowtie2 was used to map the raw data to genomic reference NC_009333.1. SNVs and small indels in characterized viral domains were detected using a combination of Strelka v.1.0.10 and Samtools mpileup v.0.1.18(7), and annotated in snpEff v.4.0E using a database constructed around the Genbank record. No recurrent variants survived filtration for minimum read depth of 10 and a variant type likely to affect the structure of the protein. Samples were searched for differential expression using HTSeq-counts v.0.6.1 and applying EdgeR v.3.6.7 (8) using the exact negative binomial test on gene counts normalized by the trimmed mean of M-values (TMM) method of tumor samples compared to BC3 normal samples. Gene expression

data from RNA sequencing was analyzed using Ingenuity Pathway Analysis software (Ingenuity® Systems, www.ingenuity.com).

References:

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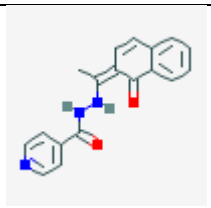
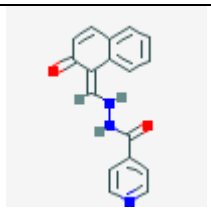
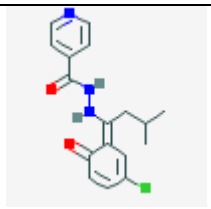
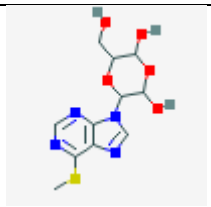
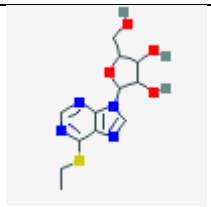
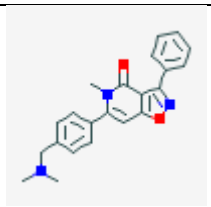
Supplemental Table 1. Small molecule screening data

Category	*Parameter*	*Description*
Assay	Type of assay	Cell-based luciferase reporter assay
	Target	NF-κB
	Primary measurement	Detection of luciferase activity
	Key reagents	BC3 NFκB-luc #6 cells, BC3 NFRen- luc #3 cells, U251-pGL3 cells, Steady-Glo luciferase assay system, Dual-Glo luciferase assay system
	Assay protocol	Cells resuspended in media plated with compounds, cell lysis and luciferase assay at 24 hours
	Additional comments	
Library	Library size	1981
	Library composition	The NCI Diversity Set was derived from the DTP small molecule library of over 140,000 compounds as a diverse set of representative compounds amenable to forming structure-based hypotheses.
	Source	NCI Diversity Set
	Additional comments	
Screen	Format	96-well plates at $7.5*10^5$ cells/mL (BC3 NFκB-luc #6)
	Concentration(s) tested	5μM compound, 0.05% DMSO
	Plate controls	No drug (0.05% DMSO-media only) treated cells and 0.05% DMSO media only (no cells)
	Reagent/ compound dispensing system	Manual
	Detection instrument and software	Tecan GENios Pro; XFLUOR4GENIOSPRO V4.53
	Assay validation/QC	Median Z´ factor = 0.83 (range 0.52 – 0.92); Assay reproducibility r^2=0.93
	Correction factors	Plate background (media only)
	Normalization	Percent Luc inhibition (=100*[1-$RLU_{test\ compound}$/$RLU_{no\ drug\ control}$])
	Additional comments	
Post-HTS analysis	Hit criteria	50% inhibition of luciferase in treated cells compared to control untreated cells
	Hit rate	Primary screen 3%
	Additional assay(s)	Primary hit confirmation in BC3 NFκB-Luc#6 by cherry-picking from the Diversity Set compounds plate (98% confirmation rate); followed by dose-response testing of confirmed hits prepared from power samples for IC_{50} determination in BC3 NFκB-luc#6, BC3 NFRen-luc #3 cells, and control U251-pGL3; 66 analogs of 3 primary hits from Phase I SAR study and 39 additional analogs from Phase II SAR study on the DTP Repository and ChemNavigator library were tested in BC3 NFκB-Ren-luc #3 cells
	Confirmation of hit purity and structure	Primary hits checked for purity by LC/MS; Primary lead compound (6-ETI) resynthesized (see Methods)

Supplemental Table 2. Hit compound IC_{50}s in primary and secondary screens

NSC#	Structure	Primary screen (5μM) % inhibition	IC_{50} (Mean±SD, μM)				
			Dual reporter BC3NFRen-luc#3			BC3NFκB-luc#6	U251-pGL3
			Firefly (NFκB)	Renilla (constitutive)	Mean IC_{50} Renilla/Firefly ratio		
54044		62.9	2.00±0.46	>20	>10	1.65	>20
51355		N/A	0.67±0.14	10.03±0.64	15.0	ND	ND
27604		N/A	1.06±0.07	>20	>18.9	ND	ND
111702		54.5	3.09±0.15	8.25±2.18	2.7	2.74	>20
39368		N/A	0.27±0.01	1.55±0.54	5.7	ND	ND
339908		55.4	0.25±0.03	>20	>80	4.5	3.69

Compounds in bold were identified in the primary screen. Compounds listed below primary hits are analogs of the respective compounds identified by re-screening. Abbreviations: N/A= not applicable, ND= not determined; Structures: Grey=H, Blue=N, Red=O, Green=Cl, Yellow=S

Supplementary Table 3. Examination of reads mapping to adenosine kinase (ADK) gene

				Number of reads in cell line								
EXON:	Start	End	Length	**BC3 #1**	**BC3 #2**	∗2-3low	♦3-5low	♥NSCE-1	NSCE-2	∗NSCE-3	NSCE-6	NSCE-7
ENSE00002319053	75910960	75911101	141	31	26	3	59	28	53	1	38	54
ENSE00001873962	75934174	75934376	202	6	6	1	0	3	3	2	5	2
ENSE00001458497	75936444	75936644	200	129	125	12	100	54	252	7	180	276
ENSE00003178627	75960522	75960596	74	110	123	10	103	74	226	3	176	274
ENSE00003157632	75984296	75984349	53	70	54	6	55	40	101	2	90	147
ENSE00001178540	76074425	76074503	78	170	174	5	157	96	314	8	296	397
ENSE00001024568	76153899	76154071	172	432	405	24	327	275	267	22	264	♣1
ENSE00001024570	76158229	76158337	108	250	259	22	213	144	179	15	172	♣0
♦ENSE00001310865	76285014	76285184	170	442	448	37	428	264	270	25	255	♣0
ENSE00001910381	76287518	76288057	539	14	13	0	17	7	2	1	3	♣0
♥ENSE00001248835	76349040	76349075	35	14	17	3	30	18	♣0	4	♣0	♣0
ENSE00001248829	76360137	76360251	114	361	345	48	358	210	♣0	33	♣0	♣0
ENSE00001248825	76429941	76430027	86	214	211	16	210	135	413	15	341	492
ENSE00001390313	76468079	76469061	982	811	770	84	945	508	1585	36	1317	1894
		Total Reads		3054	2976	271	3002	1856	3665	174	3137	3537

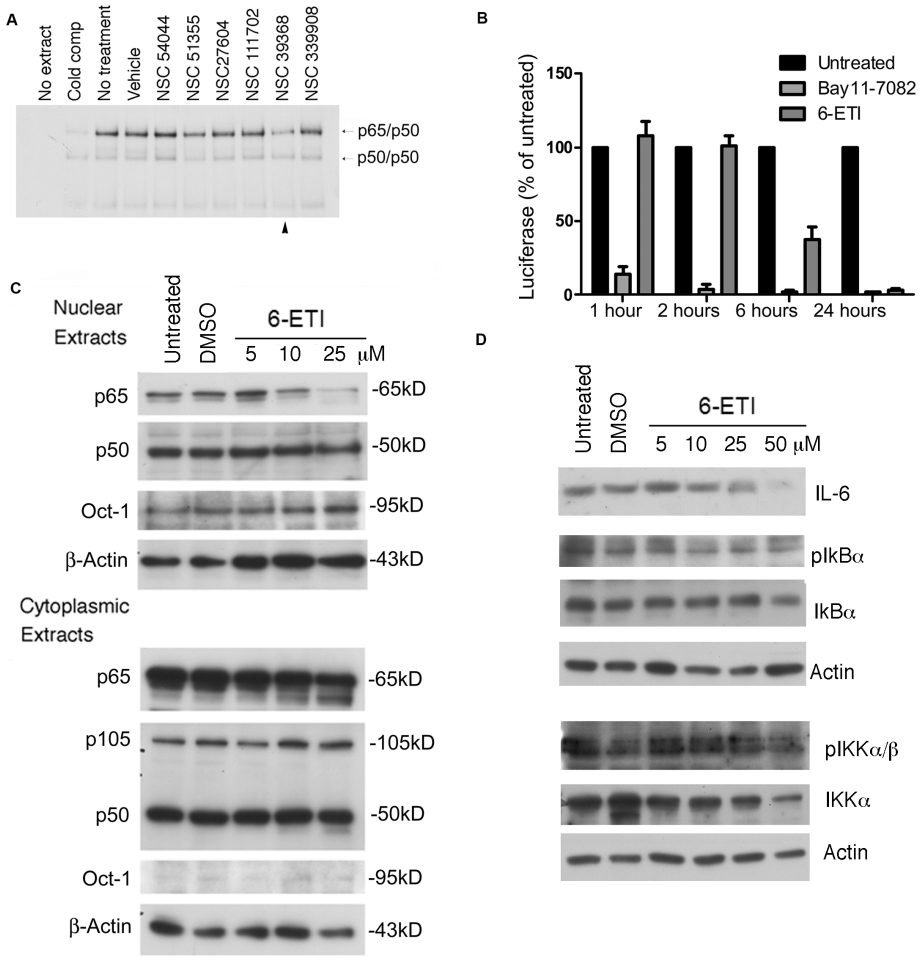
The number of reads mapping to fourteen individual exons of the ADK gene in two control parental BC3 (bold), and seven 6-ETI resistant cell lines is shown. Table indicates start and end positions, as well as length of each exon. All resistant cell lines harbor modifications, including nonsynonymous mutations (G239E in 3-5low and E243K in NSCE-1), underexpression (2-3low and NSCE-3), and exon loss (NSCE-2, NSCE-6, and NSCE-7). Key: ∗Sample with under-expression; ♦Exon / sample containing the G239E variant; ♥Exon / sample containing the E243K variant; ♣Exon lost.

Supplemental Table 4. Pathways enriched by expression analysis on transcriptome sequencing data

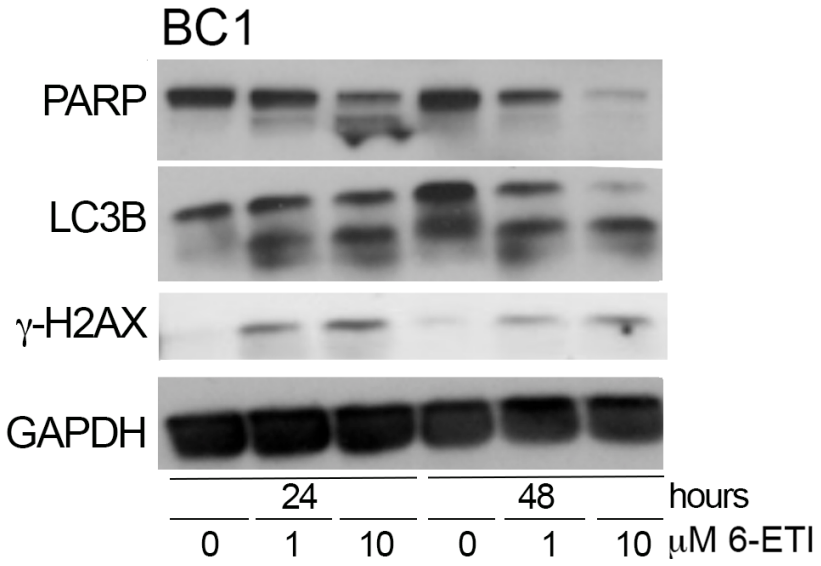
Ingenuity Canonical Pathways	-log(p-value)
Protein Ubiquitination Pathway	1.57E01
NRF2-mediated Oxidative Stress Response	6.17E00
Glucocorticoid Receptor Signaling	6.03E00
Prostate Cancer Signaling	5.81E00
Aryl Hydrocarbon Receptor Signaling	5.26E00
Antigen Presentation Pathway	4.35E00
Aldosterone Signaling in Epithelial Cells	4.07E00
Estrogen Receptor Signaling	4.03E00
Hypoxia Signaling in the Cardiovascular System	3.9E00
Androgen Signaling	3.81E00
PPARα/RXRα Activation	3.73E00
PPAR Signaling	3.69E00
Telomerase Signaling	3.5E00
PI3K/AKT Signaling	3.49E00
eNOS Signaling	3.15E00
Mitotic Roles of Polo-Like Kinase	3.05E00
Spermidine Biosynthesis I	3.03E00
S-adenosyl-L-methionine Biosynthesis	3.03E00
iNOS Signaling	3.02E00
LXR/RXR Activation	2.83E00
VDR/RXR Activation	2.6E00
Methionine Degradation I (to Homocysteine)	2.58E00
Cysteine Biosynthesis III (mammalia)	2.42E00
TR/RXR Activation	2.38E00
Neuregulin Signaling	2.36E00
Cell Cycle: G1/S Checkpoint Regulation	2.28E00
Endoplasmic Reticulum Stress Pathway	2.23E00
Chronic Myeloid Leukemia Signaling	2.22E00
Xenobiotic Metabolism Signaling	2.19E00
Polyamine Regulation in Colon Cancer	2.17E00
Huntington's Disease Signaling	2.12E00
Role of JAK1, JAK2 and TYK2 in Interferon Signaling	2.06E00
Primary Immunodeficiency Signaling	2.05E00
JAK/Stat Signaling	2.03E00
Gluconeogenesis I	2.01E00
Cell Cycle: G2/M DNA Damage Checkpoint Regulation	1.95E00
Superpathway of Cholesterol Biosynthesis	1.92E00
Amyloid Processing	1.92E00
Cyclins and Cell Cycle Regulation	1.92E00
EIF2 Signaling	1.91E00
RAR Activation	1.9E00
Hereditary Breast Cancer Signaling	1.82E00
Purine Nucleotides De Novo Biosynthesis II	1.8E00
Superpathway of Methionine Degradation	1.76E00

P2Y Purigenic Receptor Signaling Pathway	1.72E00
Chondroitin Sulfate Degradation (Metazoa)	1.66E00
Cholesterol Biosynthesis I	1.66E00
Cholesterol Biosynthesis II (via 24,25-dihydrolanosterol)	1.66E00
Cholesterol Biosynthesis III (via Desmosterol)	1.66E00
Oncostatin M Signaling	1.65E00
Stearate Biosynthesis I (Animals)	1.65E00
Role of BRCA1 in DNA Damage Response	1.63E00
Dermatan Sulfate Degradation (Metazoa)	1.6E00
ERK5 Signaling	1.59E00
Interferon Signaling	1.59E00
Nitric Oxide Signaling in the Cardiovascular System	1.56E00
Molecular Mechanisms of Cancer	1.55E00
tRNA Charging	1.52E00
Granzyme B Signaling	1.49E00
Melatonin Signaling	1.48E00
Epoxysqualene Biosynthesis	1.45E00
Spermine Biosynthesis	1.45E00
Palmitate Biosynthesis I (Animals)	1.45E00
Fatty Acid Biosynthesis Initiation II	1.45E00
Putrescine Biosynthesis III	1.45E00
Glycine Biosynthesis I	1.45E00
Glutamine Degradation I	1.45E00
Granzyme A Signaling	1.44E00
Role of IL-17F in Allergic Inflammatory Airway Diseases	1.44E00
FLT3 Signaling in Hematopoietic Progenitor Cells	1.42E00
Role of NFAT in Cardiac Hypertrophy	1.41E00
Pancreatic Adenocarcinoma Signaling	1.38E00
Prolactin Signaling	1.38E00
Regulation of eIF4 and p70S6K Signaling	1.38E00
fMLP Signaling in Neutrophils	1.37E00
Corticotropin Releasing Hormone Signaling	1.36E00
GADD45 Signaling	1.35E00
Purine Nucleotides Degradation II (Aerobic)	1.35E00
PDGF Signaling	1.31E00
DNA Methylation and Transcriptional Repression Signaling	1.31E00

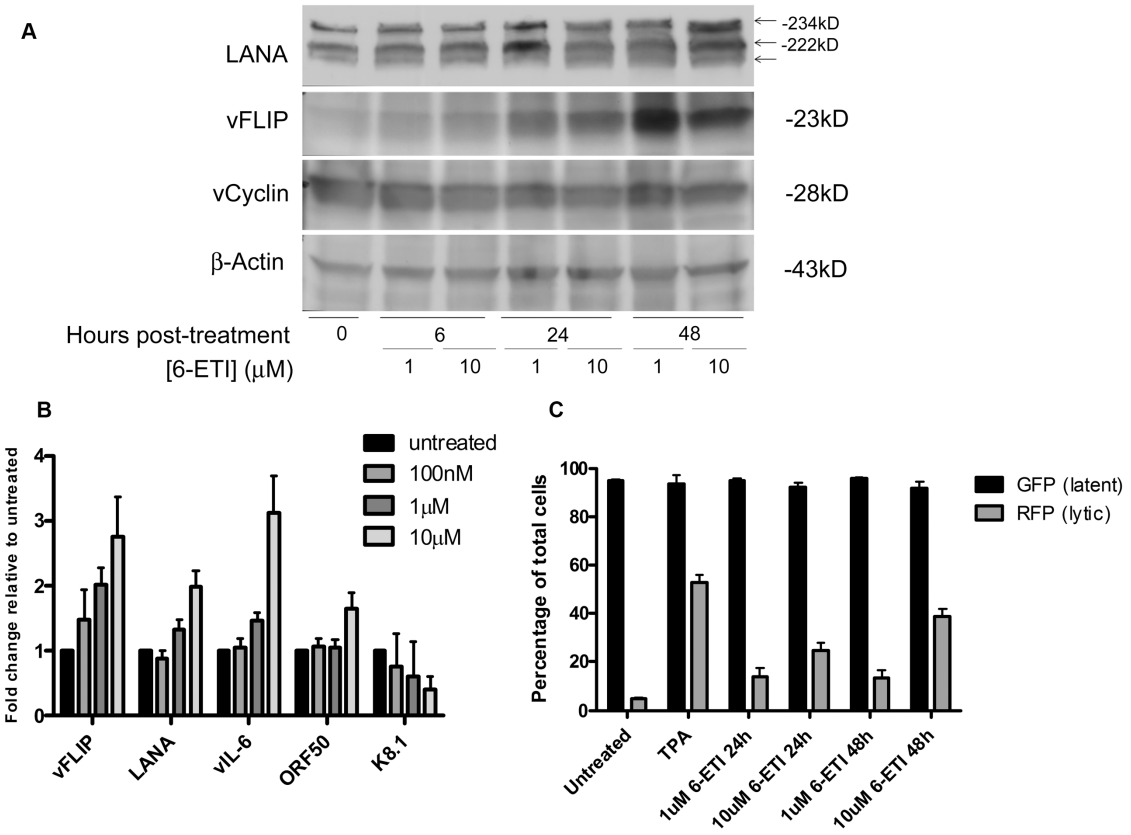
Ingenuity Pathway Analysis software was used to analyze gene expression changes from RNASeq of 6-ETI-resistant clones. Results are presented as a list of enriched pathways meeting a cutoff p-value of ≤0.05.



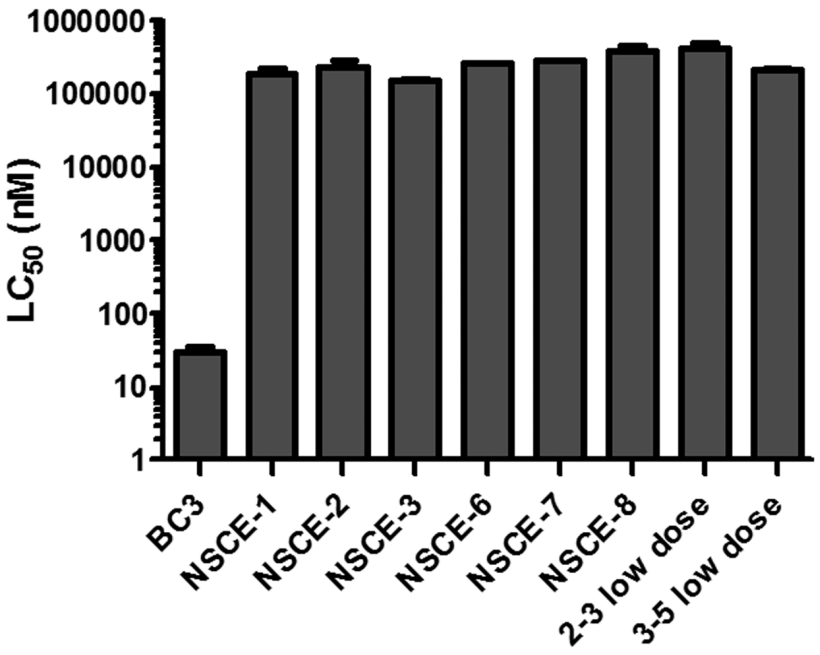
Supplemental Figure 1. 6-Ethylthioinosine is selective to PEL cells. (A) Nuclear extracts from BC3 cells treated for 24 hours with 25μM of the indicated hit compounds were analyzed by EMSA for binding to an NF-κB response element. Control lanes include no compound, competition with cold probe, untreated and DMSO-treated samples. Experiment was performed three separate times. A representative blot is shown. **(B)** BC3 NFκB-luc#6 cells were plated at $5*10^5$ cells/ml in RPMI with 15% FBS, treated with 10μM NF-κB inhibitor Bay11-7082 or 50μM NSC39368 (6-ETI). At the indicated time points, cells were lysed and analyzed by the Luciferase Assay System. Luciferase activity was normalized to untreated cells at each time point, and plotted using GraphPad Prism. Graph represents the mean±SEM of three independent experiments. **(C)** Nuclear and cytoplasmic extracts from BC3 cells treated for 24 hours with the indicated doses of 6-ETI were analyzed by western blotting for NF-κB components p65 and p50, as well as a control transcription factor Oct-1. Actin was used as a loading control, and a representative blot is shown from three independent experiments. **(D)** Whole cell extracts obtained from BC3 cells treated at the indicated concentrations of 6-ETI for 24 hours were analyzed by western blotting for upstream NF-κB pathway components phospho- and total IκBα, phospho-IKKα/β, and total IKKα. Actin served as the loading control, and a representative blot of at least three independent experiments is shown.



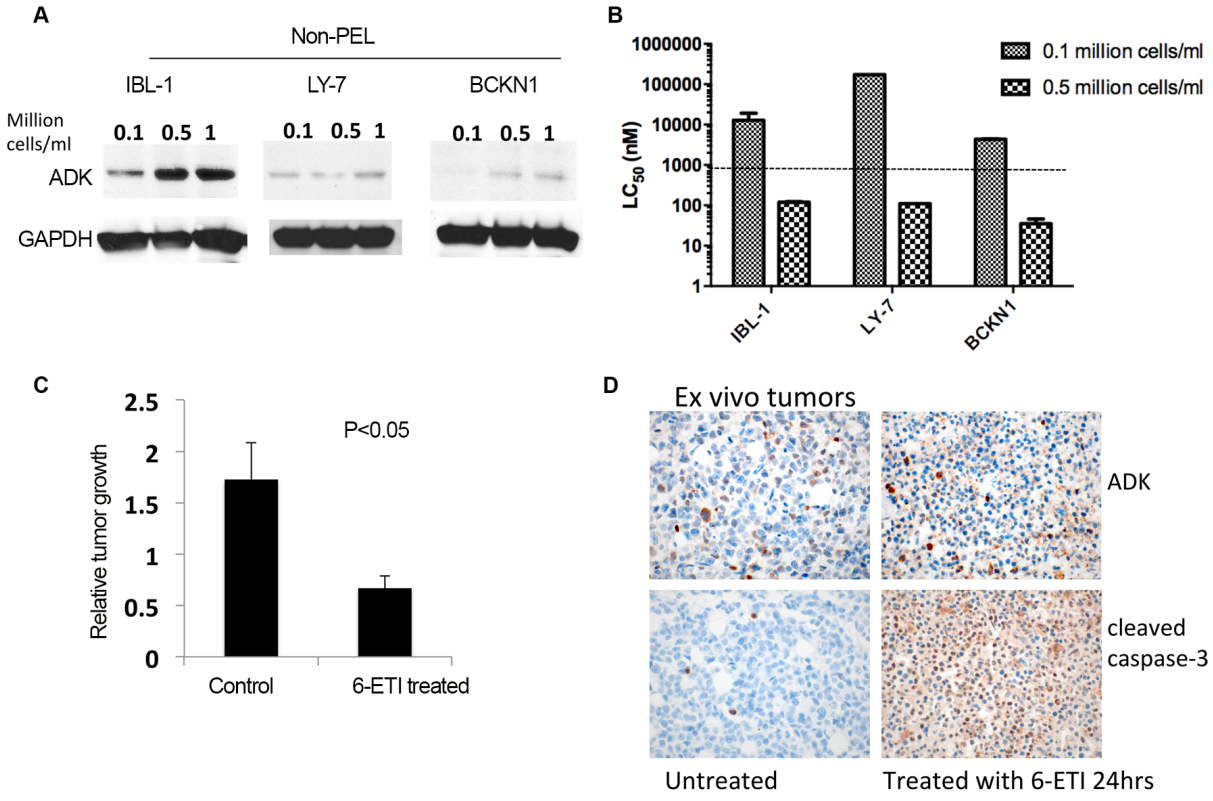
Supplemental Figure 2. Mechanism of 6-ETI induced cell death in the BC1 PEL cell line. BC1 cells treated with the indicated doses of 6-ETI at the shown time points were analyzed by western blotting on whole cell extracts for PARP cleavage as an indicator of apoptotic induction and for LC3B cleavage as an indicator of autophagosome formation. Immunoblotting for γH2AX is shown after treatment with 6-ETI at the indicated time points and doses. GAPDH was used as loading control.



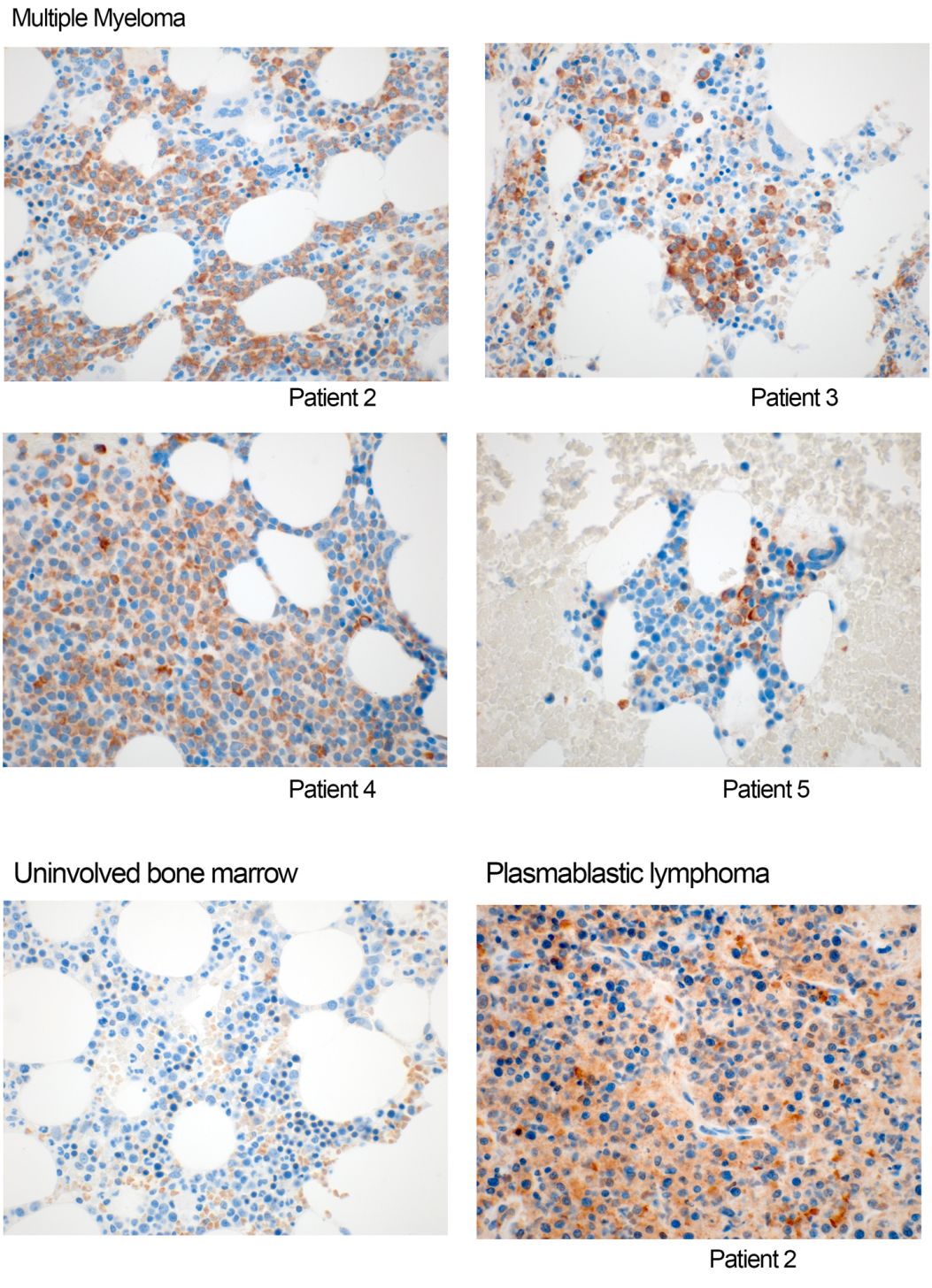
Supplemental Figure 3. Effect of 6-ETI on KSHV latent and lytic genes. (A) BC3 cells treated for the indicated time points at two different doses of 6-ETI were analyzed by western blotting on whole cell extracts for KSHV latent proteins LANA, vFLIP, and vCyclin. Results shown are representative of at least three independent experiments.**(B)** RNA was extracted from BC3 cells treated for 24 hours at three different doses of 6-ETI, followed by qPCR analysis for selected KSHV latent and lytic genes. Results analyzed using the ΔΔCt method were plotted as fold changes in expression relative to untreated cells. The graph is mean±SEM and representative of at least three independent experiments. **(C)** JSC1 B3.1 cells treated with the lytic inducer TPA (20ng/mL for 48 hours) or 1 or 10μM 6-ETI were analyzed by two-color flow for GFP (latent) or RFP (lytic) expression at 24 or 48 hours after treatment as indicated. Results represent the mean±SEM of three independent repeats.



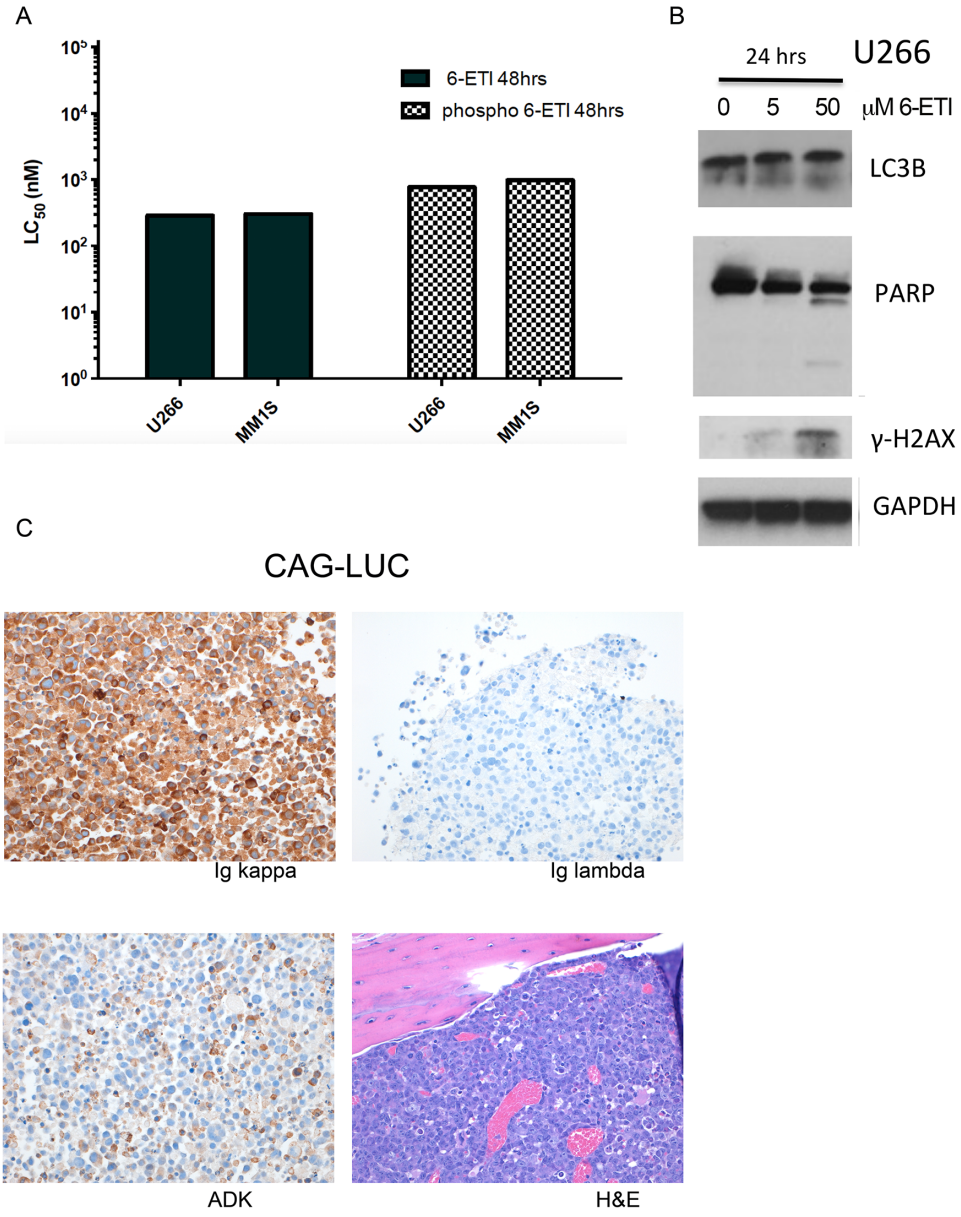
Supplemental Figure 4: Generation of 6-Ethylthioinosine resistant cell lines. 6-ETI resistant cell lines generated by growing BC3 cells under increasing doses of 6-ETI in multiwall plates were tested for 6-ETI resistance using a compound viability dose curve, with LC_{50}s determined by online EC_{50} software through the use of the Cell Titer-Glo ATP content assay at 48 hours. Results presented are the mean±SEM of at least three independent experiments.



Supplemental Figure 5. Adenosine kinase levels determine lymphoma sensitivity to 6-ETI. (A) Three 6-ETI-resistant lymphoma cell lines (IBL1, LY7 and BCKN1) were grown at various plating densities as indicated. Whole cell extracts obtained at 24 hours were examined by western blotting for ADK levels, and GAPDH was used as a loading control. A representative blot of three independent experiments is shown. **(B)** Three 6-ETI-resistant lymphoma cell lines (IBL1, LY7 and BCKN1) were plated at 1 or $4*10^5$ cells/mL in RPMI with 20% FBS, followed by treatment with 6-ETI at several doses for 48 hours. LC_{50}s were determined through online EC_{50} software, on the basis of CellTiter-Glo luciferase assay. Results shown are mean±SEM of three independent experiments. **(C)** Tumor growth in three IBL1 xenografted mice treated with 6-ETI compared to three vehicle control mice; results were analyzed by t-test ($p<0.05$). **(D)** Xenograft tumors from control and treated mice stained by immunohistochemistry with ADK and cleaved caspase 3 antibodies *ex vivo*. Original magnification is 60X.



Supplemental Figure 6. ADK expression in primary multiple myeloma and plasmablastic lymphoma patient specimens. Immunohistochemistry staining for ADK expression in four multiple myeloma samples, and additional plasmablastic lymphoma patient sample showing high levels of ADK expression in comparison to non-involved bone marrow (original magnification: 60X).



Supplemental Figure 7. Characterization of multiple myeloma. (A) Response of multiple myeloma cell lines U266 and MM1S to 6-ETI and phospho-6-ETI at 48hrs post-treatment. Results are mean±SEM of three independent experiments **(B)** U266 cells were treated with the indicated doses of 6-ETI at the shown time points. Cell lysates were subjected to western immunoblotting to detect LC3B cleavage, PARP and its cleaved form, and phosphorylated H2AX (γ-H2AX) as a marker of DNA damage response. Western blot shown is representative of three independent experiments. **(C)** Prior to injection into mice, CAG-luc cells were confirmed to express cytoplasmic immunoglobulin, as expected for a MM, and to be monotypic for kappa light chains. ADK expression was confirmed by IHC. The lower right panel shows localization to the bone marrow when injected into NOD-SCID mice (original magnification: 60X).