Supplemental Methods

CAR-T cell generation

PBMCs were stimulated by plate-bound OKT3 (1 ng/mlL) and CD28 antibodies (1 ng/mL) (BD Bio Biosciences, Mountain View, CA), and then cultured in media containing 45% Click’s media (Irvine Scientific, Santa Ana, CA), 45% RPMI-1640, 10% fetal bovine serum (Hyclone), 2 mM L-glutamine (Gibco-BRL), and 100 U/mL recombinant human IL-2 (Teceleukin; Hoffmann-La Roche, Rockville, MD). Stimulated PBMCs were then transduced with a gamma retroviral vector encoding the CD19-CAR in plates coated with a recombinant fibronectin fragment (FN CH-296; Retronectin; Takara Bio, Otsu, Japan). After transduction, T cells were fed with IL-2 (50 U/mL) twice a week and then used for the experiments. For the in vivo trafficking experiments, CARTs were retrovirally transduced with a GFP/FFLuc gene 2 days after transduction of CD19-CAR. The first generation CD19-CAR used contains the FMC63 scFv sequence\(^1\) cloned in a SFG retroviral backbone in frame with the hinge region of the human IgG1 (CH2CH3 domain) and the ζ-chain of the human TCR/CD3 complex. Second and third generation CARs incorporate additional domains (CD28 alone or both CD28 and 4-1BB, respectively).

mRNA expression of \textit{AHR}, \textit{CYP1A1}, and \textit{CYP1B1}

Quantitative polymerase chain reaction (q-PCR) was performed to determine expression of \textit{AHR}, \textit{CYP1A1}, and \textit{CYP1B1}. Briefly, total RNA from each sample was extracted by QIAamp RNeasy plus mini kit (Qiagen), as per the manufacturer’s protocol. mRNA was transcribed into cDNA by iScript cDNA synthesis kit (Bio-rad) as per the manufacturer’s protocol. q-PCR was performed with iQ5 Real-time PCR Detection System (Bio-Rad), iTaq Universal SYBR Green
Supermix (Bio-rad), and specific primer sets. The real-time PCR reaction used one cycle of 95°C for 30 sec, followed by PCR amplification with 40 cycles of 95°C for 15 sec and 60°C for 1 min. The mRNA expression was also normalized to that of GAPDH. Primers sequences were (5'-3' forward, reverse): TTCAGCCACCATCCATACTTG, GGACTCGGCACAATAAAGCA (AHR); TTGGAGCTGGGTTTGACAC, GGCCTCCATATAGGGCAGAT (CYP1A1); TGGATTTGGAGAAACGTACCG, GGTCACCCATAAGGCAGA (CYP1B1); GCACCGTCAAGGCTGAGAAC, ATGGTGTTGAAGACGCCAGT (GAPDH).

**IDO expression in B-cell lymphoma samples**

Diffuse large B-cell lymphoma (DLBCL) samples were fixed in 10% formalin-acetone and embedded in paraffin. Slides were deparaffinized in xylene and rehydrated. Each slide was heated in 0.1 M citrate buffer (pH 6.0) using the Pascal heat induced target retrieval system (DAKO, Carpinteria, CA). Endogenous peroxidase activity was blocked by incubation with 0.3% H₂O₂ in methanol for 30 min at room temperature. The primary antibody was mouse anti-human IDO monoclonal antibody (MBL) and used at a dilution of 1:100. The slide was incubated with the antibody at 4°C overnight. After washing in 50 mM Tris-buffered saline, an avidine-biotin technique in which a biotinylated secondary antibody reacted with several peroxidase-conjugated streptavidin molecules was employed for amplification using a LSAB+/HRP kit (DAKO). Diaminobenzidine tetrahydrochloride was used to visualize immunoreactive cells. Nuclei were counterstained using Mayer’s hematoxylin.
Reference List

### Table 1. Characteristics of CLL patients

<table>
<thead>
<tr>
<th></th>
<th>Age</th>
<th>Sex</th>
<th>Rai stage</th>
<th>WBC (µL)</th>
<th>%CD19(+)CD5(+)</th>
<th>Previous treatment*</th>
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<td>CLL1</td>
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<td>F</td>
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<td>FC</td>
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<tr>
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<td>98.56</td>
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</tr>
<tr>
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<td>55</td>
<td>M</td>
<td>4</td>
<td>201,100</td>
<td>97.79</td>
<td>FCR</td>
</tr>
</tbody>
</table>

NA: not available, but the lymphocyte fraction in PBMCs was 84% for this patient.

F: fludarabine, C: cyclophosphamide, R: rituximab

*: Patients were not receiving any treatment when PBMCs were collected.
Figure S1. IDO-positive Raji clone (Raji-IDO) and IDO-negative Raji clone (Raji-control). (A) An IDO-positive Raji clone (Raji-IDO) was made by retroviral transduction with human IDO cDNA, with a clone transduced with an empty vector serving as control (Raji-control). IDO expression was confirmed by western blot analysis. (B) IDO enzyme activity was evaluated by measuring the concentrations of tryptophan and kynurenine in cell culture medium using HPLC. Raji-IDO produced kynurenine and completely used the tryptophan in the medium. When Raji-IDO was cultured with 1-MT (500 μM), tryptophan accumulated and the final kynurenine concentration decreased. Data represent mean ± SD from 3 independent experiments. (C) CD19 expression of Raji-control and Raji-IDO were assayed by flow cytometry. (D) Proliferation of Raji-control and Raji-IDO were assayed by trypan blue cell count. Data represent mean ± SD from 3 independent experiments.
Figure S2. Expression of costimulatory ligand receptors. Raji-control, Raji-IDO, Jeko-1, Daudi, and BJAB were evaluated for cell surface expression of CD80, CD86, CD83, and HLA-DR. CLL patients PBMCs were cultured with or without IFN-γ for 24 hrs before assessing expression of the same markers. There was no significant difference in the level of costimulatory receptor ligands on IDO-positive and negative cells.
Figure S3. Volume of drinking 1-MT water. (A) SCID/Beige female mice were given 1-MT (5 mg/mL) solution in foil-wrapped standard autoclaved drinking water bottles without sweetener. 1-MT was prepared in 0.1 N NaOH, which was then adjusted to pH 7.4 with HCl. Water bottles were checked daily for 15 days. Data represent the average of five mice in each group. (B) The body weight of each mouse was monitored. We observed no signs of dehydration in any of the mice resulting from lower 1-MT water intake.
**Figure S4. Cell viability of samples used to determine IDO expression.** (A) Jeko-1 cells were incubated with mafosfamide (2 μg/ml), fludarabine (20 μM), or both for 24 hours. Cells were then washed with media and incubated with or without IFN-γ (50 U/mL) for 24 hours. Before extracting proteins for western blot analysis, cells were washed twice with PBS and their viability assessed by trypan blue exclusion. Each bar represents the mean ± SD of the results of 3 independent experiments. (B) PBMCs of two CLL patients were incubated with mafosfamide (2 μg/ml), fludarabine (20 μM), or both for 24 hours. Cells were then washed with media and incubated with IFN-γ (50 U/mL) for 24 hours. Before extracting proteins for western blot analysis, cells were washed twice with PBS and their viability assessed by trypan blue exclusion.
Figure S5. Immunohistochemical staining for IDO expression in diffuse large B-cell lymphoma (DLBCL) samples. DLBCL biopsy samples obtained before treatment were stained with human IDO antibody. We found partially (A) and diffusely IDO-positive samples (B), as well as negative samples (not shown) (Ref. 18).