Supporting Information

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SI Experimental Procedures

RNA Isolation, cDNA Synthesis, and Real-Time Quantitative PCR. Total cellular RNA was isolated using the RNeasy Plus Mini kit (Qiagen Nordic) or Agilent Total RNA Isolation Mini kit (Agilent Technologies) according to the instructions of the manufacturer. One microgram of total RNA was reverse-transcribed using SuperScript VILO cDNA Synthesis kit (Invitrogen), according to the instructions of the manufacturer. The relative level of the different transcripts were determined with the LC FastStart DNA Master SYBR Green I kit in a LightCycler instrument (Roche), using the standard curve method, according to the instructions of the manufacturer, with the primers listed in Table S1. Target genes were measured and normalized to the endogenous control GAPDH.

Immunofluorescence Staining. The cells were deposited on glass slides in a cytospin centrifuge and fixed in aceton:methanol (2:1). For single staining, following rehydration in balanced salt solution (BSS), the slides were incubated with primary antibody at room temperature for 60 min. For EBNA2, mouse mAb PE2 (IgG1; Leica Biosystem NovoCastra; dilution 1:50) and for LMP1, S-12 (IgG2a; culture supernatant; dilution 1:50) were used. Following washing in BSS, fluorochrome-labeled anti-mouse antibody was added, and the slides were incubated for 30 min at room temperature. EBNA2 was visualized with goat anti-mouse IgG1-Alexa Fluor 488 (Invitrogen; dilution 1:200), and LMP1 was detected with goat anti-mouse IgG2a-Alexa Fluor 594 (Invitrogen; dilution 1:200). The slides were mounted with Vectashield containing DAPI (Vector Laboratories).

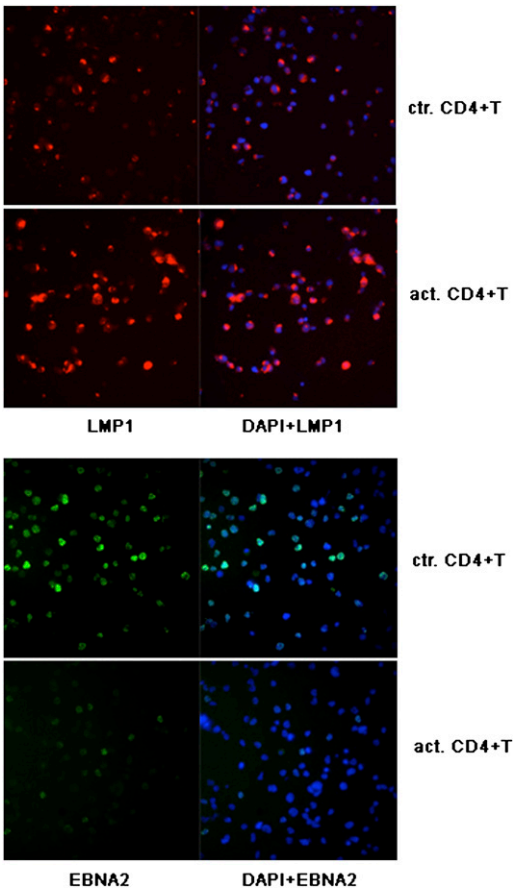

Fig. S1. Immunofluorescence staining of LCLs cultured in the TW system. LCLs were cultured in the insert with control or activated CD4 T cells for 5 d. Cells were stained for LMP1, using S12 mAb (*Upper*) and EBNA2, with PE2 mAb (*Lower*).

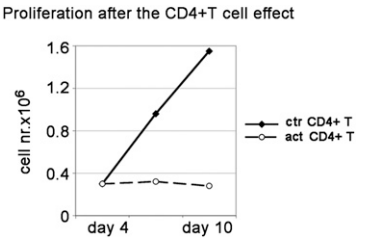

Fig. S2. Proliferation of LCLs exposed to factors produced by activated CD4$^+$ T cells. LCL07.08 was cultured in the TW system with anti-CD3/CD28 bead-activated CD4$^+$ T cells or control CD4$^+$ T cells. Cells recovered on day 3 were replated in fresh medium (day 0 for the proliferation experiment). On days 4, 7, and 10, the cells were counted.

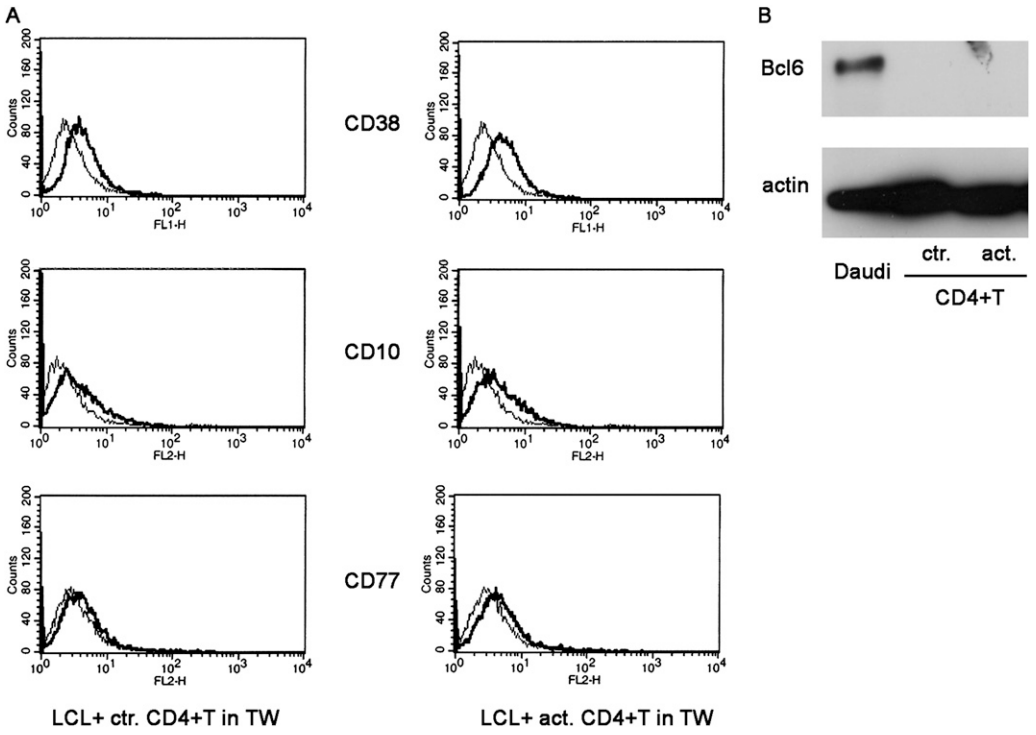

Fig. S3. Phenotypic characterization of LCLs exposed to factors produced by activated CD4$^+$ T cells. LCL07.08 was cultured in the TW system with anti-CD3/CD28 bead-activated CD4$^+$ T cells or control CD4$^+$ T cells. On day 5, LCLs were recovered and tested for CD10, CD38, and CD77 expression in flow cytometry. Bcl6 was also tested by immunoblot. Daudi is a Burkitt lymphoma line that served as positive control for Bcl6.

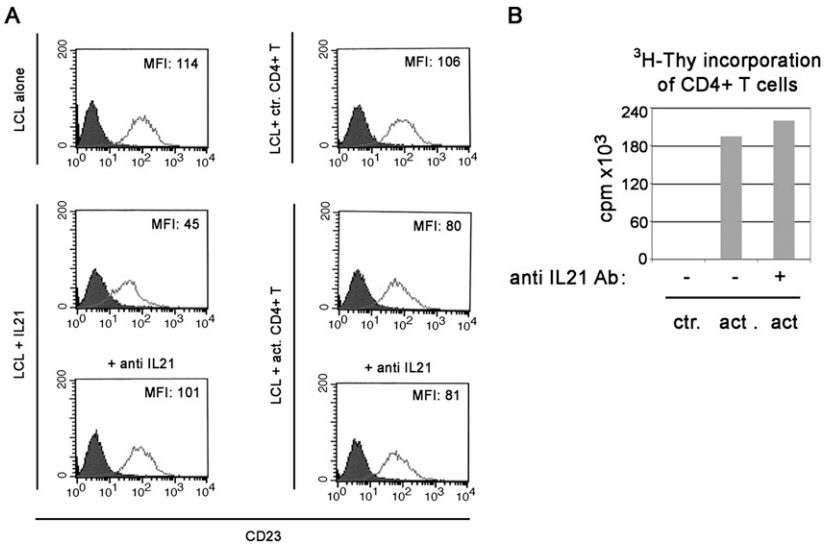

Fig. S4. Neutralization of IL21 in the activated CD4$^+$ T-cell supernatants does not inhibit down-regulation of CD23. LCL07.08 was treated with 50 ng/mL IL21 for 3 d. To parallel, IL21-treated cultures, 10 μg/mL anti-IL21 antibody was added. The same neutralizing anti-IL21 Ab was added to anti-CD3/CD28-activated CD4$^+$ T-cell cultures that were in TW with the LCL07.08. On day 3, cells were collected and tested for surface CD23 expression (*A*). The quality of activated CD4$^+$ T cells was assessed by their [^{3}H]Thy incorporation (*B*).

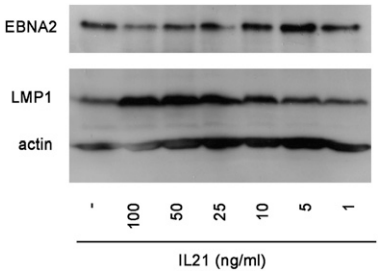

Fig. S5. Dose effect of IL21 treatment. LCL07.08 was treated for 5 d with different concentrations of IL21, after which, their EBNA2 and LMP1 protein levels were assessed.

Table S1. Primers used in quantitative PCR measurements

Primer pair	Sequence
LMP1	5′-gca gga ggg tga tca tca gt-3′
	5′-gtc ctc tat tcc ttt gct ctc atg-3′
LMP1-TR	5′-ctc tca agg tcg tgt tcc at-3′
	5′-gac gta gcc gcc cta cat aag-3′
EBNA2	5′-gga cac aag agc cat cac ct-3′
	5′-caa agc att cgc ata gca ga-3′
Cp	5′-gat cag atg gca tag aga caa gga c-3′
	5′-agg ctg ttt ctt cag tcg gtt tag-3′
Qp	5′-gat agc gtg cgc tac cgg at-3′
	5′-tgc aga atc agc tct ccc aaa c-3′
GAPDH	5′-gga agg tga agg tcg gag tca-3′
	5′-atg ggt gga atc ata ttg gaa ca-3′