Figure S1. AICAR induces apoptosis in CLL cells regardless of cytogenetic abnormalities
Cells from 28 patients (Table 1) without any genetic alteration (normal) or with 11q23 deletion (11q del), trisomy 12 (tris 12), 13q14 deletion (13q del) or 17p13 deletion (17p del) were incubated with AICAR for 24 hours. Viability was measured as described in “Patients, material, and methods” and it is expressed as the percentage of the viability of untreated cells. Black bars show the mean of cell death induced of each group.

Figure S2. AICAR-induced mRNA profile at 6 hours in CLL cells
Cells were untreated (≤) or treated (′) with 0.5 mM AICAR for 6 hours. Cells were lysed and the mRNA levels of BIM, BNIP3, BNIP3L, HRK, MOAP1, NOXA and PUMA were analyzed by RT-MLPA as described in “Patients, material, and methods”. Data represent mean ± SEM (n = 6). ***P < .005, **P < .01, *P < .05 AICAR-treated versus untreated cells.
Figure S3. Direct activation of AMPK in CLL cells does not upregulate apoptosis-related genes induced by AICAR
Cells were incubated without (Control) or with 0.5 mM AICAR or 100 µM A-769662 for 24 hours. The mRNA levels of BIM, BNIP3, BNIP3L, HRK, MOAP1, and NOXA were analyzed by RT-MLPA as described in “Patients, material, and methods”. Two representative samples are shown (n = 3).

Figure S4. Apoptosis-related gene expression induced by AICAR in patients sensitive to nutlin-3a and without 11q23 deletion
CLL cells (from patients 6, 8, 10, 11, 22 and 23) were untreated (≤) or treated (′) with 0.5 mM AICAR for 24 hours. Cells were lysed and the mRNA levels of BIM, BNIP3, BNIP3L, HRK, MOAP1, NOXA and PUMA were analyzed by RT-MLPA as described in “Patients, material, and methods”. Data represent mean ± SEM (n = 6). ***P< .005, **P< .01, *P< .05 AICAR-treated versus untreated cells.
Figure S5. Mouse T cells are more resistant than B cells to AICAR-induced apoptosis

B (•) and T (○) lymphocytes from wild-type mouse spleen were incubated with AICAR in a range of concentrations (0.05, 0.1, 0.25 and 0.5 mM) for 12 hours (solid lines) and 24 hours (dotted lines). Viability was measured as described in “Patients, material, and methods” and it is expressed as the percentage of the viability of untreated cells at each time-point. Data represent mean ± SEM (n = 6).

Figure S6. Z-VAD.fmk protects from AICAR-induced apoptosis in mouse B lymphocytes

B cells were preincubated without (−) or with 200 μM Z-VAD.fmk for 30 minutes and 0.5 mM AICAR (+) was added for 24 hours. Viability was measured as described in “Patients, material, and methods” and it is expressed as the percentage of the viability of untreated cells. Data represent mean ± SEM (n = 3). ***P< .005 AICAR-treated versus untreated, or ZVAD+AICAR versus AICAR-treated cells.