**Fig S1. Apoptotic change of KOPT-K1 cells after NDI-031301 treatment.** KOPT-K1 cells were cultured in the absence or presence of NDI-031301 (1 µM or 3 µM) for 24 h, and assessed for apoptosis by flow cytometric analysis following Annexin V-FITC and PI double staining. The panels show two-dimensional dot plot images with the percentage of cells in each colored fraction. Blue rectangles indicate the fractions of Annexin V-positive / PI-negative early apoptotic cells, while red rectangles indicate the fractions of Annexin V-positive / PI-positive late apoptotic cells.
Fig S2. Combination effect of NDI-031301 and MAPK pathway inhibitors on viability of KOPT-K1 cells. KOPT-K1 cells were treated for 72 h with varying combinations of NDI-031301 (0 - 3 µM) and each of MEK1/2 inhibitor trametinib (0 – 2.5 nM) or SAPK/JNK inhibitor SP600125 (0 - 5 µM). The relative cell viability after each combinational treatment is shown in panels (A) and (C), as mean ± s.d. percentages of the DMSO treated control value in triplicate experiment. The normalized isobolograms were obtained with the Calcusyn software. Each of the combination indices was plotted in panels (B) and (D), as a function of dose combination, and an average combination index (CI) of each drug combination is indicated in the panel.
Fig S3. NDI-031301 sensitivity and the effect of the drug treatment on cellular signaling pathways in additional human T-ALL cell lines. (A) Anti-proliferative activity of NDI-031301 on additional human T-ALL cell lines. SUP-T13, KOPT-K1, RPMI-8402 and JURKAT cells were cultured with graded concentrations of the indicated inhibitor for 72 h. Cell viability values are mean ± s.d. percentages of the untreated control value in triplicate experiments. IC$_{50}$ value in each cell line is shown. (B-D) Time-course analysis to assess the effect of NDI-031301 on cellular signaling pathways was conducted in each of SUP-T13 (B), RPMI-8402 (C) and JURKAT (D) cells, respectively. The cells were treated with DMSO or 3 µM of NDI-031301, and harvested at the indicated times. Western blot analysis was performed with each specified antibody.
Fig S4. Trends of weight of xenografted mice treated with NDI-031301. Weights of mice treated with vehicle or NDI-031301 at 100mg/kg twice daily in this study are displayed (n = 6 for each treatment group).
Fig S5. NDI-031301 reduces the appearance of human CD45 and CD2-double positive cells in peripheral blood of KOPT-K1 xenografted mice.  (A) Appearance of human leukemia cells in peripheral blood was assessed by flow cytometric analysis following human CD45 (hCD45)-PE and human CD2 (hCD2)-APC double staining. The panels show two dimensional dot plots image on a representative mouse treated with vehicle or NDI-031301 at 100mg/kg for 29 days. Red rectangles indicate the fractions of hCD45 and hCD2-double positive cells. (B) The percentage of hCD45 and hCD2-double positive cells in peripheral blood of each treated mouse (n = 4 for each treatment group). Error bars represent SEM. The statistical significance was assessed by two-sample, two-tailed t test.