Supplementary Materials

Chemotherapeutic Activity of Pitavastatin in Vincristine Resistant B-Cell Acute Lymphoblastic Leukemia

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Supplemental figures

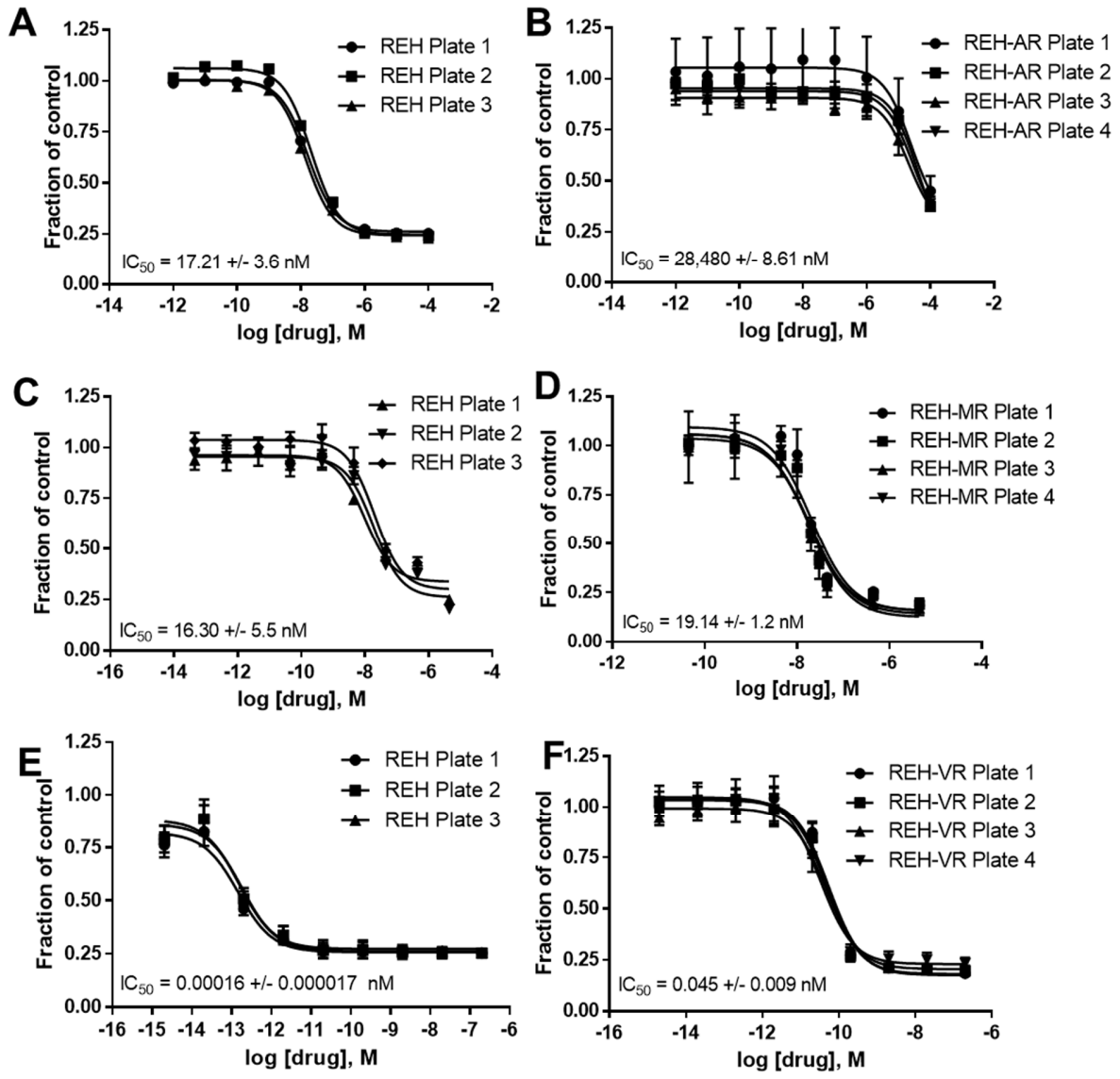

Figure S1. Dose-response effect of chemotherapeutics on cell proliferation, using CCK-8 (WST-8) reagent. Ara-C treatment in (A) REH and (B) REH-AR resistant cells; MTX treatment in C) REH and D) REH-MR resistant cells; and vincristine treatment in (E) REH and (F) REH-VR resistant cells. The IC_{50} for only Ara-C and vincristine was significantly different ($p < 0.05$; Student-t test), but not for MTX. Each data point is avg $\pm$ S.D., with N = 4. To show reproducibly of the assay, we repeated the experiments 3-4 times.

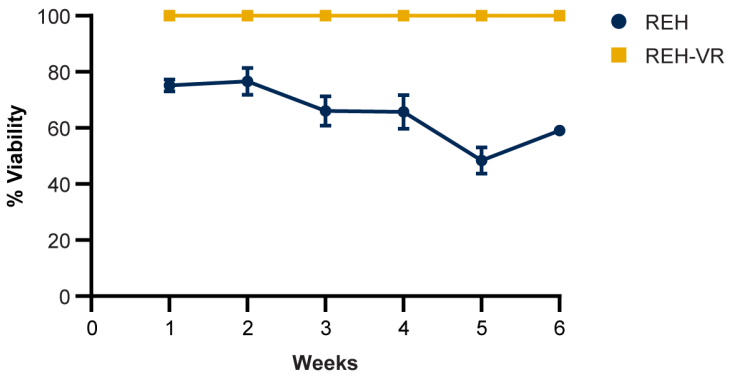

Figure S2. Stability of vincristine resistance. The vincristine resistance remained stable throughout the experimental design. Vincristine treatment was 2 nM. Percent viability determined using Trypan Blue exclusion method. N = 4. Data represents average ± S.D.

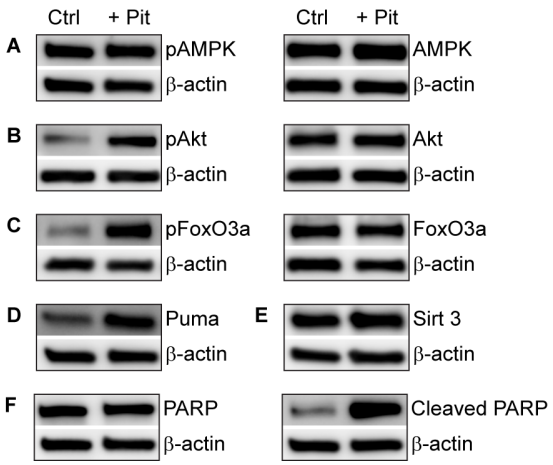

Figure S3. Pitavastatin effects on metabolic regulators. REH cells were treated with 450 nM pitavastatin (+ Pit) or DMSO vehicle control (Ctrl). Western blot analysis shows changes in phosphorylated and total protein levels following 48 hours treatment. We observed an increase in phospho-AKT, PARP cleavage, phospho-FoxO3a (Ser413), and Puma with pitavastatin treatment.

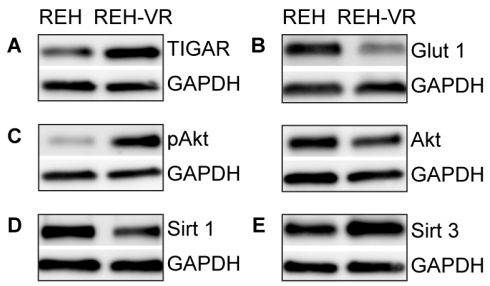

Figure S4. Differential expression of proteins in the vincristine-resistant cells. Western blot analysis of proteins in REH and REH-VR cells.

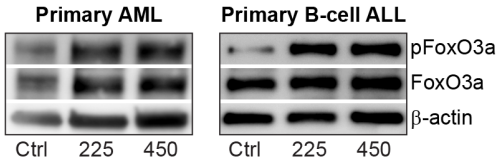

Figure S5. Pitavastatin treatment of primary leukemia samples. Primary human samples from AML and B-cell ALL patients were treated with pitavastatin at 225 nM and 450 nM and the protein expression levels of phospho-FoxO3a determined by Western Blot analysis**.** Control treatment (Ctrl) was DMSO vehicle**.**