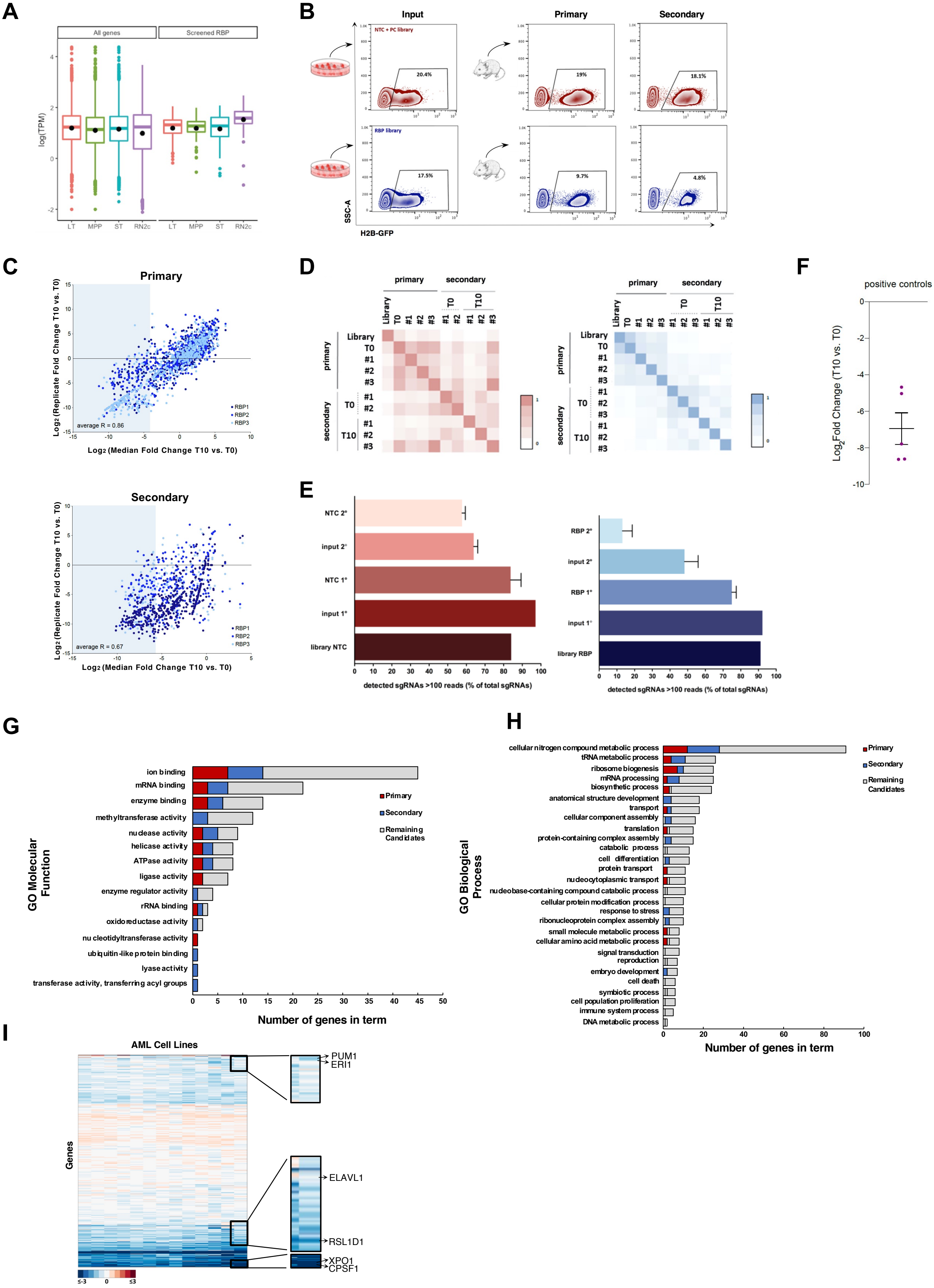
Supplemental Figure S1



Supplemental Figure S1. Two-step *in vivo* CRISPR-Cas9 screening of LSC-associated RBPs identifies drivers of AML LSC function. (A) Gene expression for all genes in comparison to the screened RBPs in control RN2c cells vs purified mouse bone marrow stem and progenitor cell fractions from[112]. (B) H2B-GFP$^+$ fractions of input RN2c cultures 48hr post-transduction (T0) and representative Ly5.2$^+$ endpoint grafts (T10) from the primary and secondary transplants. (C) sgRNA dropout reproducibility across duplicate RBP-targeting primary (top) and secondary (bottom) arms of the screen; scatter plot illustrates the correlation of normalized reads per sgRNA in three independent replicates. Blue shaded area represents sgRNAs depleting >20 fold (on average). R = pearson correlation coefficient. (D) Pearson correlation coefficients for the normalized sgRNA read counts of the plasmid library in cells 48hr after lentiviral infection (T0) and BM samples 10 days post-transplant (T10) for both primary and secondary rounds of the NTC (left) and RBP (right) arms of the screen. T0 in the secondary arm indicates the fraction of primary transplant endpoint samples that was retained and library representation at the endpoint of secondary grafts (secondary T10) was compared to this input. (E) Percentage of all sgRNAs at each sampling point in the NTC (left) and RBP (right) arms of the screen detected at a read count great than 100. (F) Log2 fold change (LFC) of positive controls targeting sgRNAs in the primary arm of the dropout screen. (G) GO annotation plots illustrating associated Biological Processes (left) and Molecular Functions (right) for hits and RBP screen candidates. (H) CRISPR score (average log2 fold-change of sgRNAs after 14 *in vitro* population doublings) of >18.000 genes (rows) in 13 human AML cell lines [40] (columns). A low CRISPR score corresponds to a high degree of essentiality. Arrows indicate CRISPR scores of select primary and secondary RN2c screen hits. (I) CRISPR score (average log2 fold-change of sgRNAs after 14 *in vitro* population doublings) of >18.000 genes (rows) in 13 human AML cell lines (Wang *et al.*, 2017) (columns). A low CRISPR score corresponds to a high degree of essentiality. Arrows indicate CRISPR scores of key primary and secondary RN2c screen hits.