## SUPPLEMENTAL MATERIAL

Quek et al., http://www.jem.org/cgi/content/full/jem.20151775/DC1

JEM S25

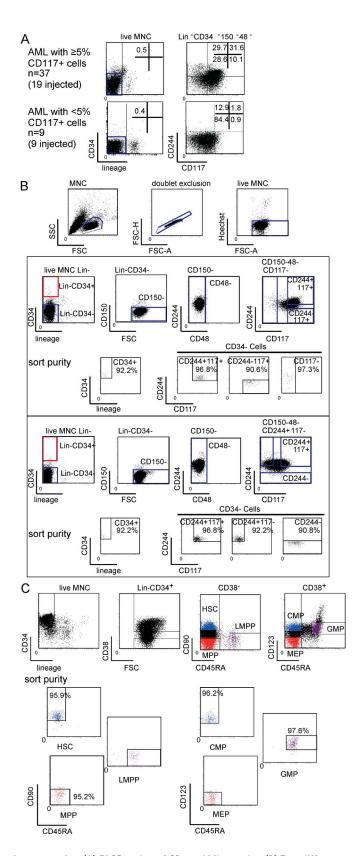


Figure S1. **FACS** analysis and sorting strategies. (A) FACS gating of CD34<sup>-</sup> AML samples. (B) Two different sort strategies of CD34<sup>-</sup> AML samples. (C) Sort strategy of normal CD34<sup>+</sup> stem/progenitor populations. FSC, forward scatter; SSC, side scatter; MEP, megakaryocyte-erythroid progenitor; CMP, common myeloid progenitor

Table S1 is provided as an Excel file and shows data on: (A) immunophenotype and (B) karyotypic and genetic analysis of CD34<sup>-</sup> AML samples. Recurrent nucleotide variants detected by targeted resequencing and/or whole-exome sequencing are filtered according to the parameters set out in Materials and methods.

Table S2 is provided as an Excel file and shows FACS data of xenotransplantation assays: (A) serial AML engraftment of unsorted and sorted CD34<sup>+</sup> and CD34<sup>-</sup> populations in CD34<sup>-</sup> AML, (B and C) sort purities of AML populations used in xenotransplantation, (D) estimation of LSC frequency of engrafting subpopulations, and (E) immunophenotype of engrafted leukemia cells compared with the primary patient AML sample.

Table S3 is provided as an Excel file and shows sort purities of normal BM MNC fractions used for in vitro culture assays.

Table S4 is provided as an Excel file and relates to RNA-seq experiments: (A) the sort purities of normal and AML samples, (B) ranked loadings scores in the first two PCs of 500 genes selected by ANOVA analysis, and (C) 547 TF genes ranked by loadings scores in PC1 and PC2.

Table S5 is provided as an Excel file and lists 300 significantly differentially expressed genes between CD34<sup>-</sup> LSC and non-LSC populations analyzed by rank product analysis (PFP <0.05).

Table S6 is provided as an Excel file and contains data from bulk and single-cell genotyping. VAF of recurrent, AML-associated mutations in samples: (A) #880, (B) #1037, (C) #044, (D) #059, (E) #875, and (F) #449. Table S6 G shows the frequencies of sorted populations for single-cell genotyping (as percentage of Lin<sup>-</sup> MNCs) and their sort purities.

JEM S27