S1. Characterization of mPML-PRflox mice. A. Spleen cells were assessed by Western blot for Pml and α-Actin expression. There was decreased Pml in heterozygous mPML-PRflox+/− mice and a loss of Pml in homozygous mPML-PRflox+/+ mice consistent with mPML-PRflox causing a Pml null allele (blot was overexposed to adequately measure the null allele in mPML-PR+/+ mice). B. RT-PCR amplification of PML-RARA and Pml in indicated bone marrow cells. C. RT-PCR amplification PML-RARA and Pml from indicated spleen cells. The LysM-Cre+/− x mPML-PRflox+/+ tumor indicates mouse 6317 (see Figure 5B). D. Western blot for PML-RARA from indicated leukemic spleen samples (see Figure 5B and 6E). E. Pml immunofluorescence in indicated bone marrow neutrophils.
S2. Q-PCR quantitation of mPML-fPR. A. Titration of mPML-PRflox with mPML-fPR. Spleen DNA from mPML-PRflox mice was titrated with spleen DNA from secondary leukemic transplants (Figure 5E). Q-PCR amplification of DNA with Flox primers (which amplify across the PGK-neo cassette) and Neo primers (which amplify across the first LoxP site) were normalized to PR primers (which amplify across the PML-RARA junction) (see Figure 2A for location of primers relative to transgene).

B. Amplification of control DNA mixtures at 10% and 90% mPML-fPR during subsequent studies. Each data point represents results from a separate experiment.

C. ERT2-Cre activation is dose-dependent following 4 mg IP tamoxifen. ERT2-Cre+/- x mPML-PRflox+/- mice were treated with indicated doses of IP tamoxifen, and bone marrow DNA harvested either 3 days (circles) or 14 days (squares) following the last dose. Each data point represents results from an individual mouse.
S3. Methylcellulose assessment of colony forming units (CFUs) in bone marrow (A) and spleen (B) from indicated mice treated with 5 doses of tamoxifen (6 mg IP every other day). Cells were harvested 24 hours after the final dose and plated in methylcellulose with IL-3, IL6, and SCF. C. Colony forming units were assessed in bone marrow and spleen cells from A & B in methylcellulose containing G-CSF (10 ng/ml). Data represent average and standard deviation from three individual mice in each genotype.
S4. Hematopoietic assessment of mice treated with tamoxifen and then followed with 18 months of observation. Mice in Figure 6A were evaluated at 18 months with complete blood counts (A) and flow immunophenotype of peripheral blood (B), bone marrow (C) and spleen (D) cells. Data represent results from individual mice.
S5. Competitive repopulation. Donor (CD45.2) and competitor (CD45.1) mice were treated with 10 doses of IP tamoxifen (4 mg) and bone marrow cells transplanted at indicated ratios into lethally irradiated CD45.1 hosts. Grey bars indicate ratios of CD45.2/CD45.1 among transplanted cells. Black bars indicated ratios of engrafted cells in host mice. A. ERT2-Cre+/− x mPML-PRflox+/− donor. B. ERT2-Cre+/− donor. C. mPML-PRflox+/− donor. Data represent average and standard deviation of indicated numbers of mice in each cohort.
S6. Competitive repopulation within hematopoietic compartments. Donor (CD45.2) and competitor (CD45.1) bone marrow cells were transplanted into lethally irradiated recipients (CD45.1). Following engraftment, all mice received 5 doses of 3 mg tamoxifen by gavage as indicated (tam). At indicated time post-transplant, the percentage of donor cells in peripheral blood was analyzed (Figure 7). Peripheral blood was also stained as indicated and the percentage of donor cells assessed within each compartment: myeloid: Gr1+, lymphoid: CD19+, CD3+, and Nk1.1+. Grey line indicates the percentage of total donor cells at 6 weeks.

A.-D. Compartment analysis of mice transplanted with donor ERT2-Cre+/- x mPML-PRflox+/- bone marrow (Figure 7A). E.-H. Compartment analysis of mice transplanted with donor ERT2-Cre+/- bone marrow (Figure 7B). Data indicated average and standard deviation.

Ave total donor cells in PB @ 6 wks n = 7

Ave total donor cells in PB @ 6 wks n = 3