Supplemental Figure 1. CD105 is expressed in normal CD34+ precursor cells.
(A-B) CD105 expression in normal BM. A) Representative FACS plots show CD105 expression in BM precursor cells, which were gated based on side scatter (SSC; y axis) and expression of CD45 (x axis), and then CD34. Percentage represents CD105 expression. B) Table summarizes CD105 expression in CD34+ progenitor cells in a total of 6 normal BM samples.
(C-D) Representative FACS profile shows CD105 expression in CD34+ progenitor cells, sub-fractionated based on CD38 expression, in BM (C) and UCB (D). Percentage represents CD105 expression in CD34+CD38- and CD34+CD38+ sub-populations.

Supplemental Figure 2. Detailed flow cytometry gating of the AML and ALL blast population.
(A-B) Representative FACS plots show characterization of the blast population for AML (A) and ALL (B). The blast population was gated based on SSC and low/intermediate expression of CD45, and then analyzed for the expression of CD117 (A) or CD19 (B), CD34, and CD105. Percentages represent the fraction of cells that express a given surface antigen.

Supplemental Figure 3. Leukemia development in a xenograft model.
(A) Representative FACS plots show leukemia development in the BM of a mouse that had been injected with primary AML blasts (A0032). Engrafted human cells were identified by staining for human CD45 and then further characterized by staining for CD105, CD117 and CD34.
(B-C) Blood analysis. FACS plot shows the presence of hCD45\(^+\) cells in the PB of a mouse that had been injected with primary A0032 AML blasts (B). Image shows representative blood smear stained with H&E, confirming the presence of immature cells (C).

(D) Spleen characterization. FACS plot shows the presence of hCD45\(^+\) cells in the spleen of a mouse that had been injected with primary A0032 AML blasts.

(E) Images show representative morphology of spleens from mice that had been injected with AML (upper) and ALL (lower) blasts, comparatively to WT spleen (middle).

(F-G) Blood analysis. FACS plot shows the presence of hCD45\(^+\) cells in the PB of a mouse that had been injected with primary P0028 ALL blasts (F). Image shows representative blood smear stained with H&E, confirming the presence of immature cells (G).

(H) FACS plot shows the presence of hCD45\(^+\) cells in the spleen of a mouse that had been injected with primary P0028 ALL blasts.

(I) Representative FACS plots show leukemia development in the BM of a mouse that had been injected with primary ALL blasts (P0028). Engrafted human cells were identified by staining for human CD45 and then further characterized by staining for CD105, CD19 and CD34.

**Supplemental Figure 4. Characterization of CD105\(^+\) and CD105\(^-\) AML and ALL blast sub-fractions.**

(A-B) Representative FACS plots show the purity of CD105\(^+\) (red gate) and CD105\(^-\) (blue gate) sub-fractions sorted from primary AML (A) and ALL (B) blasts. Percentages represent CD105 expression.
(C-D) Representative FACS plots show characterization of CD105+ (red gate) and CD105- blast sub-fractions for AML (C) and ALL (D). Blast population was gated based on SSC and low/intermediate expression of CD45, and then sub-fractioned based on CD105 expression. CD105+ (red) and CD105- (blue) sub-fractions were confirmed to express CD34 and, CD117 in the case of AML (C) or CD19 in the case of ALL (D).

Supplemental Figure 5. Detection of human CD45 following the injection of CD105+ and CD105- AML and ALL blast sub-fractions.

(A-B) Representative FACS plots shows the presence of human CD45+ cells at 4, 8, and 12 weeks following the injection with AML CD105 sub-fractions (A). Expression levels of hCD105 in the hCD45+ population, gated at week 12 for both CD105+ and CD105- cohorts (B).

(C) Representative FACS plots shows the presence of human CD45+ cells at 4, 8, and 12 weeks following the injection with ALL CD105 sub-fractions. Expression levels of hCD105 in the hCD45+ population (D).

Supplemental Figure 6. Additional data on the effect of TRC105 on the ability of AML/ALL blasts to generate leukemia in a xenograft model.

(A-D) Effect of TRC105 treatment on AML development. A) Percentage of human CD45+ cells in sub-groups was plotted individually, each data point represents a single mouse from TRC105- or IgG isotype-injected cohort. B) Bars represent average percentage of human CD45 in spleen for each cohort. Error bars indicate SEM. Spleen of TRC105-treated mice exhibited significantly lower levels of human CD45+ cells than IgG-injected counterparts. C) Spleen weight of TRC105-injected mice was lower than in IgG-injected controls. ***p<0.001 and **p<0.01 by Student’s t test. (D) Weight loss was
prevented by TRC105 treatment. Each data point represents a single mouse for TRC105 and IgG isotype groups. Measurements were performed at the beginning on day 0, and by the end of the study at week 12. ***p<0.001 for IgG Isotype control group by paired Student’s t test.

(E-F) Effect of TRC105 treatment on ALL development. E) Percentage of human CD45$^+$ cells in sub-groups was plotted individually, each data point represents a single mouse from TRC105- or IgG isotype-injected cohort. F) Splenomegaly was observed in both TRC105- and IgG isotype-treated cohorts.

**Supplemental Figure 7. Endoglin expression in human CD45$^+$ cells of leukemic mice treated with TRC105 or IgG isotype control.**

(A-B) Representative FACS plots shows the expression levels of human CD45 and human CD105 at 12 weeks following the injection with AML (A) or ALL (B) blasts. At this time point, no significant differences were observed in % of hCD45 on BM. Nearly all CD45$^+$ blasts were positive for CD105 in all experimental groups.

**Supplemental Figure 8. Additional data on the effect of TRC105 in leukemia progression.**

(A-D) Effect of TRC105 treatment on AML development. A) Percentage of human CD45$^+$ cells in sub-groups was plotted individually, each data point represents a single mouse from TRC105- or IgG isotype-injected cohort. B) Bars represent average percentage of human CD45 in BM and spleen for each cohort. Error bars indicate SEM. Spleen of TRC105-treated mice exhibited significantly lower levels of human CD45$^+$ cells than IgG-injected counterparts, however, no difference was observed in BM. C) Spleen weight of TRC105-injected mice was lower than in IgG-injected controls.
***p<0.001, *p<0.05 by Student’s t test. D) Weight loss was prevented by TRC105 treatment. Each data point represents a single mouse for TRC105 and IgG isotype groups. Measurements were performed at the beginning on day 0, and by the end of the study at week 12. ***p<0.001 for IgG Isotype control group by paired Student’s t test.

(E-F) Effect of TRC105 treatment on ALL development. E) Bars represent average percentage of human CD45 in BM and spleen for each cohort. Error bars indicate SEM. F) No difference was observed on weight loss between TRC105 (red) and IgG isotype (grey) groups. Each data point represents a single. Measurements were performed at the beginning on day 0, and by the end of the study at week 8.

**Supplemental Figure 9.** TRC105, in combination with mild myeloablation, suppresses the *in vivo* progression of AML.

(A-B) Effect of TRC105+AraC treatment on AML progression. A) Representative FACS plots show levels of human CD45 in the PB before (4th week) and after (6th and 10th week) treatment with AraC or TRC105+AraC. B) Bars represent average percentage of human CD45 in PB for each group. Red arrow indicates the beginning of treatment. Leukemia progression was significantly suppressed in the TRC105+AraC-injected cohort.* p<0.05, *** p<0.001 and **** p<0.0001 by ANOVA.

**Supplemental Figure 10.** Additional data on the effect of TRC105 in combination with mild myeloablation in the *in vivo* progression of ALL.

Effect of TRC105+CPA treatment on ALL progression. Representative FACS plots show levels of human CD45 in the PB before (4th week) and after (8th and 11th week) treatment with TRC105, CPA or TRC105+CPA.
Supplemental Figure 1

A

B

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C

D
Supplemental Figure 3

**AML engrafted mouse**

**Bone Marrow**

**Blood**

**Spleen**

**ALL engrafted mouse**

**Blood**

**Spleen**

**Bone Marrow**
Supplemental Figure 6

AML – Treatment from day 2

A

% hCD45 in PB

D0 4th week 8th week 12th week

B

% hCD45 in Spleen

20 40 60 80 100

IgG TRC105

0.0 0.05 0.10 0.15 0.20

C

Spleen weight (g)

0.0 0.05 0.10 0.15 0.20

IgG TRC105

D

Body weight (g)

10 15 20 25

Day 0 12th week

ALL – Treatment from day 2

E

% hCD45 in PB

0 50 100 150

Day 0 4th week 8th week

F

Spleen weight (g)

0.0 0.1 0.2 0.3 0.4

IgG TRC105
Supplemental Figure 7

A

AML

Untreated

91.7%

95.3%

IgG Isotype

99.3%

88.6%

TRC105

99.5%

89.3%

hCD45

hCD105

B

ALL

Untreated

99.7%

90.8%

IgG Isotype

96.7%

85.1%

TRC105

98.5%

89.1%

hCD45

hCD105
Supplemental Figure 8

AML – Treatment from day 30

A. % hCD45 in PB

B. % hCD45 in Spleen and Bone Marrow

C. Spleen weight (g)

D. Body weight (g)

ALL – Treatment from day 30

E. % hCD45 in Bone Marrow and Spleen

F. Body weight (g)
Supplemental Figure 9

A

4th week 6th week 10th week

Untreated

42.3% 60.7% 93%

AraC

33.6% 39% 69.5%

AraC + TRC105

35.7% 16.2% 19.1%

hCD45

B

Untreated AraC AraC + TRC105

% hCD45 in PB

4th week 6th week 10th week

0 20 40 60 80 100
Untreated

TRC105

CPA

TRC105+CPA

4th week

8th week

11th week

4.0%

86.3%

98.4%

3.8%

93.7%

97.0%

5.3%

13.1%

4.0%

4.2%

5.1%

3.5%

hCD45