Figure S1

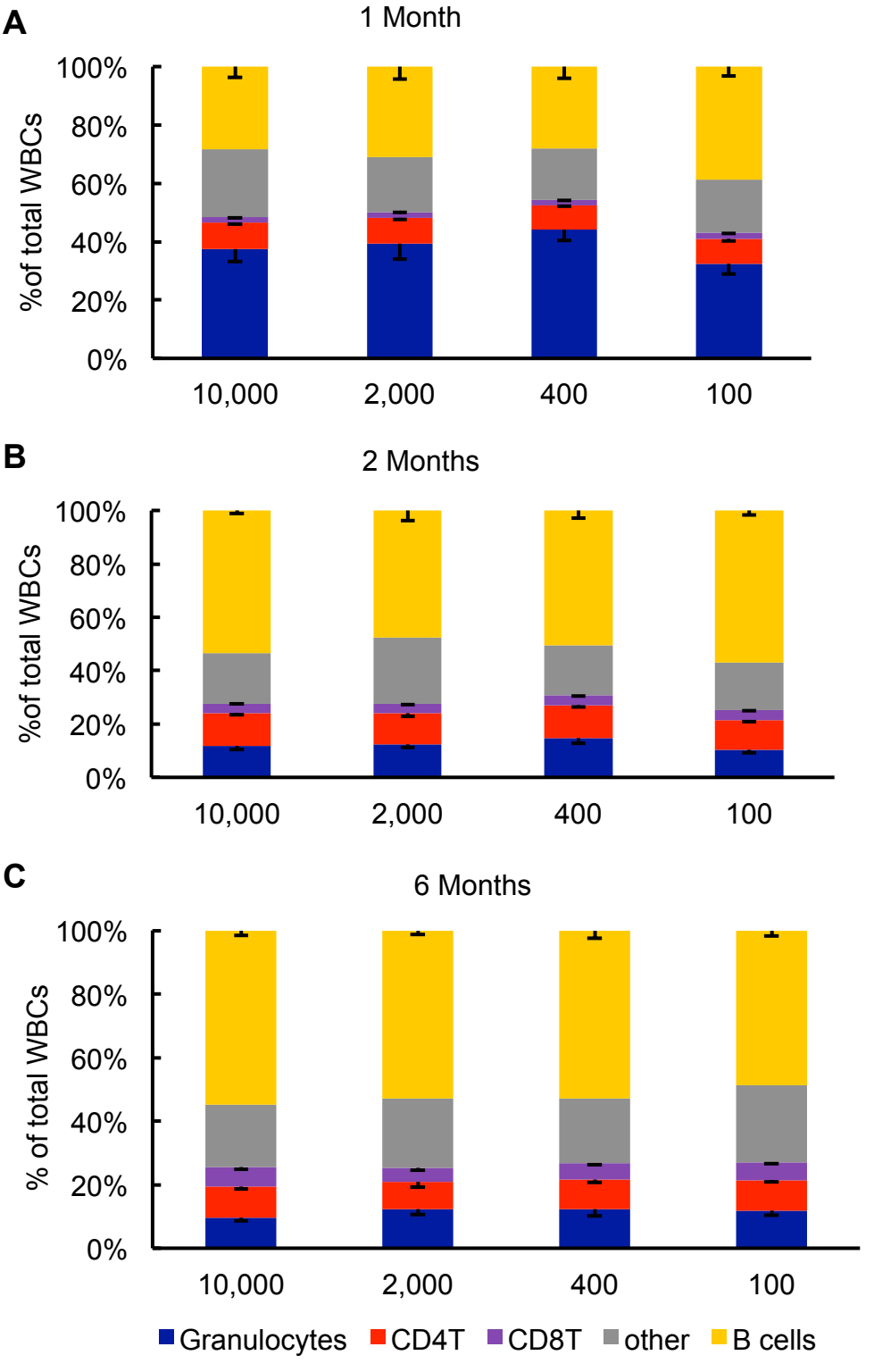
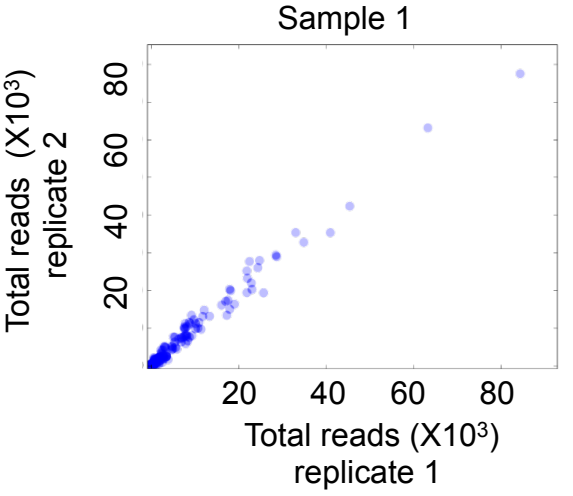
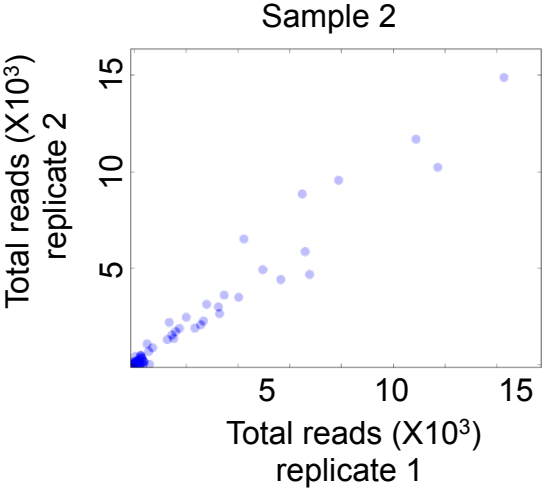

A
1 Month
%of total WBCs
100%
80%
60%
40%
20%
0%
10,000
2,000
400
100
B
2 Months
%of total WBCs
100%
80%
60%
40%
20%
0%
10,000
2,000
400
100
C
6 Months
% of total WBCs
100%
80%
60%
40%
20%
0%
10,000
2,000
400
100
Granulocytes
CD4T
CD8T
other
B cells

Figure S2


Sample 1
Total reads (X10³) replicate 2
20
40
60
80
Total reads (X10³) replicate 1


Sample 2
Total reads (X10³) replicate 2
5
10
15
Total reads (X10³) replicate 1

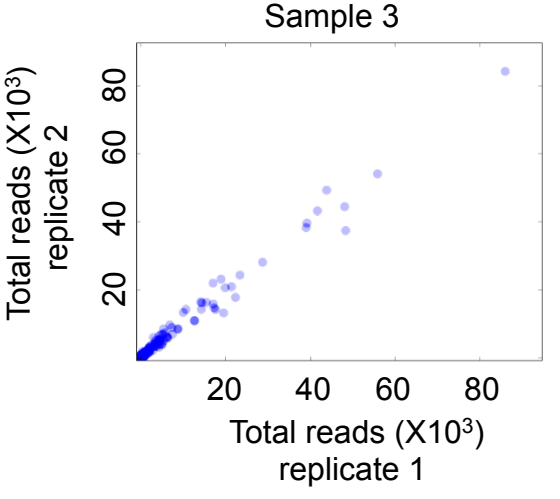
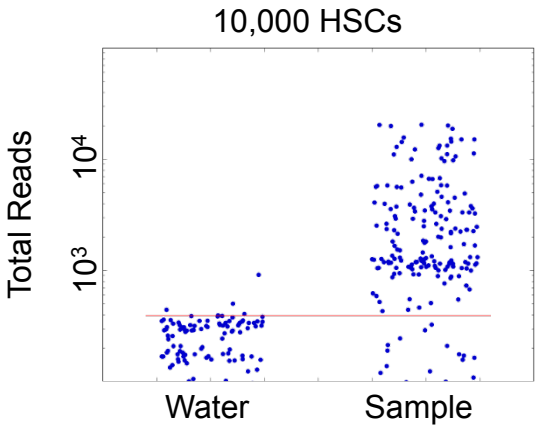
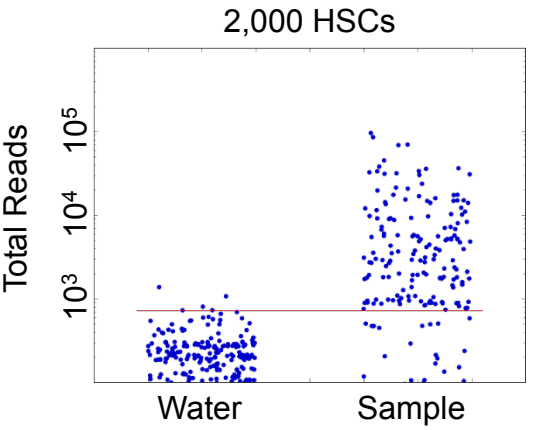
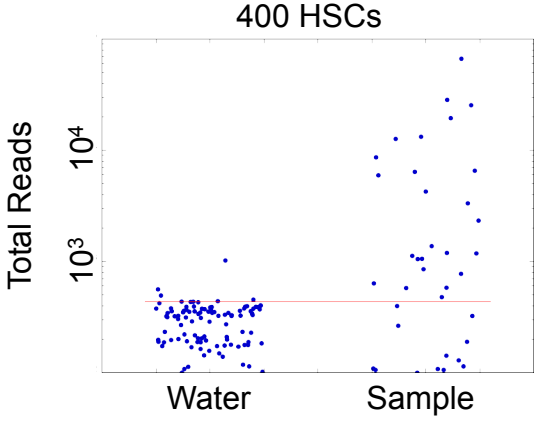

Sample 3
Total reads (X10³) replicate 2
20
40
60
80
Total reads (X10³) replicate 1

Figure S3


10,000 HSCs
Total Reads
10^3
10^4
Water
Sample


2,000 HSCs
Total Reads
10^3
10^4
10^5
Water
Sample


400 HSCs
Total Reads
10^3
10^4
Water
Sample

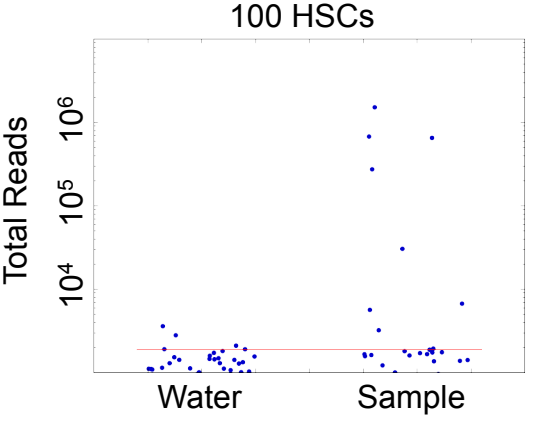

100 HSCs
Total Reads
10^4
10^5
10^6
Water
Sample

Figure S4

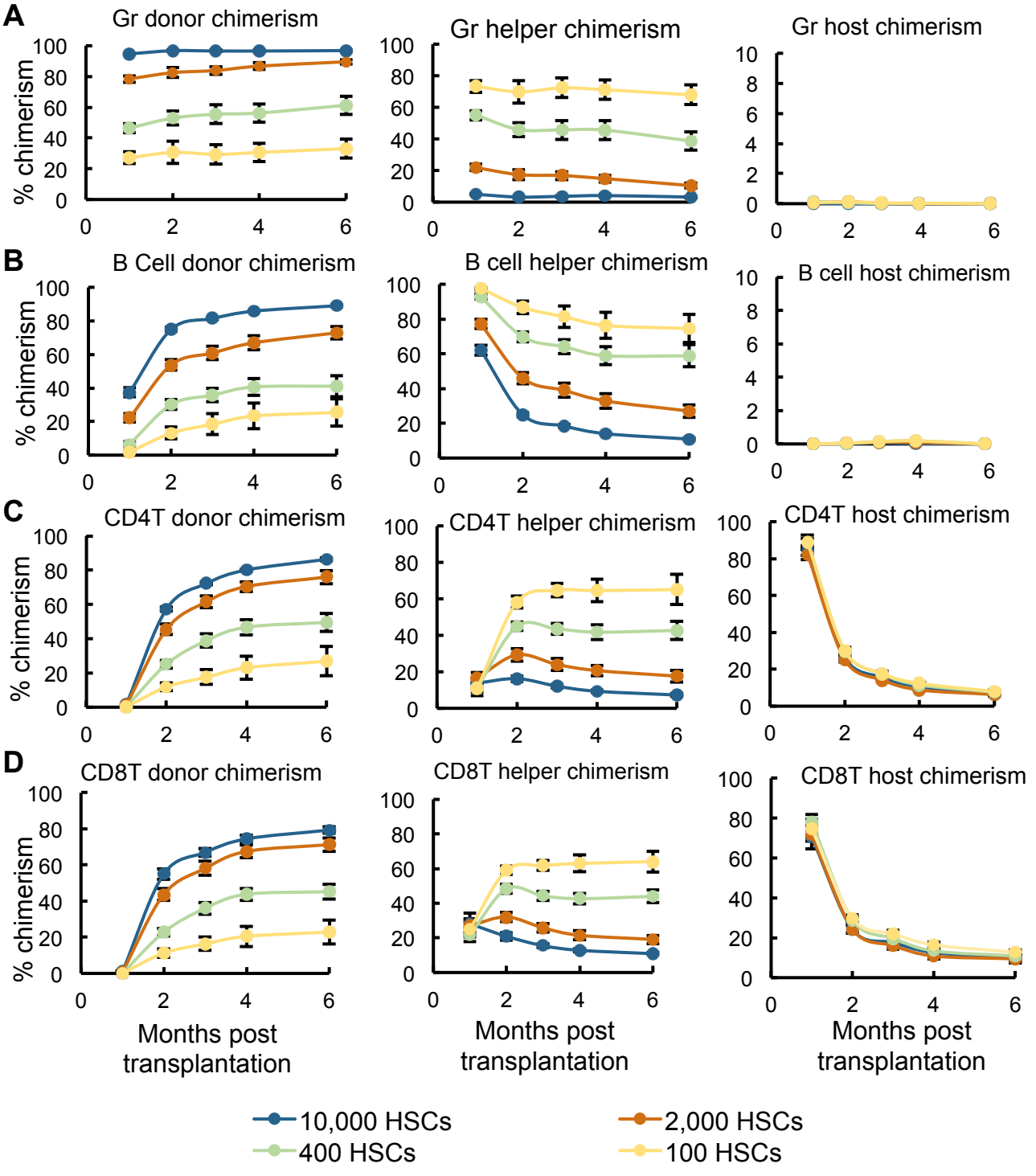

A
Gr donor chimerism
Gr helper chimerism
Gr host chimerism
B
B Cell donor chimerism
B cell helper chimerism
B cell host chimerism
C
CD4T donor chimerism
CD4T helper chimerism
CD4T host chimerism
D
CD8T donor chimerism
CD8T helper chimerism
CD8T host chimerism
% chimerism
Months post transplantation
10,000 HSCs
2,000 HSCs
400 HSCs
100 HSCs

Figure S5

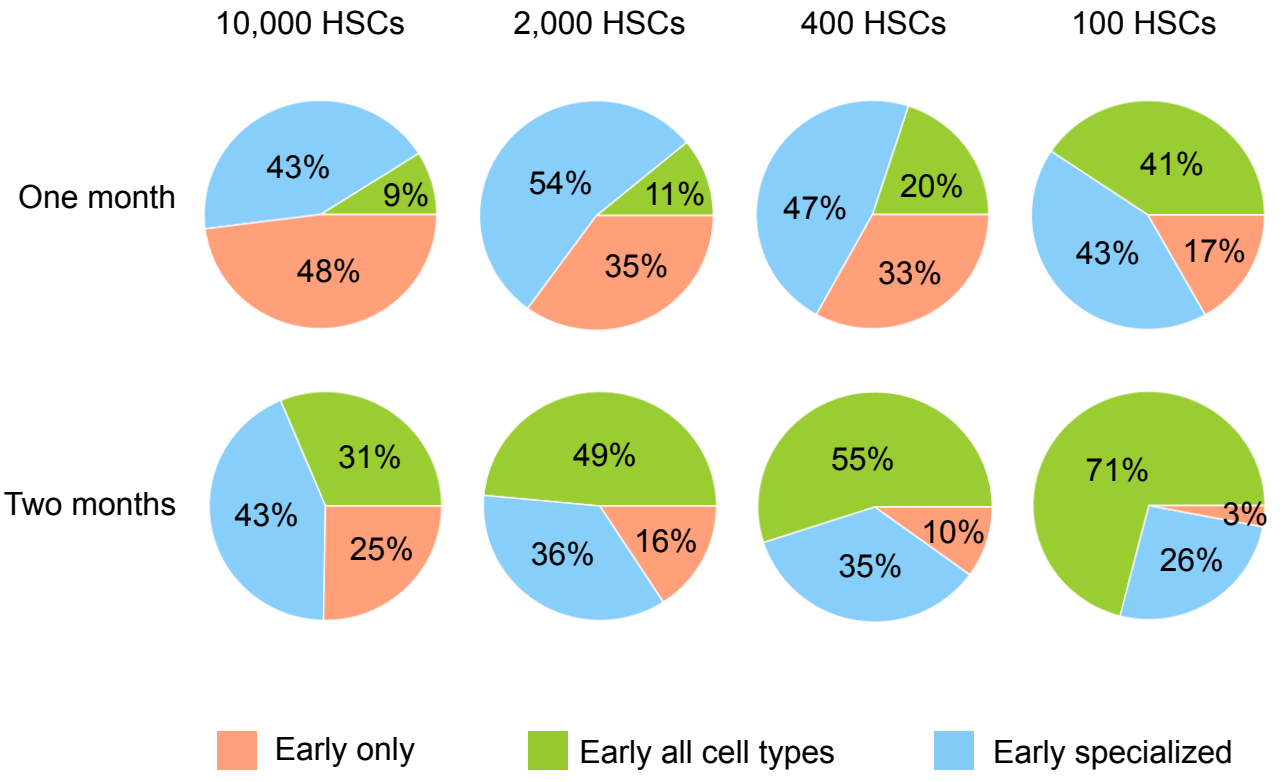

10,000 HSCs
2,000 HSCs
400 HSCs
100 HSCs
One month
43%
9%
48%
54%
11%
35%
47%
20%
33%
41%
43%
17%
Two months
31%
43%
25%
49%
36%
16%
55%
10%
35%
71%
3%
26%
Early only
Early all cell types
Early specialized

Figure S6

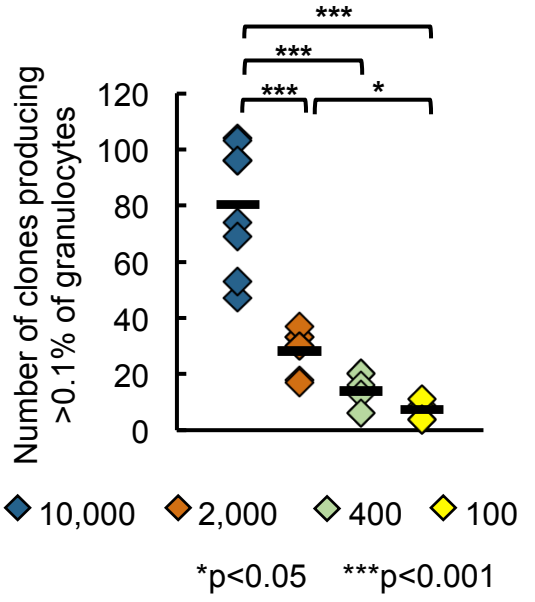

***
***
***
*
Number of clones producing >0.1% of granulocytes
120
100
80
60
40
20
0
10,000
2,000
400
100
*p<0.05 ***p<0.001

Figure S7

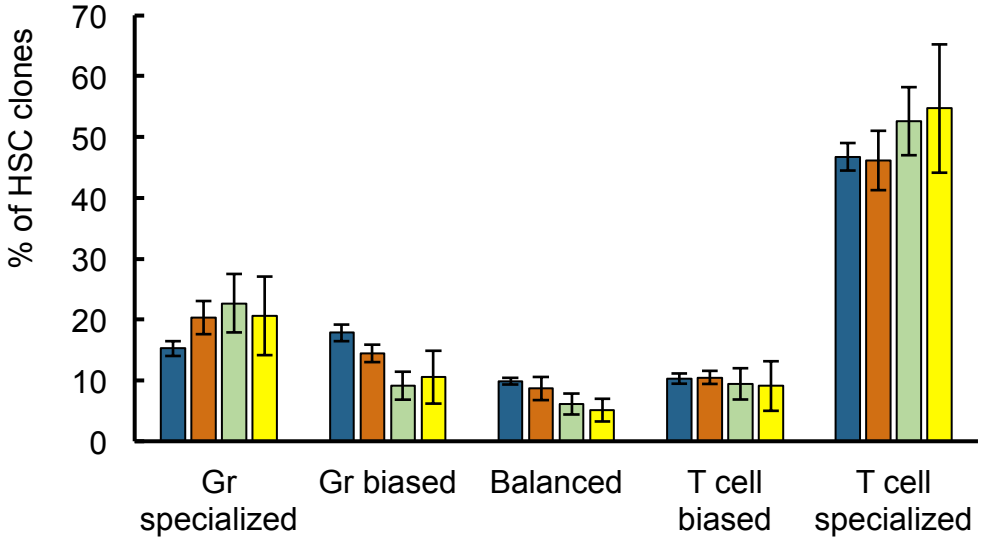

% of HSC clones
70
60
50
40
30
20
10
0
Gr specialized
Gr biased
Balanced
T cell biased
T cell specialized

Figure S1. White Blood Cell Population Size at Various HSC Transplantation Doses. ***Related to Figure 1.***

(A–C) White blood cell population sizes were measured (A) one, (B) two, and (C) six months after transplantation by flow cytometry. Bar segments represent group means. Error bars represent standard error of the mean. 7-8 mice were used per transplantation dose.

Figure S2. Replicate Analysis of barcode quantification. ***Related to Figure 1.***

Blood samples were lysed and split into replicate samples. Replicates were separately extracted for genomic DNA, amplified by PCR, and quantified using high throughput sequencing. Each data point represents a single barcode. The x and y axes represent the abundance of a barcode in each replicate sample.

Figure S3. Removal of Sequencing Background. ***Related to Figure 1.***

Molecular grade water was processed via PCR and sent for sequencing. False positive barcodes detected in the water were quantified. Barcodes from actual samples were only considered valid if they exceeded 99% of the signals from water samples. Each graph shows the barcodes of a mouse sample compared to the water sample. Each blue dot represents a barcode. The red line represents the 99% threshold.

Figure S4. Chimerism over Time at Different Transplantation Doses. ***Related to Figure 1.***

(A–D) Donor, helper, and host chimerisms in (A) granulocytes, (B) B cells, (C) CD4T cells, and (D) CD8T cells were measured monthly by flow cytometry. Data points represent group means. Error bars depict standard error of the mean. 7-8 mice were used per transplantation dose.

Figure S5. Early Granulocyte Production Changes with HSC Dose. ***Related to Figure 5.***

Pie charts show the percent of granulocyte production by each category of clones one and two months after transplantation. Values are the mean percentage for each HSC dose group. See also Tables S6-S7. 7-8 mice were used per transplantation dose.

Figure S6. Alternate Methods of Computing Clonal Dominance Produce Similar Results. ***Related to Figure 6.***

The number of clones that produced more than 0.1% of all granulocytes at each HSC transplantation dose. Each diamond represents one mouse; horizontal black lines represent group means.

Figure S7. Lineage Bias Analysis of Granulocytes vs. T cells. ***Related to Figure 7.***

Granulocyte production was compared to T-cell production for each HSC clone. A clone was considered to be "specialized" to a cell type if it only produced that cell type, "biased" to a cell type if it favored the production of that cell type, and "balanced" if its relative production of the two cell types was similar. The mean percent is displayed for each HSC dose. Error bars represent the standard error of the mean. 7-8 mice were used per transplantation dose.

Table S1

1 month				2 months				6 months			
	10000	2000	400		10000	2000	400		10000	2000	400
2000	***			2000	***			2000	***		
400	***	NS		400	***	**		400	***	*	
100	***	**	NS	100	***	***	NS	100	***	**	NS

NS, not significant

*p<0.05 **p<0.01 ***p<0.001

Table S2

Early-differentiating			
	10000	2000	400
2000	***		
400	***	*	
100	***	***	NS

Early only			
	10000	2000	400
2000	***		
400	***	*	
100	***	**	NS

All cell types			
	10000	2000	400
2000	***		
400	***	NS	
100	***	*	NS

Late and all cell types			
	10000	2000	400
2000	*		
400	***	NS	
100	***	NS	NS

Early and all cell types			
	10000	2000	400
2000	***		
400	***	*	
100	***	**	NS

Specialized			
	10000	2000	400
2000	***		
400	***	NS	
100	***	**	NS

Early and specialized			
	10000	2000	400
2000	NS		
400	**	NS	
100	***	*	NS

Late and specialized			
	10000	2000	400
2000	**		
400	*	NS	
100	***	NS	NS

NS, not significant

*p<0.05 **p<0.01 ***p<0.001

Table S3

Early-differentiating			
	10000	2000	400
2000	NS		
400	NS	NS	
100	**	**	NS

Early only			
	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

All cell types			
	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

Late and all cell types			
	10000	2000	400
2000	***		
400	***	NS	
100	***	NS	NS

Early and all cell types			
	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

Specialized			
	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

Early and specialized			
	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

Late and specialized			
	10000	2000	400
2000	NS		
400	NS	NS	
100	**	**	NS

NS, not significant

**p<0.01 ***p<0.001

Table S4

Gr 10,000	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	NS	NS	NS

Gr 2,000	L all	L Sp	E all
L Sp	NS		
E all	**	**	
E Sp	NS	NS	**

Gr 400	L all	L Sp	E all
L Sp	NS		
E all	**	*	
E Sp	NS	NS	NS

Gr 100	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	NS	NS	***

B 10,000	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	**	**	***

B 2,000	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	*	*	***

B 400	L all	L Sp	E all
L Sp	*		
E all	*	NS	
E Sp	NS	NS	NS

B 100	L all	L Sp	E all
L Sp	NS		
E all	**	***	
E Sp	NS	NS	***

4T 10,000	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	**	**	***

4T 2,000	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	**	**	***

4T 400	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	*	*	*

4T 100	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	NS	NS	***

8T 10,000	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	***	***	***

8T 2,000	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	**	**	***

8T 400	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	**	*	NS

8T 100	L all	L Sp	E all
L Sp	NS		
E all	***	***	
E Sp	NS	NS	***

L all, Late and all cell types; L Sp, Late and specialized; E all, Early and all cell types; E Sp, Early and specialized; NS, not significant

*p<0.05 **p<0.01 ***p<0.001

Table S5

Gr - Late and all	10000	2000	400
2000	NS		
400	*	NS	
100	*	NS	NS

Gr - Late and Sp	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

Gr - Early and all	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

Gr – Early and Sp	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

B cell - Late and all	10000	2000	400
2000	NS		
400	**	NS	
100	**	NS	NS

B cell - Late and Sp	10000	2000	400
2000	NS		
400	*	**	
100	NS	NS	NS

B cell - Early and all	10000	2000	400
2000	NS		
400	NS	*	
100	NS	NS	*

B cell - Early and Sp	10000	2000	400
2000	NS		
400	NS	NS	
100	**	NS	NS

4T cell - Late and all	10000	2000	400
2000	**		
400	***	NS	
100	***	NS	NS

4T cell – Late and Sp	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

4T cell - Early and all	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

4T cell – Early and Sp	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

8T cell - Late and all	10000	2000	400
2000	NS		
400	***	NS	
100	**	NS	NS

8T cell - Late and Sp	10000	2000	400
2000	NS		
400	NS	NS	
100	*	NS	NS

8T cell - Early and all	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

8T cell – Early and Sp	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

Late and all, Late and all cell types; Late and Sp, Late and specialized;
Early and all, Early and all cell types; Early and Sp, Early and specialized;
NS, not significant

*p<0.05 **p<0.01 ***p<0.001

Table S6

1 month all cell types			
	10000	2000	400
2000	NS		
400	NS	NS	
100	**	*	NS

1 month Sp			
	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

1 month early only			
	10000	2000	400
2000	NS		
400	*	NS	
100	***	*	*

2 month all cell types			
	10000	2000	400
2000	NS		
400	NS	NS	
100	**	NS	NS

2 month Sp			
	10000	2000	400
2000	NS		
400	NS	NS	
100	NS	NS	NS

2 month early only			
	10000	2000	400
2000	NS		
400	*	NS	
100	**	NS	NS

Sp, Specialized; NS, not significant

Table S7

1 month 10,000		
	all	Sp
Sp	***	
e.o.	***	NS

1 month 2,000		
	all	Sp
Sp	***	
e.o.	*	*

1 month 400		
	all	Sp
Sp	*	
e.o.	NS	NS

1 month 100		
	all	Sp
Sp	NS	
e.o.	NS	NS

2 months 10,000		
	all	Sp
Sp	**	
e.o.	NS	***

2 months 2,000		
	all	Sp
Sp	NS	
e.o.	**	*

2 months 400		
	all	Sp
Sp	NS	
e.o.	*	NS

2 months 100		
	all	Sp
Sp	**	
e.o.	***	NS

all, all cell types; Sp, specialized; e.o., Early only;
NS, not significant

*p<0.05 **p<0.01 ***p<0.001

Table S1. Significance Values for the Line Graph Shown in Figure 2A. ***Related to Figure 2.***

Significance of the comparisons between HSC dose groups for the number of granulocyte-producing clones detected at each time point. NS, not significant.

Table S2. Significance Values for the Number of Clones in Each Category of the Venn Diagrams Shown in Figure 4. ***Related to Figure 4.***

Significance of the comparisons between HSC dose groups. NS, not significant.

Table S3. Significance Values for the Proportion of Clones in Each Category of the Venn Diagrams Shown in Figure 4. ***Related to Figure 4.***

Significance of the comparisons between HSC dose groups. NS, not significant.

Table S4. Significance Values for the Comparisons of Blood Production by Each Category of the Pie Charts Shown in Figure 5. ***Related to Figure 5.***

Significance of the comparisons for each cell type and HSC dose. L all, late and all cell types; L Sp, late and specialized; E all, early and all cell types; E Sp, early and specialized; NS, not significant.

Table S5. Significance Values for the Comparisons of Blood Production at Each Transplantation Dose of the Pie Charts Shown in Figure 5. ***Related to Figure 5.***

Significance of the comparisons for each cell type and category of clonal blood production. Late and All, late and all cell types; Late and Sp, late and specialized; Early and All, early and all cell types; Early and Sp, early and specialized; NS, not significant.

Table S6. Significance Values for the Comparisons of Granulocyte Production at Each HSC Transplantation Dose Group of the Pie Charts Shown in Figure S5. ***Related to Figure 5.***

Significance of the comparisons for each category of clone and time point. Sp, Early specialized; NS, not significant.

Table S7. Significance Values for the Comparisons of Granulocyte Production by Each Category of the Pie Charts Shown in Figure S5. ***Related to Figure 5.***

Significance of the comparisons for each time point and HSC transplantation dose. Sp, Early specialized; all, Early all cell types; e.o., early only; NS, not significant.