Table S1: Data set summary and patient characteristics. HCT-graft types: TCD: T-cell depleted graft (*ex-vivo*) by CD34+selection; PBSC: peripheral blood stem cells; BM: bone marrow; cord: umbilical cord blood; Conditioning intensity: Bacigalupo classification, graded categories from most to least intense (ABLATIVE, REDUCE, NONABL).

patients		2,926
HCT therapies*		3,060
blood samples	total	450,635
	between HCT-day -21 and HCT-day 183	193,396
Disease	Leukemia	1,635
	Non-Hodgkin's Lymphoma	415
	Multiple Myeloma	170
	Hodgkin's disease	88
	other	752
HCT graft type	TCD	1,106
	PBSC unmodified	959
	BM unmodified	617
	cord	378
Conditioning intensity	ABLATIVE	65%
	REDUCE	21%
	NONABL	13%
Gender	M	58%
	F	42%
Age of adults (years)	25%-tile	39
	mean	50
	75%-tile	62
Microbiome samples	total	12,633
	- from patients with blood data	10,680
	- of those, post engraftment	4,179
	- of those with daily change in WBC	2,615
	patients with microbiome sample	1,290

*) some patient received several HCTs

Table S2: Patient and HCT characteristics of 24 patients enrolled in the randomized controlled FMT trial.

	control	FMT treated
N patients	10	14
ABLATIVE	6	7
REDUCE	4	7
BM unmodified	1	3
PBSC unmodified	3	4
TCD	5	3
cord	1	4

Table S3: Patient and HCT characteristics of the subset of patients who did not donate microbiota samples.

patients		1,010
Disease distribution	Leukemia	53%
	Non-Hodgkin's Lymphoma	17%
	Multiple Myeloma	5%
	Hodgkin's disease	5%
	other	20%
HCT graft type	TCD	40%%
	PBSC unmodified	31%
	BM unmodified	17%
	cord	12%
Conditioning intensity	ABLATIVE	65%
	REDUCE	14%
	NONABL	21%
Gender	M	58%
	F	42%
Age (years)	25%-tile	36
	mean	47
	75%-tile	59

Table S4: Patient and HCT characteristics of the subset of patients who donated microbiota samples.

patients		1,294
Disease distribution	Leukemia	51%
	Non-Hodgkin's Lymphoma	15%
	Multiple Myeloma	8%
	Hodgkin's disease	3%
	other	23%
HCT graft type	TCD	37%
	PBSC unmodified	38%
	BM unmodified	9%
	cord	16%
Conditioning intensity	ABLATIVE	55%
	REDUCE	34%
	NONABL	11%
Gender	M	59%
	F	41%
Age (years)	25%-tile	46
	mean	54
	75%-tile	65

Table S5: Patient and HCT characteristics of the Duke University patient cohort.

patients		493
Disease distribution	Lymphoma	11%
	Leukemia	50%
	Non-Hodgkin's Lymphoma	4%
	Multiple Myeloma	8%
	Hodgkin's disease	4%
	other	24%
HCT graft type	TCD	0%
	PBSC unmodified	72%
	BM unmodified	11%
	cord	16%
Conditioning intensity	ABLATIVE	92%
	NONABL	7%
Gender	M	65%
	F	35%
Age (years)	25%-tile	41
	mean	49
	75%-tile	57

Supplementary methods and results

FMT procedure

Patient sample collection protocols were approved by the Memorial Sloan Kettering Cancer Center Institutional Review and Privacy Board (ClinicalTrials.gov identifier: NCT02269150) and described in full in the original publication. Briefly, patients' stool was collected when they first entered the clinic. We chose autologous, as opposed to feces from a heterologous donor, because of potential safety concerns. The stool was tested for the presence of potential intestinal pathogens including *C. difficile* and frozen (−80°C). If a patient was randomized to receive treatment after engraftment of neutrophils, the thawed sample was re-administered via an enema. Due to the strenuous nature of this procedure, it was deemed unethical to administer a mock enema to control patients. Subjects whose pre–allo-HSCT feces demonstrated low microbial diversity (IS index < 2.0) or tested positive for the presence of an intestinal pathogen, for example, *C. difficile*, were excluded from randomization. After successful hematopoietic cell engraftment (three consecutive blood neutrophil counts ≥500 per mm3), subjects underwent testing of a fecal specimen collected after engraftment to determine the presence of the Bacteroidetes phylum via quantitative PCR. Individuals with low abundance of Bacteroidetes (<0.1% total 16S) were eligible to proceed to randomization and treatment. Eligible subjects were 1:1 randomized to undergo auto-FMT with the subject's stored pre–allo-HSCT feces versus no fecal transplantation. Randomization was stratified by cord blood source versus non–cord blood source. Subjects could be randomized within a 28-day window after engraftment. Subjects who were critically ill or required prolonged microbiota-perturbing antibiotics through the designated 28-day period were excluded from randomization.

Alternative analysis of FMT effect on white blood cell counts

In the main text we considered a random effect per day post neutrophil engraftment. Our choice of modeling day_i as a random intercept term was to allow the cell counts to follow any function of day post FMT. Alternatively, we conducted another analysis now with day as a linear predictor:

$$y_{ij} = \beta_0 + day + armpost * FMT_{ij} + patient_j + \varepsilon_{ij}, \quad i = 0, \dots, D, j = 1, \dots, P,$$

This reduced significance but did not alter our main conclusions: FMT associated with increased neutrophils (coefficient FMT: 1.44, p=0.002), lymphocyte (coefficient FMT: 0.08, p=0.13), and monocytes (coefficient FMT: 0.34, p<0.001).

Survival analysis

We sought to analyze 3-year survival following neutrophil engraftment in the patient cohort analyzed in our main analyses. We observed 668 deaths during that period (see Kaplan Meier plot Extended Data Fig. 10). We used a Cox proportional hazards model to analyze 3-year patient survival following neutrophil engraftment using the lifelines (v0.24) package for the Python programming language (DOI:10.5281/zenodo.3677104).

$$h(t) = h_0(t)\exp\left(b_1X_1 + b_2X_2 + \cdots + b_nX_n\right)$$

where h(t) is the expected hazard at time t, $h_0(t)$ is the baseline hazard. As predictors, *X*, we included the median total white blood cell count during the first 100 days following the time when FMT was usually performed (i.e. 37 days after neutrophil engraftment), sex, and age removing patients who lacked sufficient blood data during this post-engraftment period. Disease and stem cell graft source were used to stratify patients (using the "strata" parameter of the Cox proportional hazard fit function), as including them in the form of intercept terms violated the proportional hazards assumptions. This showed a mild but detectable association of total white blood cell counts during the investigated interval with improved survival:

N:	2,013
events:	668
strata:	'disease', 'hct-source'

	coef	exp(coef)	lower 0.95	upper 0.95	p-value
average total wbc count (z-scored)	-0.09	0.91	-0.18	0	0.04
age (z-scored)	0.19	1.21	0.1	0.28	<0.005
SEX:F	0.03	1.03	-0.12	0.19	0.68

Dynamic systems analyses

To investigate if the composition of the gut microbiota is associated with the dynamics of circulating white blood cells, we analyzed detailed blood and clinical metadata of HCT patients between 3 days before HCT and until 100 days post neutrophil engraftment. The blood of each patient is monitored throughout this therapy, and medications are administered to modulate the immune cell dynamics, including GCSF (Fig. 1) to increase neutrophil counts, and immunosuppressants such as mycophenolate mofetil or tacrolimus to prevent complications such as graft-vs-host disease. During this period, patients are monitored carefully, and our analysis included on average 63 longitudinal host phenotype measurements per patient in the form of complete blood counts (CBCs) which quantify the most abundant immune cells in circulation (Fig. 1, complete data description and exclusion criteria in Extended Data Fig. 1). Combining this data with detailed medication and clinical metadata—and microbiome data where available—allowed us to apply dynamic systems inference approaches on intervals during which changes in WBC counts were observed.

Covariates included in interval data

For a given daily interval, a medication was considered present for at least part of the interval when it was administered on either endpoint. Administration events were obtained from parsing the institutional task data base which contains drug and treatment administrations performed on patients. For the microbiota, data from the end day were considered for the interval, and for blood stream infection data, either endpoint was considered. Homeostatic feedback calculations used the geometric mean of the white blood cell counts between the two endpoints. We included only those covariates that were present during at least 10 intervals.

Our data comprises >1.6M recorded administrations of 806 different medications with durations provided. All patients analyzed in stage 2 had available medication records, but ~10% of the patients without microbiome data had missing medication records and/or incomplete metadata. In case of missing continuous metadata, missing values were filled with means.

The stem cell graft source is a major determinant of engraftment times and can affect recovery dynamics[1,2], and we therefore included intercept terms for unmodified peripheral blood stem cell grafts (PBSC), bone marrow (BM), T-cell depleted graft (*ex-vivo*) by CD34+selection (TCD) and cord blood (cord) in stage 1.

Patients received a variety of conditioning regimens comprised of various doses of chemotherapy and, in some cases, irradiation. There are dozens of conditioning regimes for HCT[3]. The standard approach in the allo-HCT field for observational studies is to categorize them by the Bacigalupo classification, which uses three graded categories from most to least intense[4]: Myeloablative ("ABLATIVE"), Reduced Intensity ("REDUCE"), and Nonmyeloablative ("NONABL"). We included the conditioning intensity as indicator variables.

Data exclusion for white blood cell dynamic (stage 1 and 2)

Our dynamics analyses focus on the daily changes in white blood cell counts from one day to another during recovery of the circulatory immune cell system. To analyze the kinetics of this reconstitution, we excluded data when (see also flow chart below):

- On the day of HCT, patients were younger than 18 years
- Patients did not engraft
- Patients had a second transplant within 100 days
- Samples were taken outside the window of 100 days starting from neutrophil engraftment
- In case there were multiple blood samples per patient and day, the one closest to noon was chosen
- A patient died within the first 3 months after HCT
- A sample was taken within 1 week of FMT

This data exclusion is encoded as two separate columns in the tidy data table WBC.csv, by a Boolean indicator column named "include", and the column "exclude_reason" of string type (Extended Data Fig. 1). The algorithm and code for stage 1 and 2 are available online (https://github.com/jsevo/wbcdynamics_microbiome/).

Additional results

Most of the taxa that strongly associated with white blood cell dynamics were obligate anaerobes that may affect immune homeostasis[5–8]. *Rothia*, was a notable exception: this aerobic genus is typically found in the oral cavity[9] but can become an opportunistic pathogen in immunosuppressed patients and is not known to provide metabolic functions to the host[10–12].

Shotgun sequencing data processing

We removed normal optical duplicates in paired FASTQ files using the clumpify.sh tool from the BBMap package (BBMap – Bushnell B. – https://www.sourceforge.net/projects/bbmap/), producing a pair of deduped read files. Using the bbduk.sh script in the BBMap package, we trimmed the right and left side of a read in a pair to Q10 using the Phred algorithm. A pair of reads was dropped if any one of them has a length shorter than 51 nucleotides after trimming. We trim 3'-end adapters using a kmer of length 31, and a shorter kmer of 9 at the other end of the read. One mismatch was allowed in this process, and we allowed adapter trimming based on pair overlap detection (which does not require known adapter sequences) using the 'tbo' parameter. We used the 'tpe' parameter to trim the pair of reads to the same length.

Removal of human contamination was done using Kneaddata with paired-end reads, employing BMTagger. The BMTagger database was built with human genome assembly GRCh38. After

decontamination, the paired-end reads were concatenated to a single FASTQ file as the input for functional profiling with the Humann2 pipeline (main methods).

Rationale for using estimations of total bacterial abundances for bi-directional analysis of bacterial and white blood cell dynamics

In order to assess the association of bacterial populations with antibiotics, the ecosystem of other bacteria and the count of white blood cells in circulation, we employed a similar approach to that used in stage 1. To calculate the dependent variable, i.e. the log-changes per day in total counts, we had to estimate the absolute abundances of bacteria. This is needed because inferences of dynamic equations like the ones used here cannot be conducted on observations of changes in relative abundances. Briefly, a positive change in relative abundances of a bacterium could be due to the focal bacterium's counts increasing, that of other species' decreasing, both focal and others increasing or decreasing but the focal species faster or slower so, respectively. This means that when attempting to use observations of relative abundance changes as dependent variables to infer interactions like we do here, one would try to solve one more unknown than one has information for. Unless specific assumptions can be justified, one therefore must estimate a total count. We here do this by estimating the total population density using qPCR.

This is important when estimating the sign and the effect sizes of antibiotics, other bacterial species in the ecosystem or the white blood cells in circulation on the changes of a focal taxon. We do not know what aspects of the microbiota can be affected by the immune system, and it is entirely possible that the immune system evolved to "control" relative abundances to achieve microbiota homeostasis[13]. Yet, in order to estimate in which direction the immune system's effect operates on any one taxon, for the reasons above, we require observations of total changes per taxon.

Duke data collection

Data at Duke university was collected analogously to the procedures at MSKCC. Complete blood counts were assessed on the Sysmex XN, Pro00050975 platform. Clinical metadata was obtained from the Duke Enterprise Data Unified Content Explorer (DEDUCE) and Duke Adult Blood and Marrow Transplant (ABMT) data base. All sequencing was conducted at MSKCC following the same protocols.

Bibliography

1. Anasetti, C. *et al.* Peripheral-blood stem cells versus bone marrow from unrelated donors. *N. Engl. J. Med.* **367,** 1487–1496 (2012).
2. Davies, S. M. *et al.* Engraftment and survival after unrelated-donor bone marrow transplantation: a report from the national marrow donor program. *Blood* **96,** 4096–4102 (2000).
3. Gyurkocza, B. & Sandmaier, B. M. Conditioning regimens for hematopoietic cell transplantation: one size does not fit all. *Blood* **124,** 344–353 (2014).
4. Bacigalupo, A. *et al.* Defining the intensity of conditioning regimens: working definitions. *Biol. Blood Marrow Transplant.* **15,** 1628–1633 (2009).
5. Ladinsky, M. S. *et al.* Endocytosis of commensal antigens by intestinal epithelial cells regulates mucosal T cell homeostasis. *Science* **363,** (2019).
6. Balmer, M. L. *et al.* Microbiota-derived compounds drive steady-state granulopoiesis via MyD88/TICAM signaling. *J. Immunol.* **193,** 5273–5283 (2014).
7. Yoshii, K., Hosomi, K., Sawane, K. & Kunisawa, J. Metabolism of dietary and microbial vitamin B family in the regulation of host immunity. *Front. Nutr.* **6,** 48 (2019).
8. Zaza, G. *et al.* Impact of maintenance immunosuppressive therapy on the fecal microbiome of renal transplant recipients: Comparison between an everolimus- and a standard tacrolimus-based regimen. *PLoS One* **12,** e0178228 (2017).
9. Hall, M. W. *et al.* Inter-personal diversity and temporal dynamics of dental, tongue, and salivary microbiota in the healthy oral cavity. *npj Biofilms and Microbiomes* **3,** 2 (2017).
10. Ramanan, P., Barreto, J. N., Osmon, D. R. & Tosh, P. K. Rothia bacteremia: a 10-year experience at Mayo Clinic, Rochester, Minnesota. *J. Clin. Microbiol.* **52,** 3184–3189 (2014).
11. Ulven, T. Short-chain free fatty acid receptors FFA2/GPR43 and FFA3/GPR41 as new potential therapeutic targets. *Front. Endocrinol. (Lausanne)* **3,** 111 (2012).
12. Smith, P. M. *et al.* The microbial metabolites, short-chain fatty acids, regulate colonic Treg cell homeostasis. *Science* **341,** 569–573 (2013).
13. Foster, K. R., Schluter, J., Coyte, K. Z. & Rakoff-Nahoum, S. The evolution of the host microbiome as an ecosystem on a leash. *Nature* **548,** 43–51 (2017).