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Trends in Immunology

Opinion

Hematopoiesis in numbers

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Hematopoiesis is a dynamic process in which stem and progenitor cells give rise to the ~10^{13} blood and immune cells distributed throughout the human body. We argue that a quantitative description of hematopoiesis can help consolidate existing data, identify knowledge gaps, and generate new hypotheses. Here, we review known numbers in murine and, where possible, human hematopoiesis, and consolidate murine numbers into a set of reference values. We present estimates of cell numbers, division and differentiation rates, cell size, and macromolecular composition for each hematopoietic cell type. We also propose guidelines to improve the reporting of measurements and highlight areas in which quantitative data are lacking. Overall, we show how quantitative approaches can be used to understand key properties of hematopoiesis.

Highlights

Informed by published measurements, our calculations suggest that, in mice, 5×10^3 hematopoietic stem cells (HSCs) can give rise to 10^{10} hematopoietic cells, while, in humans, 2.5×10^4–1.3×10^6 HSCs can give rise to 10^{13} mature hematopoietic cells

In humans and mice, most hematopoietic cells are short-lived, on the order of days or weeks. As such, blood cells are constantly being produced to maintain blood homeostasis. In humans, blood cells account for 90% of all cellular turnover.

Our calculations predict that the amount of murine and human myeloid and erythroid cells produced each day are approximately the same order of magnitude, despite the absolute number of erythroid cells being several orders of magnitude higher than myeloid cells in both species. This is due to large differences in the expected lifespan of each cell type.

Despite recent progress, reports on the absolute measurements of cell numbers across tissues throughout lifespans are missing for many cell types and species, particularly in humans; this represents a fruitful area of empirical investigation.

A quantitative understanding of hematopoiesis

Hematopoiesis is the process by which hematopoietic stem and progenitor cells (HSPCs) give rise to blood and immune cells [1]. Here, we argue that numbers are tools to sharpen our understanding of hematopoiesis [2], but it is challenging and time consuming to find robust values in the literature. Numbers come from various studies and it is rarely discussed how they fit into our broader understanding of hematopoiesis. To address these issues, we review existing numbers in murine and human hematopoiesis, and generate a set of reference values for murine hematopoiesis. Specifically, we discuss our current understanding of cell numbers, division and differentiation rates, cell size, and macromolecular composition for each hematopoietic cell type. We also highlight limitations of existing measurements, and instances in which quantitative data are lacking. By using simple calculations, we show examples of how this numerical reference can identify key properties of hematopoiesis and provide suggestions for future research.

How many cells are in the hematopoietic system?

Quantifying absolute cell numbers is important to help contextualize changes that occur in hematopoiesis across lifespan and disease. Here, we summarize current measurements of cell numbers across the major hematopoietic organs, lineages, and compartments, focusing on a 12-week-old, 22-g adult female mouse for which we have the most available data (Files S1, S2, and Calculation S1a in the supplemental information online [3]). We also summarize known cell numbers in humans. The criteria and sources used to generate reference values are provided in File S1 in the supplemental information online, along with metadata for each measurement.

How many cells are there in hematopoietic organs?

Consolidating cell numbers across the hematopoietic system is nontrivial because: (i) cells are heterogeneously distributed across tissues; (ii) differences in sample processing can affect cell yield; (iii) most measurements are expressed in relative rather than absolute terms; and (iv) most numbers come from samples rather than entire tissues. Reviewing existing mouse numbers and accounting for these factors (Table 1 and Figure 1; Files S1, S2, and Calculation S1a in the supplemental information online), the blood, bone marrow, spleen, lymph nodes,

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Table 1. Cellular composition of the main hematopoietic organs in the reference 12-week-old, 22-g female C57BL/6J mouse, according to calculations in Figure S1 in the supplemental information online[a,b]

Mouse organ /cell population	Value	Unit	Refs
Reference mouse			
Weight	22	g	[56]
Strain	B6		
Age	84	Days	
Sex	Female		
Bone marrow			
Total cell count	4.5E+08	Cells	[29,35,57]
Total nucleated cells	2.6E+08	Cells	[57]
Lin^- cells	2.0E+07	Cells	[10]
Phenotypic HSCs[c]	1.6E+04	Cells	[10]
Active HSCs	5.2E+03	Cells	[10]
MPPs[d] (non-HSC LSK)	1.4E+05	Cells	[10]
Erythromyeloid progenitors	1.7E+06	Cells	
CMPs[e]	4.0E+05	Cells	[10]
GMPs[f]	5.1E+05	Cells	[10]
MEPs[g]	8.0E+05	Cells	[10]
Megakaryocyte progenitors	2.6E+04	Cells	[27]
CLPs	2.5E+05	Cells	[10]
Lin^+ cells	4.3E+08	Cells	
Erythrocytes	1.8E+08	Cells	[35]
Megakaryocytes	7.7E+05	Cells	[36]
Platelets	5.6E+06	Cells	[35]
Granulomyeloids	1.5E+08	Cells	[35]
B cells	8.6E+07	Cells	[35]
Peripheral blood			
Volume	1.5E+03	ul	[58]
Total cell count	1.6E+10	Cells	
Erythrocytes	1.5E+10	Cells	[35]
Platelets	1.4E+09	Cells	[35]
Granulomyeloids	1.7E+06	Cells	[35]
B cells	8.3E+06	Cells	[35]
T cells	2.6E+03	Cells	[35]
Spleen			
Mass	7.9E-02	g	[59]
Total cell count	2.1E+08	Cells	
Erythrocytes	8.2E+07	Cells	[35]
Platelets	5.1E+07	Cells	[35]
Granulomyeloids	2.7E+06	Cells	[35]
B cells	4.7E+07	Cells	[35]
Follicular mature B cells	3.3E+07	Cells	[35]
T cells	2.0E+07	Cells	[35]

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Glossary

Capture–recapture longitudinal genomic analyses: technique to estimate population sizes when it is not possible to count each individual cell. The method involves taking a small population of cells and labeling them, then reintroducing them back into their initial environment and determining the ratio of marked to unmarked cells at a later timepoint.

$CD34^+$ progenitors: human progenitor compartment containing HSCs, MPPs, and restricted-potential progenitors.

Common lymphoid progenitors (CLPs): give rise to B and T lymphocytes. In mice, they express the following markers: $cKit^{low}$ $Sca1^{low}$ $IL7R^+$ $Flk2^-$.

Common myeloid progenitors (CMPs): give rise to erythroid, megakaryocyte, and myeloid lineages. In mice, they express the following markers: $cKit^+$ $Sca1^-$ $CD16/32^-$ $CD34^+$.

Differentiation active: stem or progenitor cell that is in the process of changing into a more mature hematopoietic cell type.

Division-linked dilution assay: a dye is split equally between daughter cells during cell division. Consequently, the amount of dye in each cell is indicative of how many times it has divided, over a small number of generations.

Endomitoses: chromosome replication in the absence of a cell division, resulting in a polypoidal cell.

Fate mapping: labeling cells with a heritable mark to understand developmental processes.

Granulocyte–monocyte progenitors (GMPs): give rise to the myeloid lineages. In mice, they express the following markers: $cKit^+$ $Sca1^-$ $CD16/32^+$ $CD34^+$.

Hematopoietic stem cells (HSCs): functionally defined as a multipotent cell capable of long-term self-renewal. In this study, we consider an immunophenotypic HSC to express the following pattern of markers: Lin^-Kit^+ $Sca\text{-}1^+CD150^+CD48^-$.

Lentiviral vector integration site: in gene therapy, a viral vector is used to introduce new genetic material into a host organism. Given that the integration of the virus is stochastic, the precise location of the new genetic material in the host genome acts as a genetic label to perform fate mapping.

Table 1. (continued)

Mouse organ /cell population	Value	Unit	Refs
Lymph nodes			
Average number lymph nodes in whole body	2.7E+01	Lymph nodes	[60,61]
Total cell count	7.6E+07	Cells	[62]
Erythrocytes	2.5E+06	Cells	[35]
B cells	3.5E+07	Cells	[35]
Follicular mature B cells	2.4E+07	Cells	[63]
T cells	3.6E+07	Cells	[35]
Others	1.5E+06	Cells	[35]
Thymus			
Total cell count	2.1E+08	Cells	[64]
Erythrocytes	3.6E+07	Cells	[35]
Platelets	2.8E+06	Cells	[35]
B cells	8.5E+06	Cells	[35]
T cells	1.6E+08	Cells	[35]

[a]Abbreviations: CMP, common myeloid progenitor; GMP, granulocyte-monocyte progenitor; HSC, hematopoietic stem cell; MEP, megakaryocyte-erythroid progenitor; MPP, multipotent progenitor.
[b]See also Files S1 and S2 in the supplemental information online.
[c]$Lin^-Kit^+Sca\text{-}1^+CD150^+CD48^-$.
[d]$Lin^-Kit^+Sca\text{-}1^+CD48^+$.
[e]Lin^-Kit^+ $CD16/32^-$ $CD34^+$.
[f]Lin^-Kit^+ $CD16/32^+$ $CD34^+$.
[g]Lin^-Kit^+ $CD16/32^-$ $CD34^-$.

and thymus have 1.6×10^{10}, 4.5×10^8, 2.1×10^8, 7.6×10^7 and 2.1×10^8 hematopoietic cells respectively (Table 1; Files S1, S2, and Calculation S1a in the supplemental information online). Adding these values, this gives 1.7×10^{10} cells in total – likely an underestimate of the true value, as organs such as the liver and gut are not considered in this calculation.

In humans, the standard reference person, historically defined as a 20–30-year-old male, weighing 70 kg and measuring 170 cm in height [4], has ~1.2×10^{12} nucleated bone marrow cells and 2.8×10^{13} blood cells (Files S1 and S2 in the supplemental information online) [5]. Surprisingly, direct measurements of human spleen and lymph node cellularity are missing. Assuming the average spleen weighs 130–150 g [6] and a single cell weighs 1 ng [7], there are ~10^{11} human splenic cells. A 1974 study extrapolating data from rats to humans reported a total of 1.9×10^{11} lymph node lymphocytes [8], distributed among 1200 lymph nodes [9]. This reported number of lymph nodes, obtained from a reference digital image library, [9] is higher than the 460 lymph nodes cited by [8], for which we could find no primary data. The lack of direct measurements of the spleen and the huge range between reported lymph node numbers illustrate that much work is needed to quantify the human hematopoietic system.

How many hematopoietic stem cells are there?

Flow cytometry analyses suggest that murine **hematopoietic stem cells** (HSCs; see Glossary) (Lin^-Kit^+ $Sca\text{-}1^+CD150^+CD48^-$) represent 0.006% of nucleated bone marrow cells and ~16 000 cells in total [10]. However, this is likely to be an overestimate, as a significant proportion (~30-70%) of immunophenotypic HSCs do not meet the functional criteria of **multipotency** and **self-renewal** [11–13]. In addition, only a subset of HSCs actively contributes to hematopoiesis (**differentiation active**), as determined by **fate mapping** [10,14] and **lineage tracing**

Lineage negative: cell that does not express surface markers associated with the mature blood cell lineages.
Lineage tracing: fate mapping carried out at single cell resolution, typically by introducing a genetic label.
Megakarocyte progenitors (MkPs): express the following markers in mice: $Sca1^-$ $FcyR^{low}$ $CD9^+$ $CD41^+$.
Megakaryocyte-Erythroid Progenitors (MEPs): give rise to erythroblasts and megakaryocytes. In mice, they expresses the following markers: $cKit^+$ $Sca1^-$ $CD16/32^-$ $CD34^-$.
Multipotent progenitors (MPPs): functionally defined as a cell capable of producing all lineages but lacking long-term self-renewal. In this study, we consider that murine MPPs are all Lin^- Kit^+ $Sca\text{-}1^+$ cells that are not within the immunophenotypic HSC compartment.
Multipotency: potential to differentiate into diverse blood cell lineages.
Post-transplantation hematopoiesis: formation of new blood cells from stem cells that have been injected into a conditioned (typically irradiated) host.
Restricted potential progenitors (RPPs): hematopoietic cells that give rise to multiple cell types, but that are incapable of creating all hematopoietic lineages. Therefore, these cells have less potential than MPPs and HSCs; population comprises CMPs, CLPs, GMPs, MEPs, and MkPs.
Self-renewal: cell division with maintenance of an undifferentiated cell state.

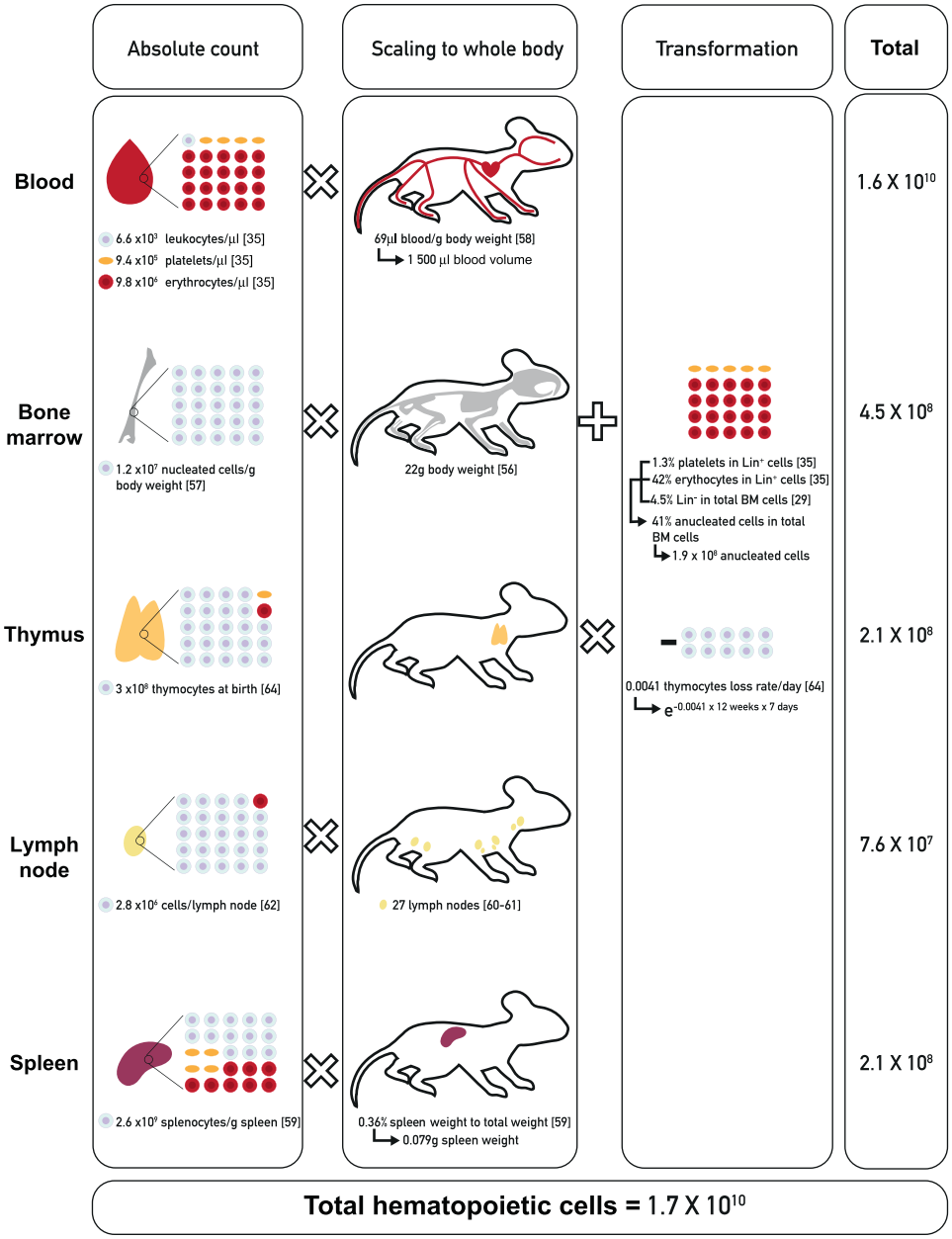


Figure 1. Generating reference values for total murine hematopoietic cell numbers. Most studies enumerate cell frequency by measuring a tissue sample. Consequently, these values need to be transformed to absolute counts, scaled up to the entire organism, and corrected for sampling and technical artefacts. All the numbers are derived for a reference 12-week-old adult female mouse, weighing 22 g. For total cell numbers in blood, we summed the total numbers of leukocytes, platelets, and erythrocytes per microliter, and multiplied this by the total blood volume for our reference mouse. In the bone marrow (BM), we scaled the measurement of the number of nucleated cells per gram of body weight to our 22-g reference mouse and we added the proportion of anucleated erythrocytes and platelets to this number. For the thymus, the total number of thymocytes at birth was corrected for the loss of thymocytes associated with aging using the rate of thymocyte loss per day for a 12-week-old reference mouse. For the lymph node, the number of cells within one lymph node was multiplied by the average total number of lymph nodes in one mouse. For the spleen, we took a measurement of the total splenocytes per gram of spleen and then scaled it by the mass of the spleen in our reference mouse. A full explanation of these calculations, along with supporting references, is provided in Files S1 and S2 in the supplemental information online. Original data from [29,35,56–62,64].

[15–17] experiments in mice. Differentiation-active murine HSC numbers range from 2770 to 22 400 [10,18,19]. Given that there are 16 000 immunophenotypic HSCs in mice and at least one-third of Tie2-YFP-labeled murine HSCs give rise to mature cells, as measured by fate mapping [10], we use ~5200 as a reference value for the number of differentiation-active murine HSCs.

In adult humans, the number of differentiation-active HSCs ranges from 25 000 to 1 300 000 based on inference from **capture–recapture longitudinal genomic analyses** of a 59-year-old male [20] and allele frequency data from a large cohort of blood cancer-free individuals [21]. In a gene therapy context, fewer human HSCs (1600–4300) actively contribute to long-term **post-transplantation hematopoiesis**, as inferred from **lentiviral vector integration site** data in patients with Wiskott–Aldrich Syndrome [22].

How many hematopoietic progenitors are there?

Downstream of HSCs are the **multipotent progenitors** (MPPs), a functionally heterogeneous population [23] that harbors multilineage potential but lacks long-term repopulating capacity post transplantation [24,25]. Flow cytometry measurements report that the MPP compartment is approximately nine times bigger than the HSC compartment [10], giving 1.4×10^5 MPPs in our reference mouse (Table 1).

Downstream of the MPP are the **restricted potential progenitors** (RPPs), which can be subdivided into the **common myeloid progenitors** (CMPs), **granulocyte–monocyte progenitors** (GMPs)**, megakaryocyte-erythroid progenitors** (MEPs), **common lymphoid progenitors** (CLPs), and **megakaryocyte progenitors** (MkPs). Based on flow cytometry measurements [10,26,27], we compute that, for each MPP, there are 2.9, 3.6, 5.7, 0.2, and 1.8 times more CMPs GMPs, MEPs, MkPs, and CLPs, respectively (Table 1; Files S1, S2, and Calculation S1b in the supplemental information online).

Summing all HSC, MPP, and RPP populations, we estimate that progenitors account for ~0.5% of the murine bone marrow (Table 1), while flow cytometry measurements suggest that **lineage-negative** cells range from 1% to 5.7% of total bone marrow [12,28–30]. Taking 4.5% as a representative value [29], we estimate that 4% of bone marrow cells (~18 million cells) are not classically defined progenitors, but do not express mature lineage markers. These cells contain both hematopoietic and nonhematopoietic cells and remain to be better functionally defined.

In humans, **CD34$^+$ progenitors** (HSCs, MPPs, and RPPs) represent an average of 2.5% of all mononucleated bone marrow cells [31,32]. Given the total number of nucleated bone marrow cells from [4], this suggests a total of ~3×10^{10} CD34$^+$ cells in our reference human. Currently, there are conflicting results about the effect of age on the frequency of CD34$^+$ cells [33,34], and it is unclear to what extent such frequencies change due to gender, ethnicity, and lifestyle factors.

How many mature hematopoietic cells are there?

At the bottom of the hematopoietic hierarchy are the terminally differentiated mature cells. To date, the most extensive measurements in mice account for all mature cells in blood, bone marrow, spleen, thymus, and lymph nodes [35]. For our reference mouse, there are 1.5×10^{10} erythrocytes, 1.5×10^9 platelets, 1.6×10^8 myeloid cells, 1.8×10^8 B lymphocytes, and 2.2×10^8 T lymphocytes (Table 1). For megakaryocytes, flow cytometry measurements (0.29% of all nucleated bone marrow cells [36]) suggest a total of 7.7×10^5 cells. Together, this yields a total of 1.7×10^{10} mature hematopoietic cells.

In humans, a recent metadata analysis [4] calculated that there are 2.5×10^{13} erythrocytes, 1.5×10^{12} platelets, 7×10^{11} T cells, 3×10^{11} B cells, monocytes 5×10^{9}, and 6.4×10^{11} neutrophils [4], consolidating measurements from human tissue samples along with measurements from rodents and primates in instances where no human data were available.

In tallying hematopoietic cell numbers (Table 1; Files S1, S2, and Calculation S1 in the supplemental information online), we find that compartment sizes vary by several orders of magnitude within and between lineages in both mice and humans. Notably, progenitors are rarer than mature cells (Figure 2, Key figure and Table 1), suggesting that significant cell expansion occurs downstream of the RPP compartment, after lineage commitment has occurred. Quantifying hematopoietic cell numbers is a topic that warrants further experimental focus (see Outstanding questions), particularly in humans. When measurements in humans are unavailable, numbers have been extrapolated from rodents. However, cross-species measurements highlight important differences calling for caution: for example, myeloid cells account for ~9–16% of all blood

Key figure

Cell numbers across different hematopoietic compartments in healthy mice

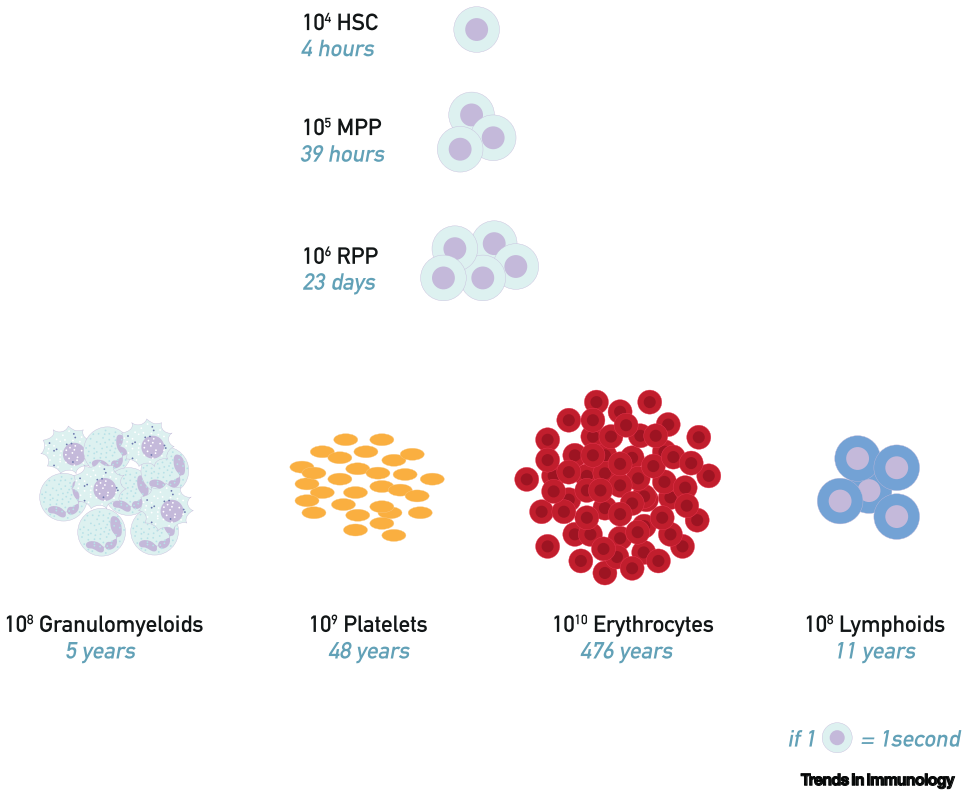


Figure 2. The total number of cells across the murine hematopoietic hierarchy were calculated using values from Table 1 in the main text. To help understand the scaling across several orders of magnitude, we place compartment sizes on a more familiar scale: time. Specifically, we place cells on a timescale in which one cell is arbitrarily equal to 1 s. All the numbers are derived for a reference 12-week-old adult female mouse, weighing 22 g (Table 1). Abbreviations: HSC, hematopoietic stem cells; MPP, multi-potent progenitors; RBC, red blood cell; RPP, restricted potential progenitor. See also Files S1 and S2 in the supplemental information online.

leukocytes in our reference mouse [3,35], but in humans, this number is ~65% [6]. Total cell numbers can help to understand how hematopoiesis changes during disease, complementing other resources, such as the human cell atlas, and facilitating cross-species comparisons to understand how the hematopoietic system has evolved.

How many hematopoietic cells do we produce per day?

The rates at which cells divide, differentiate, and die are key regulators of cell numbers [10,14] and, if perturbed, can lead to hematological malignancies [37]. Here, we discuss our current understanding of these parameters, excluding T cells, which have been reviewed elsewhere [38].

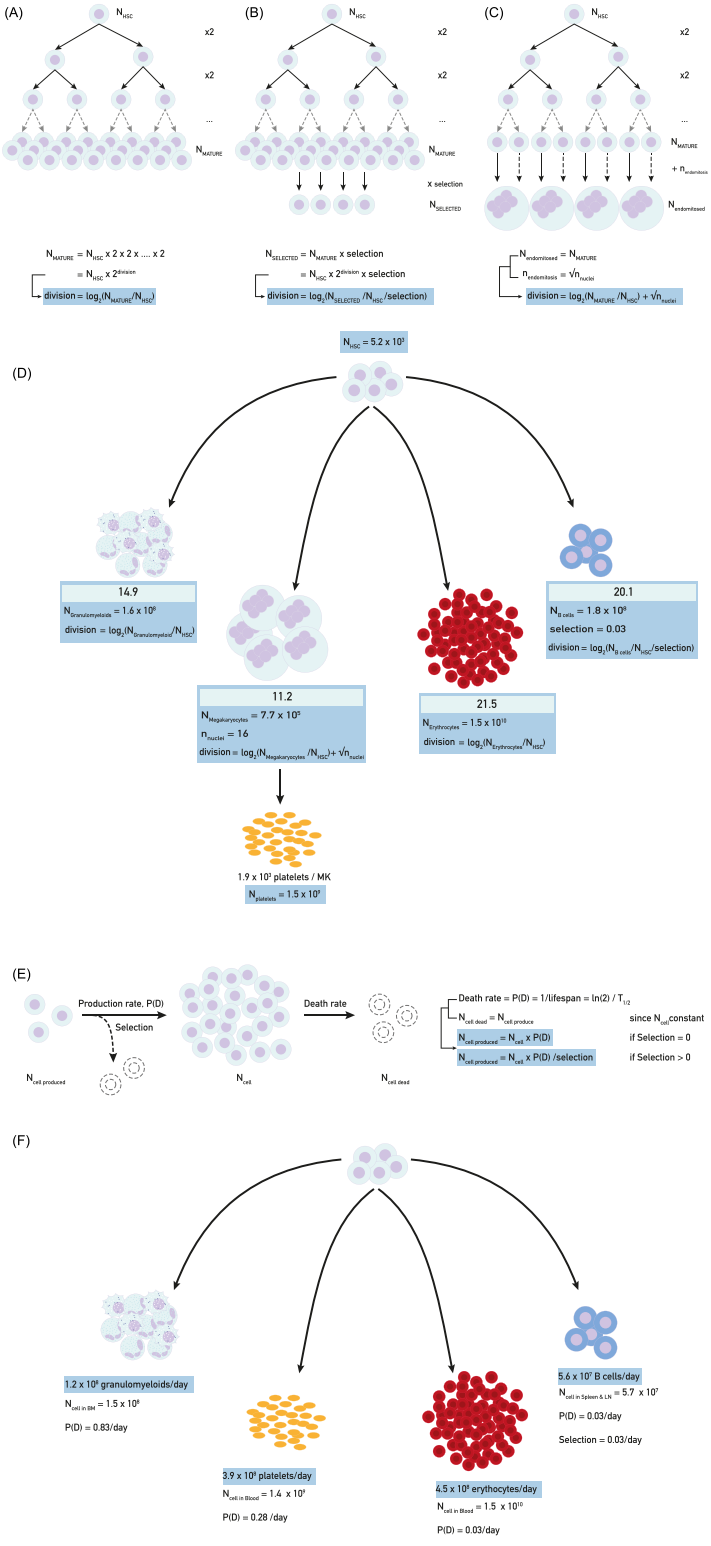
Division rate and division number

Division rates can be measured using **division-linked dilution assays**, or reporter molecules that incorporate into newly synthesized DNA [39]. In mice, HSCs are reported to divide every 145 days [40]. These values are on the same order of magnitude as estimates derived from fate-mapping studies [10,14]. However, the reported proportion of cells entering S-phase per unit time, using a BrdU-EdU sequential labeling technique, suggests a proliferation rate of approximately once every 4 days [41]. These differences in reported values probably arise due to differences in labeling techniques or heterogeneity within HSCs [14,41,42]. In mice, MPPs reportedly cycle faster than do HSCs [14,43] and *in vivo* EdU labeling showed that cycling rates for RPPs were higher than those measured in MPPs [43]. These data are consistent with a linear amplification model in which proliferation rates increase with early maturation[14]. In humans, HSCs have been estimated to divide once every ~280 days, a value inferred by mathematical modeling of X chromosome inactivation patterns in females [44]. However, *in vivo* division rates for human MPPs and RPPs are lacking.

Despite recent progress in our ability to quantify division rates, the number of divisions it takes to differentiate an HSC into a mature cell is unknown. Knowing the size of each cell compartment, it is possible to estimate the minimum number of divisions in this process (Figure 3; Files S1, S2, and Calculation S2a in the supplemental information online). Performing this estimation in mice suggests that at least 15–22 divisions are needed to account for the expansion of 5200 HSCs to 10^8–10^{10} mature cells (Figure 3; Files S1, S2, and Calculation S2a in the supplemental information online), with erythropoiesis requiring approximately seven more divisions compared with myelopoiesis. Considering the fraction of B cells that survive after negative and positive selection, we estimate that a minimum of 20 divisions are needed to produce all mature B cells (Figure 3; Files S1, S2, and Calculation S2b in the supplemental information online). We estimate that megakaryocytes derive from HSCs in seven divisions (Figure 3; Files S1, S2, and Calculation S2c in the supplemental information online), not including **endomitoses**, and produce an average of 519 platelets each day (Files S1, S2, and Calculation S2d in the supplemental information online). Importantly, these estimates consider neither HSC heterogeneity in clonal expansion nor the impact of cell death and differentiation, processes that are discussed in the following text.

Lifespans and turnover rates

A key parameter that regulates cell numbers is the rate of cell death, which can be derived from half-life and lifespan measurements (Table 2). Half-life measures are mathematically inferred by following the loss of a label over time within a cell population. Cells can be labeled *in vivo* with BrdU, EdU, deuterium isotope, or *ex vivo* [45]. Half-lives have not been measured *in vivo* in progenitor populations, because no labeling methods can currently discriminate between death and differentiation. In mature cell types of both humans and mice, results show that most cells are short-lived, on the order of days or weeks (Tables 2 and 3), with some variation within cell types, due to maturation or activation state [45], and tissue localization [46].



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(See figure legend at the bottom of the next page.)

Table 2. Total number and production rate of hematopoietic cells in the reference mouse[a,b]

Cell type	Organ	Cell count	Half-life (days)	Lifespan (days)*	Death rate (/day)	Developmental selection rate (/day)	Cells produced		Refs
							/day**	/year**	
Red blood cells	Blood	1.5E+10	24.3	**35.00**	0.03	–	4.5E+08	1.6E+11	[59]
Platelets	Blood	1.4E+09	2.5	**3.60**	0.28	–	3.9E+08	1.4E+11	[59,60]
Myeloid	Bone marrow	1.5E+08	**0.84**	1.21	0.83	–	1.2E+08	4.5E+10	[46]
B cells (follicular mature)	Spleen and lymph node	5.7E+07	24.3	**35.00**	0.03	**0.03**	5.6E+07	2.0E+10	[47,65]

[a]Based on calculations S1a and Table S1 in the supplemental information online. Half-lives or lifespans were retrieved from the given articles (as indicated in bold) and used to provide an insight of the mature hematopoietic cell population turnover using the following simple relationships: *Lifespan = 1/Death rate = $t_{1/2}$/ln(2) ** cells produced = death rate × time period in days × developmental selection rate × cell count.

While measuring death rates is challenging, estimation of turnover can help to understand the dynamics of hematopoiesis (Calculation S2e in the supplemental information online). In steady-state conditions, the turnover rate of each lineage can be estimated from population sizes and circulating half-lives (Tables 2 and 3), assuming that cell production and death rates balance each other (Figure 3; Files S1, S2, and Calculation S2e in the supplemental information online). In mice, despite different population sizes, erythroid and granulocyte turnover rates are on the same order of magnitude due to large differences in lifespan (35 days [47] versus 1.2 days [46]; Files S1, S2, and Calculation S2e in the supplemental information online). However, murine B cells have a tenfold lower turnover rate (Files S1, S2, and Calculation S2e in the supplemental information online), consistent with the relatively low number of CLPs. Importantly, the steady-state assumption we make is not always valid; different studies report that the number of phenotypic HSCs in mice increases with age [48,49].

In humans, it has been estimated that 3.3×10^{11} cells are produced each day, ~85% of which are hematopoietic [4]. Of these cells, 2.1×10^{11} are erythroid, 6×10^{10} are neutrophils, 1.5×10^{9} are monocytes, and 7×10^{9} are lymphoid (Table 3) [4]. In this study, platelets were not considered a cell type and so were not included in the final analysis. Based on available measurements of total platelet numbers (1.5×10^{12}) as well as lifespan (9.9 days) [50], we estimate a turnover rate of 1.5×10^{11} platelets per day (Table 3). In humans, as in mice, the erythromyeloid lineages dominate cell turnover.

Differentiation rates

Apart from division and death rates, the residency times within, and transition rates between, compartments regulate cell dynamics. Inference from a murine fate-mapping study reports that differentiation rates increase from HSCs to MPPs and RPPs [10], and that an individual MPP is

Figure 3. The dynamics of steady-state murine hematopoiesis. (A) The minimum division numbers between active hematopoietic stem cells (HSCs) and mature cell subsets for red blood cells and myeloid cells. For each cell type, this minimum division number was computed with the logarithmic 2 transformation of total cell numbers in the body of a certain cell type (N_{mature}) divided by the number of active HSCs (N_{hsc}). This calculation does not consider the impact of cell death and differentiation. (B) For B cells, positive and negative selection of mature naïve B cells were considered and for megakaryocytes, endomitosis was also accounted for (C), as well as platelet fragmentation (D). (D) Summary of the values for the reference mice, as described in File S1 in the supplemental information online. (E) The estimated turnover rates of the blood cell lineages, assuming that production and death rates balance in healthy young adult mice. In this scheme, the number of cells produced each day ($N_{cell\ produced}$) is the product of the total cell number (N_{cell}), the production rate, and potential selection (as for B cells) of each cell type per day, noted as $P(D)$. (F) The calculated cell population sizes in given organs, noted as expected $P(D)$ for each blood cell lineage during hematopoiesis for the reference mouse, as in Table 2 in the main text. Details of all these calculations are provided in Files S1 and S2 in the supplemental information online. Abbreviation: LN, lymph node.

Table 3. Total number and production rate of human hematopoietic cells for a reference human male 20–30 years of age, weighing 70 kg and measuring 170 cm in height[a]

Cell type	Estimated cell count	Lifespan (days)	Cells produced per day	Refs
Red blood cells	2.5×10^{13}	116	2.1×10^{11}	[4]
Monocytes	5.0×10^{9}	3.5	1.5×10^{9}	[4]
Platelets	1.5×10^{12}	9.9	1.5×10^{11}	[50]
Neutrophils	6.4×10^{11}	6.6	6×10^{10}	[4]
Mature B cells	3.0×10^{11}	63	5×10^{9}	[4]
Mature T cells	7.0×10^{11}	323	2×10^{9}	[4]

[a]See also Files S1 and S2 in the supplemental information online.

180 times more likely to transition into a CMP than into a CLP. While the confidence intervals around these estimates are large, they suggest that lymphoid commitment is rare. Based on these values of the MPP–CLP transition [10], a single-cell RNA-sequencing data set of 1000 murine MPPs contains only five or six cells that commit to the lymphoid fate, highlighting the need for enrichment strategies to study early lymphopoiesis [51]. However, estimates derived from fate-mapping studies vary depending on technical factors, such as the labeling system, the gating strategy, or the model used in parameter fitting [14]. In humans, there are no reported *in vivo* differentiation rates, although retrospective lineage-tracing methods that make use of endogenous barcodes within the genome (e.g., somatic mutations) are emerging [20].

To summarize, hematopoietic cells are short-lived, and must be constantly regenerated. In both mice and humans, the numbers of myeloid and erythroid cells produced each day are of the same order of magnitude, even though there are more erythroid than myeloid cells when we consider total cell numbers. This is due to large differences in their expected lifespan.

How big are different hematopoietic cell types? What does each cell type comprise?

As hematopoietic cells differentiate, they undergo significant remodeling, changing their shape, size, and macromolecular composition to fulfill specialized functions such as oxygen transport, blood clotting, and pathogen killing.

Cell size can be measured using synthetic beads as an internal control in flow cytometry, or by imaging [52] and results show that cell volumes vary dramatically between cell types (Files S1, S2, and Table S1 in the supplemental information online). For example, murine HSCs, CMPs, GMPs, and MEPs have a volume of 175 μm^3, 210 μm^3, 320 μm^3, and 450 μm^3, respectively [53]. Differences in cell size impact cellular composition, with protein and lipid numbers scaling linearly with cell volume for many mature blood cell types (Files S1, S2, Table S1, and Figure S2 in the supplemental information online). As a consequence, quantitative differences in cell size can have major implications for biosynthesis when we consider the large numbers of cells produced each day (see preceding text). Aside from the quantification of protein synthesis rates in progenitors [54], measurements of the ATP and macromolecular requirements for hematopoietic cell production are lacking. However, newly developed high-sensitivity mass spectrometry-based approaches are emerging to tackle this issue [55]. Using the relative protein content of the different cell types (Files S1, S2, and Table S1 in the supplemental information online) and the number of cells produced in humans per day (Table 3), we computed the amount of protein and ATP required to fuel cell turnover (Files S1, S2, and Calculation S2f in the supplemental information online). This calculation predicts that myelopoiesis requires ~2.8 times more ATP than does erythropoiesis in humans, based on only protein synthesis requirements.

To summarize, each hematopoietic cell type has a distinct size and shape, helping them to fulfill specialized functions. Consequently, the amount of energy and biomaterial it takes to regenerate each cell type may vary significantly, but many parameters have yet to be quantified. Quantitative hematometabolism is a research topic that warrants further focus and may facilitate the development of novel dietary interventions to modulate hematopoiesis.

Concluding remarks

In this article, we summarized key numbers in hematopoiesis, providing a quantitative reference for the field, and highlighting areas in which quantitative information is missing. These numbers can be accessed and updated at the Bionumbers Repository[i]. In reviewing existing numbers, we learn that, in mice, 5.2×10^3 HSCs give rise to 10^{10} hematopoietic cells, while, in humans, 10^4–10^6 HSCs give rise to 10^{13} mature hematopoietic cells. It is striking that, in both humans and mice, most hematopoietic cells are short-lived, on the order of days or weeks and, as such, hematopoiesis accounts for 90% of total blood cell turnover [4]. We have also reviewed what is known about hematopoietic cell size and composition, with numbers revealing that each cell type is likely to have distinct metabolic requirements. For example, we estimate that at least ~36 kcal per day, the equivalent of a single strawberry, is needed to maintain RBC homeostasis in humans (Files S1, S2, Figure S3, and Calculation S2g in the supplemental information online). This value is surprisingly low, given that RBCs account for 90% of all cells in the human body; this value arise from the relatively low protein content and low ATP turnover rate of RBCs compared with other cell types.

We have also illustrated how quantitative approaches may improve our ability to: (i) design experiments, as discussed earlier by the need for novel enrichment strategies to study early lymphopoiesis; (ii) consolidate and contextualize data across studies and species, identifying knowledge gaps (see Outstanding questions); and (iii) make novel predictions about how hematopoiesis is regulated.

In generating reference values, it is important to understand how each measurement has been made. Conflicting values in the literature may arise due to nuanced technical details about sample processing and analysis and may not reflect true biological variation. Unfortunately, many numbers could not be included in this article due to poor reporting with unclear tissue processing and data transformation steps. We suggest that the best practice is to make raw data available for each figure, along with metadata associated with sample processing, data acquisition, and analysis steps.

To summarize, hematopoiesis is a dynamic process, giving rise to diverse cell types with distinct population sizes, tissue localization, turnover rates, and sizes. We hope that this article illustrates how quantitative approaches can improve our understanding of hematopoiesis; we also anticipate that the numbers presented can serve as a reference starting point for the immunology and hematology scientific communities.

Outstanding questions

Can we identify and enumerate the key 'missing cell numbers' in the hematopoietic system that are needed to increase our understanding of hematopoiesis?

What are the total numbers of hematopoietic cells across all major organs, including the gut, skin, liver, lung, and brain?

What are the cell compartment sizes across developmental, adult, and aged hematopoiesis?

What are the rates of cell production, cycling, and death for each developmental compartment and cell type?

To address these quantitative gaps, we suggest that greater emphasis should be placed on performing absolute rather than relative measurements, because the same relative change can result in several orders of magnitude differences in absolute cell counts depending on the size of the cell population.

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Declaration of interests

None declared by authors.

Resources

[i]https://bionumbers.hms.harvard.edu/

Supplemental information

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References

1. Sender, R. *et al.* (2016) Revised estimates for the number of human and bacteria cells in the body. *PLoS Biol.* 14, e1002533
2. Milo, R. and Phillips, R. (2015) *Cell Biology by the Numbers*, Garland Science
3. Grubb, S.C. *et al.* (2009) Mouse Phenome Database. *Nucleic Acids Res.* 37, D720–D730
4. Sender, R. and Milo, R. (2021) The distribution of cellular turnover in the human body. *Nat. Med.* 27, 45–48
5. Lahoz-Beneytez, J. *et al.* (2016) Human neutrophil kinetics: modeling of stable isotope labeling data supports short blood neutrophil half-lives. *Blood* 127, 3431–3438
6. Valentin, J. (2002) Basic anatomical and physiological data for use in radiological protection: reference values: ICRP Publication 89. *Ann. ICRP* 32, 1–277
7. Makarieva, A.M. *et al.* (2008) Mean mass-specific metabolic rates are strikingly similar across life's major domains: evidence for life's metabolic optimum. *Proc. Natl. Acad. Sci. U. S. A.* 105, 16994–16999
8. Trepel, F. (1974) Number and distribution of lymphocytes in man. A critical analysis. *Klin. Wochenschr.* 52, 511–515
9. Qatarneh, S.M. *et al.* (2006) Three-dimensional atlas of lymph node topography based on the visible human data set. *Anat. Rec. B. New Anat.* 289, 98–111
10. Busch, K. *et al.* (2015) Fundamental properties of unperturbed haematopoiesis from stem cells in vivo. *Nature* 518, 542–546
11. Wilson, N.K. *et al.* (2015) Combined single-cell functional and gene expression analysis resolves heterogeneity within stem cell populations. *Cell Stem Cell* 16, 712–724
12. Challen, G.A. *et al.* (2010) Distinct hematopoietic stem cell subtypes are differentially regulated by TGFβ1. *Cell Stem Cell* 6, 265–278
13. Beerman, I. *et al.* (2010) Functionally distinct hematopoietic stem cells modulate hematopoietic lineage potential during aging by a mechanism of clonal expansion. *Proc. Natl. Acad. Sci. U. S. A.* 107, 5465–5470
14. Sawai, C.M. *et al.* (2016) Hematopoietic stem cells are the major source of multilineage hematopoiesis in adult animals. *Immunity* 45, 597–609
15. Pei, W. *et al.* (2020) Resolving fates and single-cell transcriptomes of hematopoietic stem cell clones by PolyloxExpress barcoding. *Cell Stem Cell* 27, 383–395
16. Rodriguez-Fraticelli, A.E. *et al.* (2020) Single-cell lineage tracing unveils a role for TCF15 in haematopoiesis. *Nature* 583, 585–589
17. Bowling, S. *et al.* (2020) An engineered CRISPR-Cas9 mouse line for simultaneous readout of lineage histories and gene expression profiles in single cells. *Cell* 181, 1410–1422
18. Sun, J. *et al.* (2014) Clonal dynamics of native haematopoiesis. *Nature* 514, 322–327
19. Abkowitz, J.L. *et al.* (2002) Evidence that the number of hematopoietic stem cells per animal is conserved in mammals. *Blood* 100, 2665–2667
20. Lee-Six, H. *et al.* (2018) Population dynamics of normal human blood inferred from somatic mutations. *Nature* 561, 473–478
21. Watson, C.J. *et al.* (2020) The evolutionary dynamics and fitness landscape of clonal hematopoiesis. *Science* 367, 1449–1454
22. Biasco, L. *et al.* (2016) In vivo tracking of human hematopoiesis reveals patterns of clonal dynamics during early and steady-state reconstitution phases. *Cell Stem Cell* 19, 107–119
23. Sommerkamp, P. *et al.* (2021) Mouse multipotent progenitor 5 cells are located at the interphase between hematopoietic stem and progenitor cells. *Blood* 137, 3218–3224
24. Morrison, S.J. and Weissman, I.L. (1994) The long-term repopulating subset of hematopoietic stem cells is deterministic and isolatable by phenotype. *Immunity* 1, 661–673
25. Morrison, S.J. *et al.* (1997) Identification of a lineage of multipotent hematopoietic progenitors. *Development* 124, 1929–1939
26. Akashi, K. *et al.* (2000) A clonogenic common myeloid progenitor that gives rise to all myeloid lineages. *Nature* 404, 193–197
27. Nakorn, T.N. *et al.* (2003) Characterization of mouse clonogenic megakaryocyte progenitors. *Proc. Natl. Acad. Sci. U. S. A.* 100, 205–210
28. Harman, B.C. *et al.* (2008) Resolution of unique Sca-1 ^{high} c-Kit $^{-}$ lymphoid-biased progenitors in adult bone marrow. *J. Immunol.* 181, 7514–7524
29. Kumar, R. *et al.* (2008) Lin–Sca1+Kit– bone marrow cells contain early lymphoid-committed precursors that are distinct from common lymphoid progenitors. *J. Immunol.* 181, 7507–7513
30. Giandomenico, S.D. *et al.* (2020) Megakaryocyte TGFβ1 partitions hematopoiesis into immature progenitor/stem cells and maturing precursors. *Blood* 136, 1044–1054
31. Kirby, M.R. and Donahue, R.E. (1993) Rare event sorting of CD34+ hematopoietic cells. *Ann. N. Y. Acad. Sci.* 677, 413–416
32. Hao, Q.-L. *et al.* (1995) A functional comparison of CD34+ CD38– cells in cord blood and bone marrow. *Blood* 86, 3745–3753
33. Farrell, T. *et al.* (2014) Changes in the frequencies of human hematopoietic stem and progenitor cells with age and site. *Exp. Hematol.* 42, 146–154
34. Povsic, T.J. *et al.* (2010) Aging is not associated with bone marrow-resident progenitor cell depletion. *J. Gerontol. A Biol. Sci. Med. Sci.* 65, 1042–1050
35. Boyer, S.W. *et al.* (2019) Clonal and quantitative in vivo assessment of hematopoietic stem cell differentiation reveals strong erythroid potential of multipotent cells. *Stem Cell Rep.* 12, 801–815
36. Niswander, L.M. *et al.* (2014) Improved quantitative analysis of primary bone marrow megakaryocytes utilizing imaging flow cytometry. *Cytometry A* 85, 302–312
37. Basilico, S. and Göttgens, B. (2017) Dysregulation of haematopoietic stem cell regulatory programs in acute myeloid leukaemia. *J. Mol. Med. Berl. Ger.* 95, 719–727
38. Krueger, A. *et al.* (2017) T cell development by the numbers. *Trends Immunol.* 38, 128–139
39. Romar, G.A. *et al.* (2016) Research techniques made simple: techniques to assess cell proliferation. *J. Invest. Dermatol.* 136, e1–e7
40. Wilson, A. *et al.* (2008) Hematopoietic stem cells reversibly switch from dormancy to self-renewal during homeostasis and repair. *Cell* 135, 1118–1129
41. Akinduro, O. *et al.* (2018) Proliferation dynamics of acute myeloid leukaemia and haematopoietic progenitors competing for bone marrow space. *Nat. Commun.* 9, 519
42. Takahashi, M. *et al.* (2021) Reconciling flux experiments for quantitative modeling of normal and malignant hematopoietic stem/progenitor dynamics. *Stem Cell Rep.* 16, 741–753
43. Signer, R.A.J. *et al.* (2016) The rate of protein synthesis in hematopoietic stem cells is limited partly by 4E-BPs. *Genes Dev.* 30, 1698–1703
44. Catlin, S.N. *et al.* (2011) The replication rate of human hematopoietic stem cells in vivo. *Blood* 117, 4460–4466
45. Borghans, J.A.M. *et al.* (2018) Current best estimates for the average lifespans of mouse and human leukocytes: reviewing two decades of deuterium-labeling experiments. *Immunol. Rev.* 285, 233–248
46. Ballesteros, I. *et al.* (2020) Co-option of neutrophil fates by tissue environments. *Cell* 183, 1282–1297
47. Verheijen, M. *et al.* (2020) Fate mapping quantifies the dynamics of B Cell development and activation throughout life. *Cell Rep.* 33, 108376

48. Bernitz, J.M. *et al.* (2016) Hematopoietic stem cells count and remember self-renewal divisions. *Cell* 167, 1296–1309
49. Chambers, S.M. *et al.* (2007) Aging hematopoietic stem cells decline in function and exhibit epigenetic dysregulation. *PLoS Biol.* 5, e201
50. Harker, L.A. and Finch, C.A. (1969) Thrombokinetics in man. *J. Clin. Invest.* 48, 963–974
51. Amann-Zalcenstein, D. *et al.* (2020) A new lymphoid-primed progenitor marked by Dach1 downregulation identified with single cell multi-omics. *Nat. Immunol.* 21, 1574–1584
52. Model, M.A. (2018) Methods for cell volume measurement. *Cytometry A* 93, 281–296
53. Hidalgo San Jose, L. *et al.* (2020) Modest declines in proteome quality impair hematopoietic stem cell self-renewal. *Cell Rep.* 30, 69–80
54. Signer, R.A.J. *et al.* (2014) Haematopoietic stem cells require a highly regulated protein synthesis rate. *Nature* 509, 49–54
55. Agathocleous, M. *et al.* (2017) Ascorbate regulates haematopoietic stem cell function and leukaemogenesis. *Nature* 549, 476–481
56. Bogue, M.A. *et al.* (2020) Mouse Phenome Database: a data repository and analysis suite for curated primary mouse phenotype data. *Nucleic Acids Res.* 48, D716–D723
57. Chervenick, P.A. *et al.* (1968) Quantitative studies of blood and bone marrow neutrophils in normal mice. *Am. J. Phys.* 215, 353–360
58. Sluiter, W. *et al.* (1984) Determination of blood volume in the mouse with 51Chromium-labelled erythrocytes. *J. Immunol. Methods* 73, 221–225
59. Menees, K.B. *et al.* (2021) Sex- and age-dependent alterations of splenic immune cell profile and NK cell phenotypes and function in C57BL/6J mice. *Immun. Ageing* 18, 3
60. Van den Broeck, W. *et al.* (2006) Anatomy and nomenclature of murine lymph nodes: descriptive study and nomenclatory standardization in BALB/cAnNCrl mice. *J. Immunol. Methods* 312, 12–19
61. Kawashima, Y. *et al.* (1964) The lymph system in mice. *Jap. J. Vet. Res.* 12, 69–78
62. Druzd, D. *et al.* (2017) Lymphocyte circadian clocks control lymph node trafficking and adaptive immune responses. *Immunity* 46, 120–132
63. Tanaka, S. *et al.* (2020) Tet2 and Tet3 in B cells are required to repress CD86 and prevent autoimmunity. *Nat. Immunol.* 21, 950–961
64. Hsu, H.-C. *et al.* (2003) Age-related thymic involution in C57BL/6J × DBA/2J recombinant-inbred mice maps to mouse chromosomes 9 and 10. *Genes Immun.* 4, 402–410
65. Allman, D.M. *et al.* (1993) Peripheral B cell maturation. II. Heat-stable antigen(hi) splenic B cells are an immature developmental intermediate in the production of long-lived marrow-derived B cells. *J. Immunol. Baltim. Md 1950* 151, 4431–4444