

Editorial

The cellular immune system comes of age

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In this issue of the *Journal*, Van den Heuvel et al¹ present data on a unique longitudinal study in which almost 1200 children were analyzed for immune cell numbers and phenotypes between birth and 6 years of age. Hierarchical clustering of 62 immune cells revealed that changes in immune cell numbers can be described in 4 distinct patterns and that nongenetic factors could be identified that codetermine immune cell numbers. Importantly but not unexpectedly, infectious disease history (notably herpesviruses) has a strong effect on numbers and phenotypes of CD4⁺ and CD8⁺ T-cell subsets and natural killer cells.²

This type of study is essential to begin to understand the gradual and ordered acquisition of immune competence. Interestingly, van den Heuvel et al¹ show that innate immune cell numbers are high at birth and decrease rapidly in the first months of life. On the other hand, development of the adaptive immune cell system follows distinct waves. Naïve lymphocytes rapidly and transiently accumulate in the first months of life, whereas likely after microbe encounter, cells with features of antigen priming emerge somewhat later, and these populations continue to increase in size. The relative predominance of innate immune cells shortly after birth corroborates the notion that these cells, also from a phylogenetic perspective, form a first line of defense, whereas it takes time for the adaptive lymphoid system to fully mature. Indeed, in the first months of extrauterine life, maternal antibodies might incite critical effector functions, thereby providing (in collaboration with the innate system) a sufficient barrier to many pathogens. Notably, this protection has limitations because, for example, infections with cytomegalovirus can be held in check by an initial natural killer cell response but depend on adaptive T-cell reactions for long-term suppression and clearance.³

The current study is highly controlled for infections with a number of specific pathogens but at the same time reveals an intrinsic problem with performing infection studies in healthy human subjects. In experimental mouse studies timing and dosage of exposure to the (primary) infectious agents are known, and

consequently, protocols can be developed that accurately depict the effect of the particular infection on different aspects of the immune system. In human studies we are faced with a number of problems. Not only can neither the dose nor exact time point of infection be determined, information about coinfections or reinfections are mostly lacking. It is not unlikely that these and a multitude of other parameters have an effect on the composition of the circulating pool of immune cells.⁴ On the other hand, the findings in this study also demonstrate again that some pathogens, such as cytomegalovirus and EBV, leave a very noticeable fingerprint on blood CD4⁺ and CD8⁺ T cells in which infection history is a major determinant for the numbers and differentiation state of peripheral immune cells.^{2,5,6}

The number and phenotype of circulating immune cells depend on multiple parameters, which makes it complicated to directly translate peripheral blood cell numbers to production, recirculation, homeostasis, extravascular homing, persistence, and apoptosis (summarized in Fig 1). After production in the bone marrow and thymus, mature but (where it concerns lymphocytes) mostly naïve immune cells recirculate. Homeostasis of naïve T cells depends in part on influx from *de novo*-generated naïve T cells from the thymus but in human subjects also on homeostatic division of the mature recirculating naïve populations.⁷ Inflammation will change recirculation properties and attract phagocytes directly to sites of infection (eg, neutrophils from the bloodstream). During their usual recirculation, T and B cells will extravasate to secondary lymphoid organs to patrol and check antigen-presenting cells for whether their specific antigenic determinants are present. If so, these naïve cells become activated, expand, and acquire effector functions, after which they are able to invade the infected tissue from which the microbial antigens were derived to start the reaction in the local draining lymphoid organ and deal with the infectious agent itself. After recovery from a primary infection, antigen-specific T cells remain in the system, possibly though the effect of homeostatic cytokines acting on a pool of memory stem cells. Regarding the recirculation time, organ- or tissue-specific dwelling time outside of the bloodstream, usual half-life, or survival of memory lymphocytes and their subsets, not much is known in kinetic terms about the dynamics of this process in human subjects. Finally, it has become evident that a subset of the initially activated pool of T cells takes up permanent residency in solid tissue, such as the gut, skin, and lung. These cells, which form a kind of frontline defense system, are believed to be important in the initial response to reinfecting pathogens.⁸ It is conceivable that this population also slowly builds up at these epithelial surfaces after birth, thereby shaping on-site protection at these barriers. Ideally, not only would one like to be able to gather information about the dynamics of the circulating and tissue-resident populations, but also novel techniques should be developed to assess these parameters in human subjects. Until these data become available, it is difficult to determine how representative the peripheral blood cell count is for the actual number of immune cells in the body.

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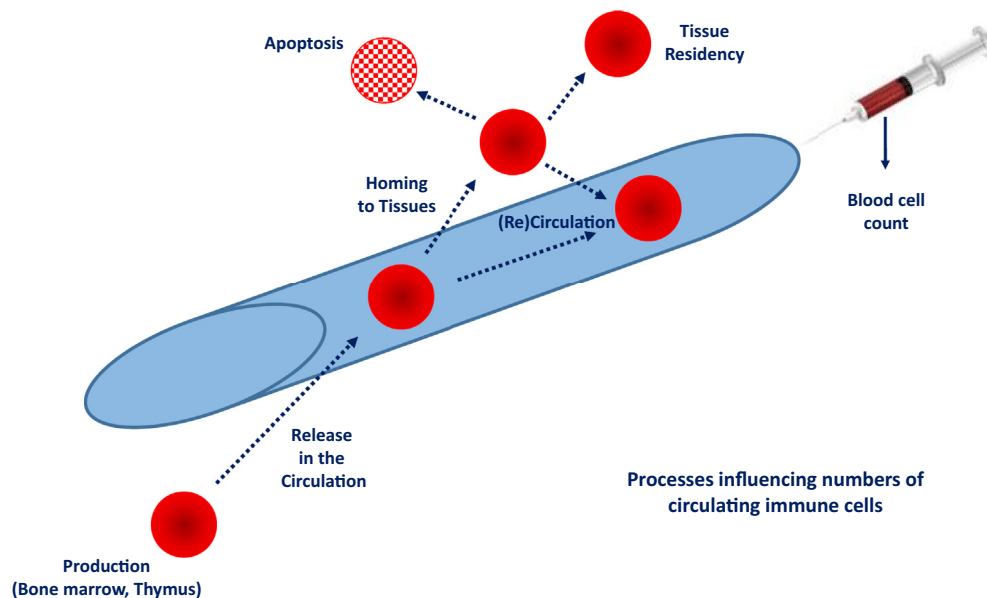


FIG 1. Schematic representation of the different mechanisms that influence peripheral blood immune cell counts.

All of these observations and considerations, together with the findings of Van den Heuvel et al,¹ raise interesting questions. Which rheostats operate to regulate immune cell numbers? For phagocytes, it is known that inflammatory cytokines, such as IL-17, and growth factors, such as granulocyte colony-stimulating factor, will boost the formation of granulocyte production in the bone marrow, leading to an increase in circulating cell counts.⁹ Still, in the absence of overt infection, granulocyte production and numbers in the circulation are kept at a certain steady-state level, and it is not understood how this process is regulated in human subjects. For lymphocytes, the common γ -chain cytokines, such as IL-7 and IL-15, are important homeostatic cytokines for naive and primed cells,¹⁰ but the precise mechanism through which they balance numbers of circulating and (lymphoid tissue) resident cells remains to be established.

It is important not only to understand how the immune system develops at a relatively young age but also to start gathering this type of data in aging populations. To understand, for example, responses to vaccination and susceptibility to infection, analyses for the adaptive immune system should perhaps even take a step to the next level of determining the putative changes in time in the diversity of antigen receptor repertoires because these, better than mere numbers, will determine the immunologic flexibility of the host to respond adequately to pathogens.

Is the article of direct value to those working in clinical immunology? This question is difficult to answer because the mechanisms that operate to control immune cell numbers are complex, interrelated, and not fully understood.¹¹ As the authors mention themselves, “Although we defined whether determinants had a positive or negative association with leukocyte numbers, we were unable to identify the exact nature of the effect, ie, exactly when these effects presented and whether these effects will be transient, persistent or potentially even increasing over time.” Ethnicity and different environmental exposures (eg, parasites, pathogen-associated molecular patterns, and toxins) might be as

influential; we could predict these to be strong drivers, but this is uncertain at the moment. On the other hand, the study indicates that apart from infection, factors other than genetics play a role in or at least correlate with shaping the cellular composition of the blood compartment, such as sex, breast-feeding, and type of infection (where the age at primary infection might be another critical factor to consider the early phase of immune determination), for better or worse.

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