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HEMATOPOIESIS AND STEM CELLS

Comment on Pagliuca et al, page 2781, and Zaimoku et al, page 2799

HLA in AA: innocent bystander or culprit?

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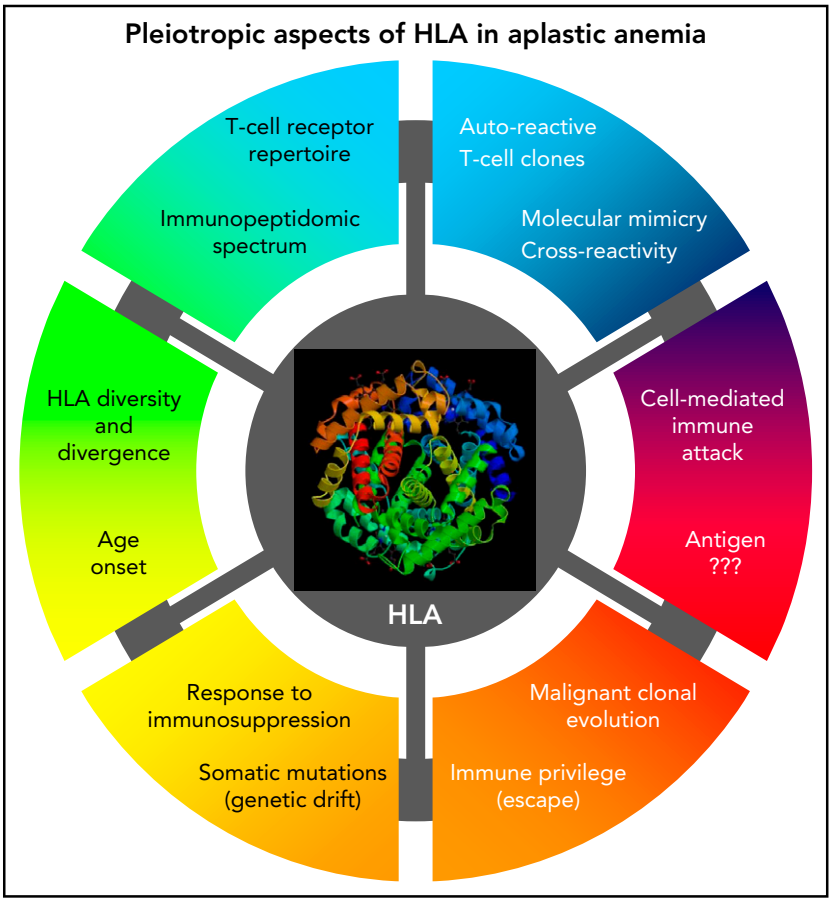
In this issue of *Blood*, 2 papers deal with HLA in patients with acquired idiopathic aplastic anemia (AA). Pagliuca et al[1] show that patients with AA have a reduced structural divergence of homologous HLA alleles, possibly contributing to reduced T-cell receptor repertoire diversity, cross-reactivity, and emergence of autoreactive T-cell clones. In parallel, Zaimoku et al[2] document that somatic mutations in HLA genes leading to functional loss are frequent and correlate with clinical manifestations of AA, such as age onset and risk of clonal evolution.

These 2 independent experimental works highlight that HLA plays a major role in AA pathophysiology and clinical course. Both papers ultimately confirm the well-established immune-mediated pathophysiology of acquired AA.[3] The 2 papers describe different aspects of HLA involvement and are complementary in shaping a fuller story for HLA in AA. The full story will only be known when the key player, the target antigen in AA, is identified. That acquired idiopathic AA is a T cell–mediated disorder is clearly proven by the clinical efficacy of antithymocyte globulin[4] and by a number of experimental observations.[3] In particular, autoreactive T-cell clones have been identified as the key pathogenic effector in patients with AA.[5] Even if the identification of the target antigen(s) remains elusive, it is plausible that these auto-reactive T cells recognize specific epitopes presented on hematopoietic progenitor/stem cells (HSCs) through HLA molecules. Thus, HLA is necessarily involved in the immune-mediated damage of hematopoietic progenitors, either as an innocent bystander or as a culprit. The 2 manuscripts both support a scenario where HLA is not an innocent bystander, but rather, it appears to be a vicious culprit involved during the whole course of the disease.

In their work, Pagliuca et al show that evolutionary divergence in class II alleles is lower in patients with AA. This is associated with a smaller size than predicted of the immunopeptidomic spectrum, thus accounting for decreased T-cell receptor (TCR) diversity. In addition to the evolutionary divergence assessed through metric quantification of the Grantham distance, the reduced diversity of homologous class II HLA alleles is also caused by a higher frequency of homozygosity at these loci. In evolutionary biology, this condition is supposed to confer a disadvantage because of a less efficient T-cell response against tumors and infectious agents. Thus, in humans, the HLA loci have evolved, developing the greatest diversity. Therefore, in individual subjects with reduced HLA divergence, the immune response remains globally efficient, but it may resort to molecular mimicry and cross-reactivity, which may favor autoimmune phenomena. The work of Pagliuca et al is a very elegant attempt to investigate immunogenetic risk in the development of AA, with the caveat of the understanding of the metric quantification of divergence (variations are quite small, with large overlap between patients with AA and a control population). Nevertheless, they could not make further meaningful steps toward the Holy Grail of AA: namely the target antigen. Pagliuca et al exploit an in silico approach trying to identify recurrent amino acid structures in the peptide binding site of HLA and trying to model antigen interaction and subsequent TCR binding. However, even looking at the HSC-specific proteomic reference, they could only find a lower peptide binding capability, without identifying putative target peptides bound by recurrent HLA binding site structures. Finally, high-throughput analysis of the TCR repertoire confirmed that TCR diversity is lower in patients with AA, but it did not correlate with class II HLA evolutionary divergence. However, patients with AA showed increased frequency of T-cell clones harboring TCR CDR3 sequences with known autoreactive specificity, supporting the authors' hypothesis that molecular mimicry and cross-reactivity may result in pathogenic events.

In the other paper, Zaimoku et al point out that HLA molecules do not remain passive during the clinical course of AA, because somatic genetic lesions ranging from locus deletion (ie, 6p loss of heterozygosity) to single nucleotide mutations may affect about half of patients with AA, leading to functional HLA loss (seen as lack of surface expression). The occurrence of these mutations seems consistent with the normal somatic mutation rate of rapidly replicating cells, but their emergence over normally polyclonal hematopoiesis can be caused by a specific immune privilege of HSCs harboring HLA loss (ie, multiple mutant clones are possible), similar to *PIGA*-mutated cells.[6] Nevertheless, in contrast with paroxysmal nosturnal hemoglobinuria (PNH), in the case of HLA loss, mutant clones usually are unable to effectively replace normal hematopoiesis (variant allele frequency is relatively low and tends to decrease after immunosuppressive treatment). Zaimoku et al also demonstrate that HLA loss and the presence of specific HLA alleles frequently found with HLA loss (irrespective of their loss, such as *HLA-B*1402*) are associated with a high risk of clonal evolution. Thus,



The pleiotropic aspects of HLA in AA.

albeit both selected because of immune escape (*PIGA* and *HLA* mutations are always mutually exclusive), PNH and HLA-deficient clones have a different evolution in terms of propensity to transformation into myeloid malignancies.[7,8] An easy explanation is that HLA-mutated clones may escape the mechanism of cancer immune surveillance once they harbor a concomitant oncogenic mutation (as shown in a few cases of monosomy 7). Alternatively, the association between HLA loss and malignant clonal evolution may not imply any causal role of HLA, which would just represent a kind of sentinel gene, helpful to track intrinsic genomic instability of residual HSCs surviving to the immune-mediated attack. In this scenario, we should not forget that in AA, the emergence of clones carrying somatic mutations may also be simply a consequence of genetic drift (ie, neutral mutations becoming detectable because of the extreme oligoclonality).[6] Indeed, a recent phase 3 trial recently documented that somatic mutations in so-called "myeloid cancer genes" (which do not include HLA) can be detected in about one-third of patients with AA at diagnosis, and then in two-thirds of patients at 6 months after treatment, without leading to obvious transformation to myeloid malignancy.[9]

Dr. Young and Dr. Maciejewski have driven the understanding of AA for about 4 decades,[10] and in this issue of *Blood*, their research teams dissect the pleiotropic roles of HLA in AA (see figure). Hopefully merging high-dimensional data (which often are not easy to read) with functional data may help to continue this tremendous journey with the identification of target antigens in AA.

Conflict-of interest disclosure: The author declares no competing financial interests. ■

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