Playing musical chairs with bone marrow transplantation to eliminate leukemia stem cells

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Increasing attention has been focused on the interactions between leukemia cells and their bone marrow (BM) microenvironment. We have recently shown that leukemic stem cells (LSCs) share common BM “niches” with their healthy counterparts. As a result of these shared niche requirements, human LSCs can be mobilized using existing pharmacological agents that facilitate competitive healthy reconstitution, leading to eradication of leukemia during BM transplantation.

Acute myeloid leukemia (AML) is an aggressive hematological neoplasm characterized by the rapid growth of abnormal blood cells, for which no novel therapies have been clinically adopted in decades.1 In current therapeutic practice, risk stratification and treatment decisions are mainly guided by cell-intrinsic cytogenetic and molecular profiling of leukemic blasts. However, few of the recognized aberrations represent easily druggable targets and more than 50% of AML cases do not possess cytogenetic abnormalities.1 More importantly, non-genetic features may be of greater therapeutic significance, as AML has been described to exhibit profound functional heterogeneity within genetical identical cells of the same patient. The majority of leukemic blasts are believed to be short-lived descendants of rare, self-renewing leukemic stem cells (LSCs) that perpetuate long-term disease progression and initiate relapse following conventional treatment.2,3 Because it is unclear to what extent the molecular basis of LSC self-renewal differs from that of healthy hematopoietic stem and progenitor cells (HSCs), selective LSC elimination has remained a challenge.

Previous work by our group has emphasized the cell-extrinsic basis of human HSC self-renewal regulation through in vivo functional assays and high resolution in situ localization analysis of both human biopsies and human xenografted tissue.4 This work clarified that the preservation of self-renewing HSCs with long-term repopulation potential is dependent upon their residence within specialized trabecular bone marrow (BM) regions, particularly in close association with the endosteum versus other cell types in the BM space. We have extended these analytical techniques to evaluate leukemia-niche dynamics toward the critical goal of determining whether LSCs possess distinct microanatomical positions and/or unique external influences.5 We found that phenotypically primitive leukemic cells recapitulated the non-uniform BM distribution previously observed in the context of healthy hematopoiesis, with their preferential detection within the same trabecular endosteal sites as healthy human HSCs. We developed a unique competitive human xenotransplantation model that allowed functional testing of this apparent physical overlap between HSC and LSC niches by concurrently tracking cells from healthy HSC donors versus AML patients within the same recipient mice. Remarkably, increasing numbers of co-transplanted HSCs were able to provide sufficient competitive pressure to undermine the BM colonization and long-term persistence of leukemic cells. This finding suggests that AML-LSCs are in fact narrowly restricted to HSC-specific niches (Fig. 1A) and cannot improvise by relocating to alternative BM sites. This contrasts with the plastic and

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adaptable qualities often thought to typify cancerous cells and presents a novel vulnerability for LSC targeting.

To further exploit the therapeutic value of these observations, we confirmed that, like healthy HSCs, BM-resident LSCs are sensitive to niche dislodgement using mobilization agents such as granulocyte-colony stimulating factors that are already used in human patients to mobilize HSCs. We then paired LSC mobilization with carefully timed HSC transplantation (HSCT), ultimately orchestrating a cellular “musical chairs” scenario in vivo that operated at the expense of LSC niche retention and self-renewal (Fig. 1B). Importantly, the effectiveness of this therapeutic technique was independent of any differential phenotypic or functional qualities between HSCs and LSCs; instead, critical competitive repopulation factors were the relative ratio of healthy versus malignant repopulating cells and precise temporal coordination. In fact, leukemic cells could reciprocally displace healthy HSCs if the sequence of events was reversed. The integrity of the BM microenvironment also proved to be a significant determinant of transplantation success following mobilization conditioning, as HSCs failed to reclaim BM niche territory if leukemic dissemination had already saturated the BM space prior to therapeutic intervention. Such insights will be essential for the development of appropriate protocols and selection criteria for novel reduced-intensity transplantation procedures. This strategic approach could benefit routine HSCT efforts and might be particularly suitable for elderly AML patients or those with poor prognosis, who typically suffer high levels of treatment-related mortality that currently limits their eligibility for transplantation.6

In addition to new therapeutic prospects, the functional and spatial similarities observed between HSCs and LSCs advocate for adjusted perspectives of our abilities to reliably distinguish these cell types. The conventional practice of evaluating HSC and LSCs within independent recipient hosts overlooks any differential feedback signals from normal versus leukemic cellular progeny,7 which would potentially form divergent BM niches over time. Differences between HSCs and LSCs may therefore become less apparent when all aspects of the microenvironment are kept constant. For example, not only do LSCs exhibit impaired differentiation during disease conditions, but the multilignage maturation of healthy murine HSCs also becomes suppressed when exposed to leukemic microenvironments in vivo.8 However, with few exceptions,9 molecular analyses typically compare primitive leukemic cells to matched healthy populations acquired from disease-free individuals, which may not provide an accurate representation of HSC states within leukemia-infiltrated BM. In AML patients, HSC molecular signatures would predictably become altered in reflection of their perturbed environment, and may more closely resemble those of LSCs than currently anticipated. Therefore, rather than focusing on selective molecular stem cell targets, it may be more practical to pursue pharmacological mechanisms to correct the corrupted niches that discourage HSC and LSC differentiation alike.

Our findings complement recent murine genetic studies,10 collectively suggesting that AML-LSCs rely on HSC-like niches and share common extrinsic influences. Importantly, our functional studies have demonstrated that LSC–niche associations are dissociable, providing a promising axis to benefit cell-based therapies. This refined understanding of LSC niche requirements reinforces the need to more broadly apply innovative transplantation models in preclinical settings and to prioritize the development of a novel class of niche-centric pharmacological therapies.

Disclosure of Potential Conflicts of Interest
No potential conflicts of interest were disclosed.

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