The doctor prescribed fat-free diet for stem cell mobilization

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Mobilized hematopoietic stem and progenitor cells (HSPCs) are widely utilized for clinical stem cell transplantation. HSPCs mobilization by the cytokine G-CSF is clinically used for several decades, however, the underlying mechanisms and factors leading to big variations in mobilization yields in healthy donors, are poorly understood. In this issue of Haematologica, Suzuki and colleagues shed light on the effect of dietary fat content on G-CSF-mobilization deciphering the regulatory role of ω3-polyunsaturated fatty acids (PUFA) processed by bone marrow (BM) neutrophils (and to a lower extent by other cell types) as part of the mobilization process in mice. The authors show that G-CSF-mediated activation of PPARδ signaling requires first cues from the sympathetic nervous system via β1/2-Adrenergic Receptors in BM neutrophils, which in turn increases PPARδ expression and activity. However, PPARδ is a negative regulator of HSPC mobilization. A shrewd approach to bypass this negative regulation was to feed mice short term with fat-free-diet. Owing to the low ω3-PUFA content in this diet, the lack of ω3-PUFA/PPARδ activation decreased transcription of the negative regulator angiopoietin-like protein 4 (angptl4), which in turn increased BM vascular permeability and facilitated enhanced HSPC mobilization. This simple albeit novel approach could be easily assessed in order to address the problem of poor clinical HSPC mobilization in some healthy donors.

However, BM neutrophils are not the only players in the complex multifaceted process of HSPC mobilization. Hence, the intriguing study by Suzuki et al, in addition to its novelty regarding the machinery activated in BM neutrophils during HSPC mobilization, opens new research directions, as to HSPC cell-intrinsic signaling.

Signals driving HSPC retention in the BM versus their egress to the blood are tightly balanced during steady state homeostasis in order to facilitate blood and immune-cell production on demand along with preservation of the undifferentiated HSPC BM reservoir. Thus, physiologic HSPC egress to the blood is dynamically modulated by homeostatic light/dark cycles and circadian rhythms involving β1/2-Adrenergic Receptor signaling as well as BM blood vessel permeability and hormone/cytokine secretion. These signals in mice balance the daily rhythms of BM HSPC differentiation and egress during daylight to replenish the blood. While melatonin secretion at night reduces BM vessel-permeability and egress, exerting anti-inflammatory conditions, which reprogram
stem cell self-renewal. Pro-inflammatory signals enforced by bacterial-mimicking LPS challenge and by G-CSF-clinical treatment in mice, modulate this balance skewing it towards differentiation and mobilization, to address the urgent need for immune-competent cells. The negative-regulatory function of angptl4 seems to be part of the balance machinery addressing the need for BM HSPCs preservation from exhaustion and hematopoietic failure. G-SCF is known to evoke pro-inflammatory stimuli in the BM which involve signals from the nervous system, that exert dramatic changes in myeloid cells, osteolineage cells, bone metabolism, and blood vessel permeability. HSPCs are also responding to G-CSF-induced signals showing robust metabolic changes, which prepare them for the dynamic state, essential for taking the active journey to the blood. An interesting question is, can HSPCs “sense” changes in BM lipid mediators during daily light/dark cycles, and moreover, following G-SCF stimuli? Furthermore, what signaling do these mediators induce in HSPC and particularly, do they involve changes in ROS levels, as well as in angptl4 expression and activity? Some hints for this notion come from several reports. HSPCs bear the lipolytic machinery (phospholipase C-β2) to control pharmacological (G-CSF and AMD3100) HSPC mobilization. Angiopoietin-like proteins play multiple roles in the regulation of hematopoietic stem cell activity including quiescence, expansion, self-renewal, and homing. A major candidate for such future studies could be angptl4. In the human settings, angptl4 maintains in vivo repopulation capacity of CD34+ cord blood HSPC. In mice, the PML-PPARδ-FAO pathway influences ROS generation and stem cell division. Depletion of PPARδ that serves as a fatty acids nutrient sensor, reduced stem cell quiescence, and their repopulation potential since it controls asymmetric divisions that are essential for HSC maintenance. Interestingly, angptl4 is upregulated in the BM under inflammatory conditions induced by bacterial LPS challenges, leading to increased secretion of G-CSF and angptl4 from BM stromal cells, which also expand BM myeloid progenitors. Thus, Angptl4 in HSPC balances their response to pro-inflammatory effects in order to preserve their BM maintenance and long-term function. Suzuki et al suggest that temporal attenuation of angptl4 upregulation may further increase the efficiency of G-CSF-induced mobilization (Figure 1).

Another physiologic life condition is aging, which is associated with stress and pro-inflammatory cues, an increase in marrow vascular permeability, adipocytes, and a decrease in hematopoietic cellularity. Adipocytes are accumulated in the marrow during obesity and aging, and notably also following high-fat diet in mice. This change in the ratio of adipocytes/hematopoietic cells reprograms mesenchymal stem cells towards adipogenic rather than osteogenic differentiation, which reduces the rates of bone regeneration and hematopoiesis recovery. In addition to pro-inflammatory signals, G-SCF induces in human individuals also a pro-coagulative state and increased thrombin activity. Moreover, the efficiency of G-CSF-induced mobilization in healthy donors for clinical HSPC transplantation can be predicted by the surface expression levels of the major coagulation- and inflammation-related thrombin receptor, PAR1 on mature
peripheral blood leukocytes and CD34+ HSPC before mobilization is conducted. Importantly, this surface PAR1 expression also predicts HSPC repopulation potential in transplanted patients and PAR1 signaling in mice is essential for steady state egress and for directional in vitro migration of HSPC to a gradient of the major stem cell chemokine CXCL12. It would be of high interest for future studies to elucidate a potential cross-talk between these two axes: the coagulation and inflammation-related thrombin/PAR1/nitric oxide axis and ω3-PUFA/PPARδ/Angptl4 signaling for the sake of improving G-CSF-induced mobilization.

Taken together, the manuscript by Suzuki et al provides important insights as to the signaling pathways activated in BM neutrophils by G-CSF stimuli, and the cross-talk with the lipid content in the BM as a major driving force for the intensity of HSPC mobilization from the BM to the blood.

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Figure 1: Fatty acid content of the diet, affects G-CSF-induced mobilization of HSPC. In addition to signaling in BM neutrophils as reported by Suzuki et al\(^1\), fatty acid content of the diet may affect G-CSF-induced mobilization also via cell-intrinsic signaling in HSPC. A. G-CSF-induced mobilization under normal diet provides fatty acids including the key \(\omega_3\)-PUFA that activates PPAR\(\delta\)/Angptl4 signaling in order to balance and maintain BM HSPC despite the pro-inflammatory cues. B. G-CSF-induced mobilization under fat-free diet deprived of the key \(\omega_3\)-PUFA, prevents the activation of PPAR\(\delta\)/Angptl4 inhibitory signaling that yields higher rates of HSPC mobilization.
