To be or not to be a stem cell: dissection of cellular and molecular components of haematopoietic stem cell niches

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Recent studies have identified multiple cell types that regulate haematopoietic stem cells (HSCs); however, proof that a specific cell type produces a specific factor important for HSC function and maintenance is largely lacking. Ding *et al* (2012) reported recently that conditional deletion of stem cell factor (SCF) in *Leptin receptor* (*Lepr*) expressing perivascular cells or endothelial and haematopoietic cells resulted in significant reductions in number but less profound reduction in function of HSCs. Although the long-term fate of HSCs in these models is largely unexplored and an underlying mechanism for reduction in HSCs not yet reported, these findings further implicate the vascular niche in the functional maintenance of HSCs *in vivo* and also raise intriguing questions for future studies in this field.

The haematopoietic stem cell (HSC) niche has traditionally been considered a discrete site within the bone marrow; however, recent studies have shown that numerous cell types are critically important for HSC regulation and maintenance (Wang and Wagers, 2011). Imaging studies have shown that phenotypic HSCs can be found adjacent to osteoblasts or osteoprogenitor cells on the inner surface of trabecular bone, and genetic studies have further shown that expansion of trabecular bone, leads to expansion of HSCs (Calvi *et al*, 2003; Zhang *et al*, 2003; Lo Celso *et al*, 2009; Xie *et al*, 2009). Other studies have found that phenotypic HSCs

reside in both osteoblastic and vascular niches. The vascular niche is juxtaposed with the osteoblastic niche and includes endothelial cells, CAR cells, Nestin⁺ cells, Lepr⁺ perivascular cells and other cell types. Ding *et al* show that SCF is predominantly provided by endothelial and perivascular cells. (B) Cell-specific deletion of *Scf* in endothelial and Lepr⁺ cells results in reduced HSCs; however, other HSCs are maintained, possibly from a quiescent reserved population that is less dependent on SCF, providing significant levels of haematopoiesis.

Figure 1 (A) HSCs reside in both osteoblastic and vascular niches. The vascular niche is juxtaposed with the osteoblastic niche and includes endothelial cells, CAR cells, Nestin⁺ cells, Lepr⁺ perivascular cells and other cell types. Ding *et al* show that SCF is predominantly provided by endothelial and perivascular cells. (B) Cell-specific deletion of *Scf* in endothelial and Lepr⁺ cells results in reduced HSCs; however, other HSCs are maintained, possibly from a quiescent reserved population that is less dependent on SCF, providing significant levels of haematopoiesis.

are frequently localized to the central marrow, specifically near endothelial or perivascular cells (Kiel *et al*, 2005). Recently, endothelial cells have been shown to support the *ex vivo* expansion of HSCs (Butler *et al*, 2010); however, it was so far not known whether endothelial or perivascular cells functionally maintain *in vivo* HSCs.

Ding and colleagues used knockin reporter mice for *Scf* expression and found that stem cell factor (SCF) was produced predominantly by endothelial and perivascular cells but was not concentrated near the bone surface. To investigate which cellular sources of SCF are important for HSC maintenance, they conditionally deleted *Scf* specifically in haematopoietic cells, osteoblasts and *Nestin-Cre* expressing cells but found no significant effects on HSC maintenance. In contrast, conditional deletion in both haematopoietic and endothelial cells or in *Leptin receptor (Lepr)* expressing perivascular stromal cells significantly reduced phenotypic and, to a lesser extent, functional HSC frequency—thus further demonstrating that the vascular niche plays a role in functionally supporting HSCs (Figure 1). These findings underscore the complexity of the HSC niche and raise crucial future questions.

The nature and specific identity of *Lepr* expressing cells is uncertain. This population appears to partially overlap with *Nestin-Cre* expressing cells, and it is not clear to what extent *Lepr* expressing cells might identify with *Cxcl12*-abundant reticular (CAR) cells, both of which have been previously identified as HSC niche components (Sugiyama *et al*, 2006; Mendez-Ferrer *et al*, 2010). Although phenotypic HSC frequency (determined by the cell-surface markers lineage⁺, Sca-1⁺, Kit⁺, CD150⁺, CD48⁺) is dramatically reduced, functional HSC frequency is only mildly compromised following conditional deletion of *Scf* either ubiquitously or in endothelial/perivascular cells. This indicates that other factors or sources of SCF maintain substantial numbers of HSCs independent of SCF produced by the vascular niche or

elsewhere. Indeed, these results may be consistent with the coexistence of quiescent and active HSC populations—with the quiescent, reserved population serving as a backup HSC source to support life-long haematopoiesis, especially following the loss of active HSCs in response to stress (Li and Clevers, 2010). Considering the role of SCF in promoting proliferation (Broudy, 1997), it would be interesting to know the long-term effects of cell-specific deletion of *Scf* on HSC maintenance. Cell-specific deletion in *Nestin-Cre* expressing cells apparently did not affect HSC frequency long-term (5 months); however, such long-term data were not presented for osteoblast-specific knockout of *Scf*. It would also be interesting to know the mechanism for HSC loss following endothelial/perivascular-specific deletion of *Scf*. Interesting topics to address in the future are whether HSC quiescence is compromised, or whether apoptosis or differentiation is increased.

As the authors note, multiple cell types are involved in HSC maintenance. Given the juxtaposition of endothelial and perivascular cells with the bone surface, the osteoblastic and vascular niche represent not always mutually exclusive entities (Lo Celso *et al*, 2009). We have recently proposed that stem cells may reside in special zones, where active stem cells may provide for the daily replenishment of tissues while quiescent, reserved stem cells serve as a backup sub-population to ensure life-long tissue maintenance and replenishment of the stem cell pool following stress (Li and Clevers, 2010). It remains for future studies to continue to determine which specific niche cells produce which particular factors for maintaining long-term quiescence versus those for supporting proliferation and survival of stem cells. The results published by Ding *et al* present a significant step towards this goal.

Conflict of interest

The authors declare that they have no conflict of interest.

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