A novel growth factor encoded by an old gene

Imagine you are asked to complete a jigsaw puzzle with an unknown number of pieces. You start off from scratch putting pieces together that fit, but you'll never know when you're done.

In this issue of the journal (see pages 157) Xu and colleagues present interesting data on the activity of a protein, which they refer to as hemangiopoietin (HAPO), on early human hematopoietic progenitor cells. This study, together with a recent publication by the same group in which the activity of HAPO was tested *in vivo* in mice,[1] convincingly demonstrates that HAPO is a potent stimulator of both hematopoietic and progenitor endothelial cells. As our understanding of proteins that regulate blood cell production is still limited, the identification of any molecule with proven hematopoietic activity is always welcome; it adds another piece to the puzzle.

However, there are several reasons why HAPO is a special case. First of all, the gene that encodes HAPO has been well known for over 10 years. Different groups of researchers, working in unrelated fields, stumbled at some time during their studies on this gene, which is therefore also known by a variety of other abbreviations, such as *CACP*, *MSF*, *SZP* and *PRG4*.

For example, it was shown that patients with camptodactyly-arthropathy-coxa vara-pericarditis (CACP) syndrome, an autosomal recessive condition characterized by various clinical symptoms that all relate to problematic joint or intimal cell lubrication, carry a mutation in this gene, which was therefore also called articular cartilage superficial zone protein (*SZP*) gene.[2,3]

The gene that encodes HAPO is also known as proteoglycan 4 (*PRG4*), as it is actually part of a larger family of so-called proteoglycans, which form an important constitutent of the extracellular matrix.[4] PRG4 is a multifunctional proteoglycan with potential growth-promoting, cytoprotective, and lubricating properties in cartilage metabolism.[3,5] One previous publication reported on the megakaryocyte-stimulating activity of a protein encoded by this gene, which was therefore referred to as megakaryocyte stimulating factor (*MSF*) gene.[6]

CACP/SZP/PRG4/MSF/HAPO is expressed in many different tissues, which explains why mutated alleles of this gene can lead to a plethora of seemingly unrelated clinical features. So, what's new ? Xu and colleagues purified a protein from the urine of patients with aplastic anemia that stimulated proliferation of both human hematopoietic progenitor cells and endothelial cells, hence the name hemangiopoietin. After multiple purification steps they were able to use protein sequencing to identify the exact nature of this molecule. It turned out that hemangiopoietin is encoded by the *CACP/SZP/PRG4/MSF* gene locus. However, whereas the full PRG4 protein is translated from a mRNA molecule that contains all 12 exons of the gene, HAPO is an alternatively spliced isoform and consists of only exons 2, 3, 4 and 6. This is yet another example of a single gene encoding multiple proteins, with apparently highly variable functions. It is this small soluble protein, which is detectable in relatively high levels in normal serum, that has interesting hemangiopoietic activity.

Over the last decade it has become increasingly clear that close developmental ties exist between the blood-forming tissue and the vascular system. Cells have been identified that have the potential to give rise to both hematopoietic and endothelial lineages.[7] These hemangioblasts do not only exist in the embryo, at a time of development when blood cells and endothelial cells first emerge, but have also been found in adult tissue.[8] Whereas the number of growth factors that act on hematopoietic stem or progenitor cells is large, growth factors that act on these hemangioblasts have remained elusive. The data presented by Xu *et al.* suggest that HAPO may be precisely one such factor.

In an extensive set of experiments presented in both papers the authors show that HAPO has *in vitro* stimulatory activity on immature human hematopoietic, but also primitive endothelial, progenitor cells when added as a soluble molecule to colony assays. In addition, they show that stromal cells that are engineered to overexpress HAPO have enhanced ability to support human hematopoiesis *in vitro*. Finally, when HAPO was injected into irradiated mice, enhanced survival was documented.

As with any good experimental study, many questions remain. First, it will be important to establish the phenotype of the HAPO-responsive cells. As the studies by Xu *et al*. were carried out with populations of cells, it cannot be ruled out that only a small subset of the purified cells (the hemangioblast?) is responsible for the *in vitro* effect. Secondly, there is no information on the molecular mechanism by which HAPO exerts its activity: for which molecule (receptor?) on stem/progenitor cells does it show affinity? The identification of the HAPO receptor could potentially be an important hemangioblast marker.

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