GENE THERAPY

Promising delivery

Much effort has gone into developing efficient gene therapy protocols for sickle cell disease (SCD) because of its impact, especially in Africa, and because SCD is caused by a mutation in a single gene, making it a suitable candidate for gene therapy. Until now, the results have been disappointing, but in a recent issue of *Science*, Pawliuk *et al.* report success in correcting SCD in mice by delivering a variant of β-globin to the haematopoietic cell lineages using an improved viral vector.

SCD is caused by a single amino-acid substitution in β-globin, which leads to the formation of an abnormal form of haemoglobin, known as HbS. Polymerization of HbS into long fibres causes red blood cells (RBCs) to adopt a characteristic sickle shape, and makes them rigid and adhesive, which leads to circulation blocks and organ damage. Although wild-type β-globin can prevent HbS polymerization when present in high concentrations, γ-globin — another type of globin chain — is an even more efficient antipolymerizing agent. Therefore, Pawliuk *et al.* engineered a modified β-globin, into which they introduced a γ-globin-specific amino acid, which is believed to be responsible for its antipolymerizing properties.

In the past, progress in SCD gene therapy was hampered by low expression of the globin constructs and inefficient gene transfer to haematopoietic stem cells. To overcome these problems, the authors included in their viral vector the β-globin locus control region (LCR), a large regulatory region that enhances gene expression and makes it specific to the haematopoietic lineage. But to ensure efficient chromosomal integration of such a large insert, they used a vector derived from HIV with greater gene transfer capability. They also included additional DNA transport sequences from HIV and optimized the LCR sequences for transfer and expression by a method called recombinase-mediated cassette exchange.

The finished construct was tested in two different transgenic mouse models of SCD. Bone marrow of these SCD mice was transduced with the modified β-globin and was then transplanted into normal mice, whose own haematopoietic stem cells had been destroyed by irradiation. Negative control mice had up to 80% sickling in their RBCs, but those that received the modified β-globin-containing bone marrow had only 26% sickling in RBCs. Important confirmation of the therapeutic potential of this construct came when Pawliuk *et al.* found no evidence of SCD-associated pathology in the mice that had been transduced with the modified β-globin.

The results from Pawliuk *et al.* offer great hope for effective gene therapy for SCD. Although the safety of HIV-based viral vectors needs to be confirmed, and efficient ways of removing diseased stem cells from the patient need to be developed, the authors are optimistic that clinical trials might be feasible in a few years' time.

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References and links

ORIGINAL RESEARCH PAPER Pawliuk, R. *et al.* Correction of sickle cell disease in transgenic mouse models by gene therapy. *Science* **294**, 2368–2371 (2001)