

SMaRT Technology Enables Gene Expression Repair in Skin Gene Therapy

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In this issue, Wally *et al.* (2008) report successful gene expression repair by spliceosome-mediated RNA *trans*-splicing (SMaRT), a novel achievement in molecular medicine. In their model, SMaRT was able to replace a mutation of the plectin gene in epidermolysis bullosa simplex with muscular dystrophy. This approach is particularly attractive for skin gene therapy of dominant-negative mutations present in a number of blistering genodermatoses.

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Wally *et al.* (this issue, 2008) describe successful execution of gene expression repair, a novel achievement in molecular medicine. The authors have used spliceosome-mediated RNA *trans*-splicing (SMaRT), which is able to replace certain mRNA exonic segments of defective genes. This approach is particularly attractive for the correction of dominant-negative mutations present in a number of blistering genodermatoses (Uitto and Richard, 2004). This technology will restore not only the expression of the corrected mRNA from the dominant-negative gene but also the production of the corresponding protein. Although *trans*-splicing has been successfully used for the correction of the cystic fibrosis transmembrane receptor and the β -globin gene (Liu *et al.*, 2002; Chao *et al.*, 2003), it has not been pioneered for correcting dominant-negative mutations of genodermatoses such as the plectin gene in epidermolysis bullosa simplex with muscular dystrophy.

Advantages of this technology include the possibility of utilizing small corrective RNA sequences that target exonic sequences within the mutated gene with high specificity as well as the natural regulation of gene expression. Consequently, these small corrective

RNA sequences may be efficiently packaged in retroviral or lentiviral vectors that have a limited capacity to carry large genes, such as the plectin gene that codes for a 14.2-kDa RNA. In this study, retroviral gene transfer has yielded increasing amounts of correct plectin protein. Several private mutations contained in hot-spot areas can be targeted with a few pre *trans*-splicing RNAs, thus making SMaRT technology applicable to a larger number of patients.

Repairing dominant-negative mutations of genodermatoses

Limitations of the SMaRT technology are its relatively low efficiency and the optimal design of the pre *trans*-splicing molecules that must specifically recombine with target intronic pre RNA, e.g., using functional splice sites (Mansfield *et al.*, 2003).

Short interfering RNA has previously been used to knock out the mutated gene/gene product that exhibited off-target effects and was found to silence expression from the normal allele, which

counteracts normal protein expression in dominant-negative diseases.

The new technology must compete with other molecular therapies such as gene replacement or supplementation therapy in which, for example, *Sleeping Beauty* transposons or Φ C31 bacterial phage integrases were used to integrate wild-type gene copies into the genome (Ortiz-Urda *et al.*, 2003a,b). Alternatively, zinc finger proteins have recently been used for true gene correction and were found to restore normal gene expression (Urnov *et al.*, 2005).

Last, although the achieved progress is greatly appreciated—and necessary if skin gene therapy is to be kept alive—some obstacles to successful gene therapy remain unresolved. The first is the random insertion of retroviral vectors that has corrected the life-threatening severe combined immune deficiency syndrome (“bubble babies”) in more than 70% of treated individuals throughout the world (Hacein-Bey-Abina *et al.*, 2003) but has also caused several cases of leukemia when vectors inserted into cellular oncogenes (e.g., *LMO2* or *evi-1*). Second, the inactivation of gene expression following retroviral or lentiviral gene transfer limits the longevity of the desired effects to several months. Ultimately, the use of suitable animal models will reveal the full benefit of this new technology (Arin and Roop, 2004).

CONFLICT OF INTEREST

The author states no conflict of interest.

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COMMENTARY

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