Replication-defective adenoviruses have become popular gene therapy vectors because of their ability to efficiently transduce most cells, resulting in a few weeks or months of episomal expression, terminated by (*a*) immune attack on host cells expressing viral antigens, (*b*) DNAses, and/or (*c*) methylation (1, 2). In this issue of *The Journal,* carefully controlled experiments by Feldman et al. (3) demonstrate that high titers of the Ad 5 vector (lacking E1A and part of E1B and E3, and driven by the Rous sarcoma virus long terminal repeat) transduced 0.2% of smooth muscle cells (SMC) in the balloon-injured iliac artery of cholesterol-fed rabbits, vs 2% in balloon-injured normal arteries. Potential mechanisms for this difference include (*a*) immune destruction of virions or host cells by plaque macrophages and T cells (4); (*b*) trapping of virions in the abundant proteoglycans and collagens of the plaque (5); (*c*) down-regulation of receptors required for viral binding (unknown in the case of Ad 5, but most viruses attach to both a protein receptor and a glycoprotein or proteoglycan (see reference 6), all of which are differentially expressed in atherosclerosis (4, 5); (*d*) competition by fibronectin and fibrin(ogen)—abundant in atherosclerosis—(4, 5) with the viral penton's RGD attachment sequence (6); (*e*) less efficient escape of the virus from the liposomes, an event triggered by a drop in pH, which could be opposed by the alkalinization of proliferating cells in G_1 (7); (*f*) increased methylation or destruction of viral DNA by dividing SMC; (*g*) failure of proliferating (or apoptotic) SMC, both of which are more abundant in atherosclerotic vessels (8), to transcribe the viral DNA; (*h*) cytopathic effects of the virion, particularly for macrophages, which must have taken up more virions than did the SMC, and which showed no transgenic expression whatever.

Without the permeabilizing effect of balloon delivery (9), the efficiency might have been 0.02%, yet still more efficient than lipofection. This is probably not limiting for the production of practical amounts of prostacyclin (10) or nitric oxide (11), but transgenes whose products act within the cells need to be expressed in most of the cells, not 0.2%.

Once the mechanisms of reduced efficiency have been identified, solutions may be found by reengineering Ad 5 for less matrix-binding, toxicity, or immunogenicity (12). Alternatives include (*a*) replication-defective retroviruses, which transduce only proliferating cells and insert into the chromosomes, yielding the potential for long-term expression and a slight risk of mutagenesis (13); (*b*) parvoviruses, which target proliferating cells (14); adeno-associated viruses, which tend to integrate into a silent region of human chromosome 19 but carry only 4.5 kb DNA (15); (*c*) herpes viruses, which target SMC (but will have to be engineered to be less toxic and still target SMC, and may be immunoneutralized in most patients [16]); (*d*) the HVJ-liposome construct (11); or (*e*) other ligands (17), such as basic fibroblast growth factor (FGF-2) bound to DNA (or liposomes or virions). The up-regulation of SMC and macrophage FGF receptor 1 in injured vessels, and FGF-2's DNA-binding (pI 10.5), lysosomal escape, and transport into the nucleus (especially in proliferating cells) recommend FGF as a vector, though matrix-binding and mitogenesis could pose problems (16, 18–20). Conceivably, each of these approaches may eventually find a clinical niche.

Finally, atherosclerosis may require optimization of local delivery protocols (9) (pressure, volume, and dwell time) and new techniques, such as polymers and iontophoretic catheters (21).

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