Crippled HIV debuts as gene therapy tool

If you talk to gene therapists long enough, the word "vector" always comes up. What's more, it is often accompanied by a sigh or a frustrated shake of the head. Vectors are delivery vehicles for genes, and investigators simply have not had effective ones.

For inspiration, vector designers have now turned to a surprising source—HIV, the deadly virus that causes AIDS. Crippled versions of HIV can efficiently insert new genes into chromosomes, report investigators from the Salk Institute in La Jolla, Calif., and the Whitehead Institute for Biomedical Research in Cambridge, Mass., in the April 12 SCIENCE. The modified virus can perform that task even in cells that are not dividing, such as the neurons in a rat's brain.

Currently, no other gene therapy vector can make that claim. And since most cells in the human body are not dividing or are dividing very slowly, the novel vector has prompted renewed hope that gene therapy may someday fulfill its promise of curing genetic diseases such as cystic fibrosis and treating illnesses such as Alzheimer's.

"I think it's one of the most important vector developments in years," says gene therapist James M. Wilson of the University of Pennsylvania in Philadelphia. "It was a concept that made sense, and a lot of people were thinking about it, but [the Salk and Whitehead teams] have reduced it to practice, and it works."

Successful gene therapy, many investigators contend, will depend on finding vectors that insert therapeutic genes into a cell's chromosomes. This integration ensures that the genes will code for their proteins indefinitely and will be copied whenever a cell divides.

Some currently used vectors cannot carry genes into chromosomes, although they can target nondividing cells. Others integrate genes into a cell's DNA only when the cell divides and the nuclear membrane around the chromosomes breaks up.

In contrast, lentiviruses, including HIV, have evolved a variety of means for sneaking genetic material into the nucleus of nondividing cells. "We took advantage of that property of HIV," says Inder M. Verma of the Salk Institute.

To create the new vector, Verma and his colleagues kept the genes that enable HIV to integrate its genetic material but threw away those that enable it to reproduce and bud from an infected cell. Furthermore, instead of using the gene for the protein that makes up the outer envelope of HIV, they borrowed an envelope protein gene from a virus that targets more than just immune cells, which HIV normally infects.

From that mix of genes, plus the genes the scientists wanted the vector to deliver, the investigators churned out copies of the new vector. "We made a debilitated HIV which can infect a large number of cell types," says Verma.

The vector bears little resemblance to the AIDS virus, say researchers. The only feature of HIV that remains is the ability to slip genetic material inside the nucleus of a nondividing cell, observes gene therapist Joseph C. Glorioso III of the University of Pittsburgh.

Nevertheless, there remain legitimate safety concerns about using the new vector in humans, concedes Verma. For example, the investigators have not established how often, if ever, such an HIV-based vector might recover the ability to replicate by borrowing genes from other viruses in the body or even from the host's own genome.

Verma suggests that these initial cell and animal experiments are best seen as proof of the principle that lentiviruses can provide effective vectors for nondividing cells. He and his colleagues started with HIV because so much more is known about it than any other lentivirus. They contend that vectors based on other lentiviruses, such as the monkey or cow forms of HIV, should prove safer and just as effective.

"People will feel better about vectors that are not associated with human disease," agrees Glorioso. —*J. Travis*

Kinky business: Watching atoms wriggle

What's more annoying than a bump in a rug? Step on it, and it shifts beneath your feet but doesn't flatten out.

At the molecular level, the surfaces of ductile materials were thought to harbor similar structures. Heat a sheet of metal or silicon enough to soften its brittleness, bend it ever so slightly, and tiny kinks, or dislocation lines, could form on the curved surface, looking somewhat like a series of speed bumps.

Until recently, this analogy—devised to explain how soft materials bend—had never found observational support. Now, John C.H. Spence, a physicist at Arizona State University in Tempe, and his colleagues have developed a technique—called forbidden reflection electron microscopy—to watch such kinks form and move on a flexible surface.

"We've been able to see a kink at atomic resolution, watch it move, and determine how fast it moves in response to a given pressure," Spence said this week at a meeting of the Materials Research Society in San Francisco. "Until now, no one's actually observed these structures in motion."

Although researchers have assumed that the motion of these kinks controls the strength of ductile materials, Spence says, "it's all been theory. No one's seen it happening."

Robert Hull, a materials scientist at the University of Virginia in Charlottesville, agrees. "This is the first time that anyone has directly seen kinks moving in a material. That's really quite remarkable."

According to the new observations, it is the speed with which those bumps grow and move that controls how easily the material yields to pressure. The softer and more pliant a material is, the more easily the kinks form and roll along. In contrast, a brittle material resists forming kinks and stays stiff in the face of pressure.

As a surface bends, the kinks look like "strands of spaghetti zipping around," says Spence. "The million-dollar question is how fast those dislocation lines move in response to a given pressure and what impedes their motion."

Working with physicists Helmut Alexander and Harold R. Kolar at Arizona, Spence's team applied pressure to a hot strip of silicon under the gaze of an atomic-resolution electron microscope that they adapted to detect electrons bouncing off the bumps that form as the material bends.

Ordinarily, scientists scan an ordered crystal lattice of atoms to study the material's regularity. Certain reflections are referred to as "forbidden" because they never are produced by these lattices. Spence's group adjusted the microscope to capture such forbidden reflections, which arise from the irregularities caused by the pressure wave moving through the silicon.

The new observations help to explain, for instance, why adding impurities to a material can strengthen it. Pure aluminum is "almost too soft to be useful," Spence says. "But if you add impurities, it becomes stiffer."

Spence has observed that impurities block kinks from forming and moving, so the material won't bend easily. "If we can block the motion of dislocations, then a material can be strengthened," he says. "One might want to do this by introducing obstacles on the atomic scale."

Although the aim of these experiments is to understand the behavior of ductile materials, Spence says, the new technique may help in designing flexible alloys less likely to fracture under intense heat or stress—like the conditions that can cause jet engine turbine blades to fail. —*R. Lipkin*