Gene therapy escapes the immune response

Lately, enthusiasm over gene therapy has given way to skepticism. The National Institutes of Health, for instance, is evaluating whether investigators are rushing too quickly to start human trials of gene therapy.

As this reflection goes on, researchers continue to challenge the barriers to gene therapy's success. According to a report in the September NATURE MEDICINE, investigators have taken a crucial step toward overcoming one such barrier. They've found a way to sneak genes past the body's defenses more than once, a feat that might allow repeated gene therapy efforts in the same patient.

One of the largest obstacles to gene therapy is the immune response, a usually welcome defense against viruses or bacteria. Researchers often use crippled viruses, ones that can't replicate, to ferry healing genes into cells. For example, adenoviruses, which cause respiratory infections, are a popular method of targeting the lungs.

But the immune system makes no distinction between good and bad viruses. In animals, immune cells eventually kill any cells infected by the engineered adenoviruses, limiting how long the genes that they have carried in will be active. Moreover, the immune response generates antibodies that neutralize adenoviruses, preventing gene therapists from using that delivery method more than once. That's a major problem for diseases, such as cystic fibrosis, where patients will need repeat doses of curative genes, says James M. Wilson of the University of Pennsylvania Medical Center in Philadelphia.

Therapists are therefore scrambling for ideas to sneak gene-carrying viruses past the immune system. One method may be to briefly suppress immune responses when they introduce gene-loaded viruses, Wilson and coworkers report in NATURE MEDICINE.

Previously, Wilson's group injected mice with antibodies that target CD4 cells, a type of white blood cells active in immunity. They synchronized these injections with the administration of virus-carried genes. The antibodies prevented the immune system from attacking infected cells, as demonstrated by the fact that the introduced genes were still active one month later. Furthermore, since CD4 cells are vital to the production of virus-neutralizing antibodies, the treatment's attack on CD4 cells allowed Wilson's group to effectively use their virus a second time.

Since the treatment with antibodies to CD4 cells may itself generate an immune response in humans, Wilson and his colleagues have explored another concept. When they administered gene-transporting adenoviruses into the lungs of mice, they also injected the animals with either interleukin-12 or gamma-interferon, two natural chemicals that the immune system uses to communicate between cells.

These so-called cytokines prevent the deployment of certain immune cells that activate antibody-producing machines known as B cells. As a result, mucus lining the lungs of these mice produced just one-twentieth as many adenovirus-neutralizing antibodies as those of mice not given cytokines, Wilson's group reports. About 1 month later, the investigators successfully used an adenovirus to deliver a second gene into the lungs.

The central unresolved question, says Savio L.C. Woo, a gene therapy expert at Baylor College of Medicine in Texas, is "Can this be done multiple times?"

In the coming months, predicts Wilson, gene therapy groups will explore many other methods intended to temporarily and safely suppress the immune response against virus-ferried genes.

Investigators, notes Woo, are also designing gene-carrying viruses that elicit much less of an immune response. "In the end, it may take a combination of both strategies," says Woo. — *J. Travis*

Unraveling role of the breast cancer gene

Last October, an army of collaborating researchers from five North American medical institutions pinpointed the genetic flaw associated with some five percent of all breast cancers (SN: 9/24/94, p.197). Scientists around the world had hoped that this gene—a mutant form of *BRCA1*—would provide insight into the causes of most other breast cancers as well. Those hopes were dashed, however, when *BRCA1* proved to be an unusually complex gene associated only with a small proportion of inherited forms of breast and ovarian cancers.

Now, work on the mouse equivalent of *BRCA1* offers the first clues to the role played by the normal form of this gene. It appears to help control cell growth and maturation throughout the body, according to a report in the September NATURE GENETICS.

"We weren't really expecting [the *BRCA1* gene] to be this broadly expressed," says Lewis A. Chodosh of the University of Pennsylvania School of Medicine in Philadelphia, an author of the new study. "It makes us think that *BRCA1* is not just playing a role in the breast."

Women who inherit a flawed copy of the *BRCA1* gene not only face an 85 percent chance of developing breast cancer at some point during their lives, but also experience a substantial increase in ovarian cancer risk. In recent months, researchers have also noted that people carrying a mutant copy of the gene experience more colon and prostate cancer. Although some researchers suggest normal *BRCA1* may suppress tumors, no one knows how *BRCA1* flaws might foster cancer.

To get an idea of just when the *BRCA1* gene "turned on" and where it was expressed, the new study—conducted by researchers from Penn, the University of Michigan School of Medicine in Ann Arbor, and the National Center for Human Genome Research in Bethesda, Md.—studied embryonic, pubescent and adult mice. Surprisingly, the gene proved active in nearly all the immature tissues of very young embryos, but as the tissues matured, the embryos developed a specific pattern of *BRCA1* expression which included the liver, lung, salivary gland and thymus.

In adult mice, the gene proved most active in the testes, which produce sperm, and in the thymus, which makes T cells, the immune system's laborers. While the breast, ovary, uterus and liver also expressed significant levels of the gene, *BRCA1* appeared inactive in the kidney, heart and brain. This activity pattern suggests *BRCA1* is most active in tissues that produce many developing cells, says Chodosh.

Because the breast develops largely after birth, the researchers investigated whether *BRCA1* expression increased during times of breast development as a result of stimulation by the hormones estrogen and progesterone. And they indeed found substantial increases in *BRCA1* activity during puberty, when mammary ducts are beginning to form, as well as during early pregnancy, when those ducts mature to express milk.

Remarkably, *BRCA1* activity levels remained elevated long after pregnancy. Giving estrogen and progesterone in amounts typical of pregnancy also increased *BRCA1* activity in mice that cannot produce those hormones. If hormones turn on *BRCA1*'s tumor suppressing activity, "we could potentially use currently available [hormone-mimicking] drugs to protect against breast cancer," speculates Chodosh. However, he notes that work is still preliminary.

Myles Brown of the Dana Farber Cancer Institute in Boston says, "The work is an important first step, but only a first step, in understanding *BRCA1*."

— *L. Seachrist*