RESEARCH BRIEFS

## Shuffling viral tropism

Current viral vectors for gene therapy have many disadvantages: they cannot efficiently infect certain types of cells, they often stimulate an immune response, and they do not typically accommodate large genes. Nay-Wei Soong and colleagues now have developed a general strategy for creating "better" viruses (*Nat. Genet.* **25**, 436–439, 2000). Using in vitro molecular breeding to mimic genetic variation and recombination, followed by selection, they "evolved" a virus for new tropism toward a specific target cell. The researchers combined DNA encoding the envelopes from six strains of a virus in nearly one million configurations, and tested whether the recombinant viruses could infect a cell type that none of the six strains would normally infect. By repeating the infection process five times and giving the best virus ample opportunity to outcompete the others, they managed to recover a new subtype of virus that had the ability to infect new target cells efficiently. David Curiel of the University of Alabama (Birmingham, AL) comments that although the technology might have some limitations, it has great potential in "pushing the envelope" of vector development. *MR*

## Teaching introns new tricks

Researchers have developed a general technique for specific gene targeting using a mobile bacterial intron that copies itself into genomic DNA (*Science* **289**, 452–457, 2000). The group II intron from *Lactococcus lactis* is a catalytic RNA that splices itself out of an RNA transcript with the help of an intron-encoded protein (IEP). The excised intron—bound to the IEP—then inserts into the genome at a particular sequence and is reverse transcribed by the IEP into cDNA. To better understand the rules of target-site recognition, the researchers devised a genetic assay that could measure the contribution of individual nucleotides in the recipient DNA to intron mobility, and found they could redirect the intron by mutating a small region expected to base-pair with the target. They also selected introns capable of inserting at a desired destination from a combinatorial library of randomized sequences. The researchers have achieved integration into bacterial genomes and are now taking aim at eukaryotic chromosomes. "If the technique can be made to work for chromosomal insertion in human cells, then the full range of gene therapy applications would become possible," says senior author Alan Lambowitz. *KA*

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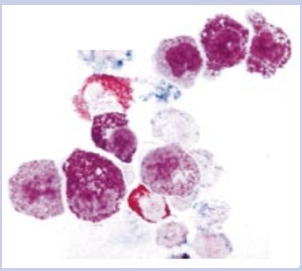
## Reaching a critical mast



By deriving mast cells in vitro from genetically manipulated embryonic stem cells, a team of researchers have overcome the problem of analyzing embryonic lethal mutations. The work also paves the way for tissue engineering, in which highly differentiated cell types could be generated in vitro and transplanted to restore specific functions in vivo. Previously, mast cells were shown to be generated from hematopoietic precursors in vitro, making mast cells an attractive target for the new work, which is described in a recent issue of *PNAS* (**97**, 9186–9190, 2000). The researchers generated mast cells from both wild-type embryonic stem cells and stem cells carrying a homozygous deletion of SEK1. Although SEK1 has been implicated in certain mast cell activities, the knockout has been difficult to study because of its embryonic lethal phenotype. The team found that both wild-type and SEK1 knockout mast cells appear to function normally after transplantation into a mast cell–deficient mouse strain. Stephen Galli, a researcher at the Stanford University School of Medicine (Stanford, CA) and senior author on the study, explains that the new approach has broad potential, because "mast cells are thought to participate importantly in many biological responses ... such as angiogenesis, wound healing and tissue remodeling, responses to neoplasms, and non-immunological forms of chronic inflammation, to name just a few." *AD*

## Gene therapists install insulation to boost efficiency

In an approach that could significantly increase the efficiency of a variety of gene therapies, researchers at the University of Washington (Seattle, WA) have demonstrated that the addition of a chromatin insulator can protect a retroviral gene vector from position effects that otherwise tend to silence inserted genes. Although they are used widely in gene therapy, murine retrovirus-based vectors integrate randomly and frequently insert themselves into the transcriptionally silent heterochromatin that makes up the bulk of a cell's genome.

In the new work, described in the 1 August issue of *PNAS* (**97**, 9150–9155, 2000), the researchers flanked a reporter gene in a retroviral vector with the HS4 chromatin insulator from the chicken β-globin locus control region. Chromatin insulators, which shield promoters from the activity of nearby regulatory elements, have been found in a variety of species. In cultured cells and mice transplanted with transduced bone marrow, the flanked vector was expressed in a significantly higher fraction of transduced cells than a control lacking the insulators. David Emery, a researcher at the University of Washington and first author on the paper, cautions that "it is possible this and other chromatin insulators may not work with every promoter and in every tissue," but adds that the team is now testing a variety of constructs for possible clinical use. *AD*

## Humanized mice for toxin sensing

Ronald Evans and colleagues at the Salk Institute (La Jolla, CA) report the first transgenic mouse that can sense and react metabolically to human toxins (*Nature* **406**, 435–439, 2000). Animal species respond differently to foreign substances, an observation that prompted the researchers to identify species-specific receptors for xenobiotic sensing mechanisms. Previously, the researchers identified SXR, a human nuclear receptor that responds to xenobiotic compounds by activating cytochrome CYP3A gene expression, leading to toxin degradation. Hypothesizing that evolutionary divergence of this receptor could be responsible for species-specific xenobiotic sensing, they generated mice with a targeted disruption in the mouse homolog, PXR, and showed they could no longer activate CYP3A gene expression in response to rodent-specific CYP3A inducers. Next they introduced an activated form of the human SXR gene into the PXR-null background, and showed that the mice could now sense and degrade human xenobiotics to which rodents are naturally susceptible. Evans believes that the model could be used to screen drugs for spurious activation of SXR, leading to drug–drug interaction. "Antibiotics and even herbal remedies such as St. John's Wort can trigger SXR," asserts Evans, "and cause women taking oral contraceptives to deliver 'miracle babies'." *AJB*