A kinder, gentler war against hepatitis B

Virologists have long known that cells called cytotoxic T lymphocytes (CTLs) battle viruses by latching onto an infected cell and delivering the kiss of death—an outburst of compounds that destroys both the cell and its infectious cargo.

Now, investigators are realizing that these immune cells may sometimes have a gentler bedside manner. In fighting the hepatitis B virus, for example, these lymphocytes appear to secrete compounds that provoke infected cells into destroying the viral molecules being produced inside them. This activity suppresses the infection without harming the infected cells.

"This appears to be a mechanism vertebrates have evolved to deal with infections without killing themselves," says Francis V. Chisari of the Scripps Research Institute in La Jolla, Calif.

For the last 2 decades, Chisari has studied how the immune system combats the hepatitis B virus. Though an effective vaccine for the virus exists, its distribution is limited and an estimated 300 million people worldwide are already infected. Since the virus infects liver cells called hepatocytes, these people face a significant risk of cancer or long-term liver damage, says Chisari.

Because it doesn't infect the mouse, the animal most widely used in biomedical research, hepatitis B virus has been a difficult infectious agent to study. To bypass the problem, Chisari and his colleagues have gradually added all the genes from the virus into the mouse genome. As if infected by hepatitis B itself, the liver cells of such a mouse turn into viral factories, producing new copies of the virus and secreting them into the animal's bloodstream.

Working with these mice, Chisari, Luca G. Guidotti of Scripps, and their colleagues have recently discovered that they can completely eliminate the hepatitis B virus from a mouse's bloodstream by injecting CTLs that target the hepatocytes producing viral proteins.

Because the CTL injections killed only about 5 percent of the hepatocytes, the investigators began to suspect that CTLs do more than slay cells, says Chisari. His group then found that when CTLs recognize infected cells, they secrete two soluble compounds, tumor necrosis factor-alpha and interferon-gamma. These compounds diffuse through the liver and stimulate antiviral activity in distant liver cells.

Chisari's group has described the nature of that activity in several reports over the last few years, including articles in the Dec. 15, 1995 PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES and the January IMMUNITY.

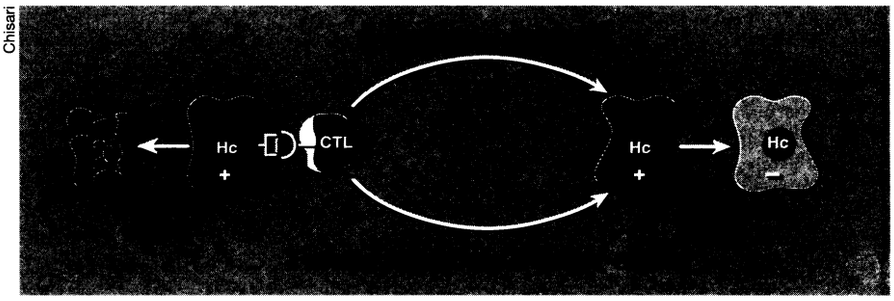
In their first response, stimulated liver cells produce proteins that degrade the molecules that would make up hepatitis B's protective viral shells.

Shortly afterward, these liver cells make proteins that interfere with the replication of the virus' genetic material. In order to create new viruses, some of the viral genes in the mice produce RNA, a molecule similar to DNA. The new liver proteins appear to destroy specifically the viral RNA.

Chisari contends that human liver cells may attempt to suppress hepatitis B in a similar manner. In addition to identifying the antiviral proteins produced by hepatocytes, his group would like to develop methods to stimulate the hepatocytes' antiviral response directly.

Chisari also suggests that what his group has learned about how CTLs confront hepatitis B may help researchers tackling other viruses, including the AIDS-causing HIV. "It gives us a little more information on how cytotoxic T cells can contribute to defense by causing cells themselves to become more resistant," agrees Barry T. Rouse, a viral immunologist at the University of Tennessee in Knoxville. —*J. Travis*

Chisari



Immune cells called CTLs can either destroy infected liver cells (Hc+) or secrete compounds that help infected cells eliminate the viruses within (Hc-).

Cancer gene found vital to mouse embryos

Like no other bit of DNA, the gene behind breast cancer inspired a worldwide quest. After victoriously nabbing it, however, scientists found themselves confronting the laborious task of determining what role this gene, *BRCA1*, normally plays and how mutations in it lead to cancer.

Now, a newly developed strain of mice genetically engineered to be defective in *BRCA1* is beginning to offer some clues. It seems the gene may play a vital role in nervous system development.

"With this mouse, we have a potentially powerful model system" for studying *BRCA1*, says project leader Beverly Koller of the University of North Carolina at Chapel Hill Medical School. "And, for the mouse, *BRCA1* is essential. Without it, they die before birth."

After a group of Utah researchers announced that they had found *BRCA1* (SN: 9/24/94, p. 197), three other groups identified a total of 22 mutations in the gene. Any one of these mutations confers an 85 percent lifetime risk of breast cancer (SN: 12/3/94, p. 372).

The mutations have one thing in common. They stop cells from making a functional BRCA1 protein.

Everyone inherits a copy of *BRCA1* from each parent. A single normal copy of *BRCA1* is enough to put the brakes on abnormal cellular growth. But if the normal copy is damaged, cancer can arise.

Koller and her colleagues attempted to make a mouse that begins life with two defective copies of the gene. The team first created mice that carried a mutation in one copy of *BRCA1*, then bred those mice in order to produce some offspring with mutations in both copies.

The matings failed to create any of the desired *BRCA1* knockout mice.

Koller and her team hypothesized that the mouse embryos had died before birth. As the researchers report in the February NATURE GENETICS, they found that many of the knockout embryos had a spine that didn't seal completely or no brain. In addition, cells in the embryos had overgrown and failed to mature.

The embryos died after 10 to 13 days, about halfway through normal gestation. Because certain breast and nerve cells mature from the same type of embryonic cells, Koller suggests that the BRCA1 protein may help both cell types mature.

These findings may not be directly applicable to humans. Scottish researchers described in the June 15, 1995 NATURE a breast cancer patient with a normal nervous system who inherited two mutant *BRCA1* genes. Koller points out, however, that the second mutation may have arisen in the individual's lifetime and that researchers need to identify other such patients.

The mouse development "offers an important tool for studying the function of *BRCA1*," says Barbara L. Weber of the University of Pennsylvania in Philadelphia. Koller notes that the mice with single mutations are still too young to have developed tumors. If they do, Weber points out, they would provide a model for developing breast cancer treatments and preventive measures. —*L. Seachrist*