

With each large and small victory against HIV—and there have been many in the last two decades—the virus reinvents itself and evades total defeat.

By Marlene Cimon

Is
HIV
Smarter
Than
We
Are





A masked technician uses a centrifuge to separate viruses such as HIV or hepatitis from blood cells. A high-speed spin causes the higher density blood cells (red) to separate from the virus, seen as an opaque layer within the less dense, clear fluid.



What Robert F. Siliciano saw when he entered his lab five years ago triggered conflicting emotions. He was elated because the clear substrate in the plates had turned blue, confirming his hypothesis. But he also felt a deep sadness because it meant bad news for those AIDS patients who had been enjoying remarkable clinical benefits from new protease inhibitor drugs.

His tests showed that the human immunodeficiency virus (HIV) was still in their bodies, despite tantalizing hope that the drugs could eradicate HIV.

“That was a moment that really shook us up,” says Siliciano, an HHMI investigator who conducts AIDS research at The Johns Hopkins University School of Medicine. “It was the realization that this virus will remain in their bodies forever.”

In the more than 20 years since the earliest AIDS cases were documented, HIV has proved a formidable foe, thwarting some of the sharpest scientific minds in the world. The National Institutes of Health has poured \$24.3 billion into HIV research since 1982, including \$2.77 billion this year alone, with impressive results that have often translated into widespread benefits reaching far beyond the boundaries of AIDS (see sidebar, page 15). Yet for each hurdle scientists have overcome, HIV, in maddening fashion, seems to devise another to throw in their path.

Each new promising drug, for example, has brought elation with its efficacy, only to be followed by despair as the virus developed resistance to it. “We know so much more about HIV than any other virus,” says HHMI investigator Bruce D. Walker, director of the Harvard Medical School’s Division of AIDS. “But HIV is a very tough nut to crack. It ultimately gets the upper hand.”

Is HIV truly smarter than we are? To be sure, a virus has no innate intelligence; it’s just a bit of genetic material surrounded by a protein coat, trying to stay alive. “It may look like it’s smart, but it’s doing what it is programmed to do, which is to replicate and adapt,” says Anthony S. Fauci, director of the National Institute of Allergy and Infectious Diseases (NIAID). Certainly, other viruses do that too, but HIV, a retrovirus, has the insidious habit of permanently integrating itself into the host’s cells, unlike flu viruses, for example, which wreak their temporary havoc and leave. Because HIV is a retrovirus, it contains RNA rather than DNA, and uses an enzyme known as reverse transcriptase to convert its RNA into DNA as it integrates, turning the body’s own cells into little virus factories.

One reason the virus is so difficult to fight, says Robert C. Gallo, director of the Institute of Human Virology at the University of Maryland in Baltimore (and HIV’s co-discoverer, along with France’s Luc Montagnier), is that “the ability of HIV to replicate itself is tremendously greater than that of other retroviruses. And it’s a newer infection in mankind, so we are less adapted to it.”

“Every time the virus replicates, it is capable of changing,” Fauci says. “And every time the virus mutates, it has the potential of assuming a form that can avoid destruction” by drugs or a vaccine. Also, the virus does not exist as one universal strain. There are subtypes, and people can become infected with more than one. New subtypes could present “an even bigger problem for an AIDS vaccine than the simple mutation problems people talk about now,” says June E. Osborn, a virologist who chaired the U.S. National Commission on AIDS and now runs the Josiah Macy Jr. Foundation in New York City. “When I want to worry late at night, that’s what I worry about.”

Still, a tremendous amount of understanding has accumulated about the behavior of HIV, including the various ways in which the virus infects



BILL DENISON

Robert Siliciano has seen HIV held in check, but the drugs are still too toxic.

immune-system cells. For example, HHMI investigator Dan R. Littman at New York University School of Medicine and colleagues have focused on how HIV invades helper T lymphocytes, the cells that are destroyed by HIV. Elimination of helper T cells leaves the body vulnerable to life-threatening infections. Most recently, in a paper published January 2002 in the journal *Immunity*, the researchers showed that for potent infectivity, HIV must be taken up by the dendritic cells on mucosal surfaces. “The virus hops onto the dendritic cell to hitch a ride to the lymphatic tissues, where the T cells are, and then infects these cells very effectively,” Littman explains. However, at this point, he adds, “I’m not aware of any approaches to exploit the dendritic cell work to develop therapies or vaccines.”

PROGRESS AMID PROBLEMS

Despite the roller-coaster nature of progress against AIDS, researchers point to great advances in drug development during the past 15 years. The growing arsenal of drugs has transformed an AIDS diagnosis from an automatic death sentence; patients now expect to manage the disease as a chronic illness.

The first anti-AIDS drug, AZT, was approved by the Food and Drug Administration in 1987 with much fanfare. Until then, antiviral drugs were nearly nonexistent, so the rapid development and licensing of AZT was regarded as a triumph. It prolonged survival and improved quality of life. But AZT, like every AIDS drug that would follow, proved problematic, bringing nasty side effects and eventual viral resistance. It did,

however, offer an important lesson—that viruses could be attacked and controlled, even if the effect was only temporary.

With that hopeful note, pharmaceutical companies pumped more money, time and energy into studying other compounds, while clinicians tried innovative ways to use them, going beyond single-drug therapy to the approach that is practiced today—cocktails of different compounds that hit the virus in multiple ways.

Today, 16 drugs in four different classes are licensed to treat HIV. Three of the classes block reverse transcriptase; the fourth, which blocks another enzyme, called protease, dramatically changed the landscape of AIDS treatment when it was introduced in 1995. Used in cocktail combinations known as “highly active antiretroviral therapy” (HAART), the protease-inhibiting drugs proved much more potent than the earlier drugs; the annual numbers of U.S. deaths plunged 60 percent—from 38,100 in 1996 to 15,300 in 2000, according to the Centers for Disease Control and Prevention.

The early impact of the protease inhibitors led some researchers, including David D. Ho, director of the Aaron Diamond AIDS Research Center in New York City, to suggest for a time that HAART might be able to completely eliminate HIV from the body.

Not so. The research of Siliciano and other groups revealed hidden reservoirs of HIV. Later, Ho’s work showed that low-level viral replication continues, even when patients are on HAART. Today Ho says: “It is clear that eradication is going to be very, very difficult.” Siliciano agrees. “I don’t think it will be possible to hit the resting cells,” he says. “They

are rare, one in a million, and they are not making any RNA or protein. They are indistinguishable from the rest.”

BLOCKING ENTRANCE

Although no one has any illusions at present that some particular drug combination will be a panacea, researchers remain convinced that newer and better drugs are still the best chance of helping those who are already infected, so the work on new drugs continues. In July, researchers at the International AIDS Conference in Barcelona heard encouraging news about the first new AIDS drug in six years—T-20, the first member of a long-anticipated family called fusion inhibitors. Unlike current drugs, which block enzymes needed for replication, fusion inhibitors keep HIV from entering cells.

The drug, developed by Trimeris of Durham, North Carolina, and to be marketed as Fuzeon by Hoffmann-La Roche, was tested in patients who no longer responded to other AIDS drugs. It reduced their blood-borne virus levels by three-quarters, and doubled the percentage of patients in whom the virus fell to undetectable levels.

Researchers are also trying to improve existing drugs by changing their formulation to encourage compliance; regimens of at least 3 to at most 20 pills per day have sometimes been difficult to follow. Depending on the regimen and the patient population, anywhere from 5 to 35 percent of patients abandon multidrug therapy.

“With some of these drugs formulated in the same capsule, we can sup-

Lessons Learned—and Shared

T

he early burst of activity in support of AIDS research occurred against a backdrop of resentment among advocates for research on other diseases. Many complained that AIDS afflicted far fewer people than their own conditions of interest, yet it received far more research funding.

In recent years, however, such criticism has been muted by the growing applications of AIDS research to other medical areas. Despite the dogged inclination of HIV to elude scientists’ best efforts, their work has produced a wealth of knowledge that goes well beyond the scope of AIDS; in particular, it has provided models for fighting other chronic viral infections, historically among the toughest to treat.

As researchers began to better understand how HIV worked, they discovered that the more hits a virus took during different stages of replication, the more effective the result. This led to the development of protease inhibitors and other drugs that could deliver one-two punches at different phases in HIV’s cycle.

Scientists are now applying that same approach to other viral diseases. Both hepatitis C and hepatitis B, which together infect an estimated 600 million persons worldwide, are prime candidates. Like HIV, hepatitis C makes a protease enzyme that is neces-

sary for viral replication. And hepatitis B makes an enzyme that is similar to HIV’s reverse transcriptase.

“We are learning in hepatitis C treatment, as we learned with HIV, that hitting multiple metabolic pathways of the virus using multiple drugs is effective,” says Lawrence Deyton, who directs HIV and hepatitis C programs for the U.S. Department of Veterans Affairs. Moreover, “the knowledge gained in understanding virology and the viral/immune-system interactions—everything we didn’t know before HIV—will really help us leapfrog in [hepatitis C research],” Deyton says. “We are just taking all our cards off the HIV table.”

The same goes for hepatitis B. Patients who suffer from chronic hepatitis B have a new drug, adefovir dipivoxil, made by Gilead Sciences of Foster City, California. The Food and Drug Administration recently approved the drug to help treat this life-threatening infection. Adefovir was initially tried in AIDS patients but was rejected as too toxic for the kidneys. In lower and safer doses, however, it is effective against hepatitis B.

The other major hepatitis B drug, lamivudine (also known as 3TC and epivir), made by the U.K.-based GlaxoSmithKline, also got its start as an AIDS treatment. In fact, it is still used against HIV in combination with other drugs.

For hepatitis C, the combination of interferon and ribavirin—also originally an HIV drug—has produced a better sustained viral response than single-drug therapy.

“Before HIV, many drug companies were scared of researching antiviral drugs,” says Vicki L. Sato, president of Vertex Pharmaceuticals in Cambridge, Massachusetts. But lessons learned from HIV “gave us the confidence to try.” The company has a protease inhibitor candidate against hepatitis C, called VX-950, which could go into clinical trials sometime next year.

Researchers also are looking to other areas to apply AIDS advances. Among those considered the most promising: blood disorders, autoimmune diseases and cancer. For example, in cervical cancer, non-Hodgkin’s lymphoma and Kaposi’s sarcoma, both viruses and the immune system are believed to play a role.

Similarly, research into HIV vaccines, however frustrating, may open the door to developing vaccines against other infections. “All sorts of novel strategies are being pursued in HIV vaccine research,” says David D. Ho, director of the Aaron Diamond AIDS Research Center in New York City. “If this ultimately results in a protective vaccine, the lessons will be most useful for other vaccines, particularly for malaria and tuberculosis.” —M.C.

press viral replication for years with two pills a day, with very few side effects,” says Robert T. Schooley, an AIDS specialist who heads the infectious diseases division of the University of Colorado Health Sciences Center. “If they are used the way we prescribe them, resistance will be a rare event because the virus won’t be replicating fast enough to develop mutations.”

And some patients continue to do well on existing HAART. In these individuals, Siliciano has seen viral evolution stop. “Those who can maintain below 50 copies [of viral RNA] per milliliter of plasma have halted replication and resistance,” he says. “This means, at least in principle, that it is possible to permanently hold the virus in check.” Still, the drugs are toxic. But “if we can develop nontoxic drugs, it should be possible for a patient to have a normal life,” he says.

TIMING IS EVERYTHING

One way researchers are trying to make the drug regimens more tolerable and reduce side effects is through carefully timed drug interrup-

tions, which offer a reprieve from drug toxicities, save on drug costs and, in some cases, even train the body’s immune system to kick in and gain control of the virus.

Researchers at NIAID have been experimenting with a seven-day-on/seven-day-off cycle in which patients go off treatment but then return to it before the virus starts to bounce back. Fauci and NIAID colleague Mark Dybul have followed 10 patients for two years on this routine, and “they are still doing very well,” Fauci says. The researchers are doing an expanded study of a similar protocol now in the United States and, soon to start, in Uganda.

Walker and colleagues at Massachusetts General Hospital have also tried a novel interruption approach. They’ve been studying 14 patients who started HAART within weeks of learning they were infected. After about 18 months of continuous treatment, they stopped the drugs, returning to treatment only when the virus began to rebound. Over the course of three years, the researchers found that the time between

Turning the Tide

“T reatment is important, and we should take care of people who are sick,” says Barry R. Bloom, dean of the Harvard School of Public Health and HHMI Medical Advisory Board member. “But if we don’t prevent transmission, there will be even more people sick. The challenge is to get the most effective balance.” The global fight against AIDS is not about treatment or prevention, but about both.

Indeed, a report released last July in Barcelona by UNAIDS (the Joint United Nations Programme on HIV/AIDS) recommended a minimum of \$10 billion annually, largely subsidized by the world’s wealthier nations, to achieve two critical goals: more affordable drugs and beefed-up prevention programs. Otherwise, it warned, more than 68 million people worldwide could die from AIDS over the next 20 years.

While the rate of new infections and mortality has been leveling off in the United States and Europe—thanks to prevention and the availability of therapy—the same cannot be said of the epidemic in developing nations. In sub-Saharan Africa, for example, 28.5 million people are living with HIV, but fewer than 30,000 are getting AIDS drugs. The disease is decimating the adult population, having already orphaned an estimated 11 million children in the region.



Children who lost their parents to AIDS gather at the Tithanizane Orphan Care Center in Ndirane Township, Malawi. HIV infects 13 percent of Malawi’s population.

ALEXANDER JOE/AFP PHOTO

Jr. Foundation in New York City. She cites Thailand, Uganda, Senegal, Ivory Coast and Brazil as examples of nations that have been able to turn the tide.

In Thailand, for example, “where young men in the military have a long tradition with sexual commerce, the yearly new-infection rate among military recruits was 20 percent,” Osborn says. “The government did a turnaround on preventive messages about safe sex, and the rate dropped to 3 percent almost instantly.”

In Brazil, the world’s fifth-largest country, aggressive prevention messages and access to free drugs have resulted in a drop of HIV-related hospital admissions by 75 percent since 1997 and a decline in AIDS deaths of 50 percent, according to a 2002 Ford Foundation report.

“Curtailing HIV is in our grasp,” Osborn says. “In those countries where they realize it’s a matter of life and death, the simple messages that we have had from the beginning have worked.”

Bloom agrees. “As primitive as the tools for prevention may be—condoms and exhortation—we know they are working in places like Senegal, Thailand and Uganda,” he says. “HIV never rose in Senegal, and dropped dramatically in Thailand and Uganda. As nontechnical, unsophisticated and unappealing as it sounds, prevention can work. But it takes a huge effort.”

—M.C.

Structures Revealed

stopping treatment and viral rebound had lengthened. They theorize that early treatment helped preserve killer immune-system cells that recognize HIV and that each time the virus returned, those cells increased in number.

“It’s our hope that since we are dealing with a lifelong infection, we can get the immune system to do a better job. If so, we might be able to limit treatment and toxicities,” Walker says. Despite promising results in boosting immunity for patients who receive therapy soon after they become infected, this approach does not appear to work in patients who have been infected for longer periods of time.

IN PURSUIT OF A VACCINE

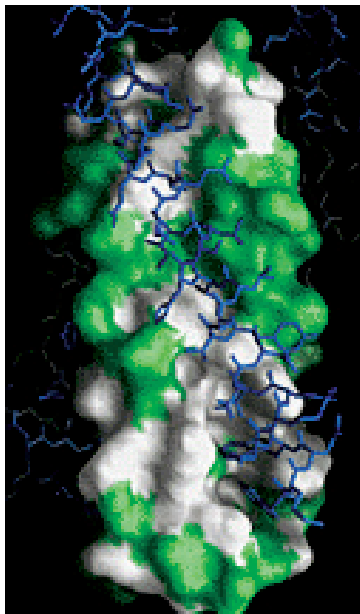
Most AIDS researchers acknowledge that advances in drug therapy have moved at a much faster pace than vaccine research. Nevertheless, no one is giving up the chase. More than two dozen candidate vaccines have undergone early (phase I and phase II) studies in humans for safety and efficacy, and some have progressed to large, phase III studies.

Farthest along of all experimental AIDS vaccines is AIDSVAX, a product developed by VaxGen of Brisbane, California, and manufactured by Genentech. It was the first to enter phase III trials—enrolling nearly 8,000 volunteers in the United States, Canada, the Netherlands and Thailand—and early data are expected in 2003.

AIDSVAX is based on the traditional approach of trying to prevent the virus from establishing an infection by generating antibodies that will immediately bind to the virus and neutralize it. AIDSVAX uses HIV’s surface protein, gp120. Experts generally remain pessimistic, however, believing that a truly effective vaccine against HIV needs to both prevent infection and provide what is known as cell-mediated immunity, which is the ability of immune-system cells to kill those cells that are already infected.

Merck & Co. is testing two versions of its candidate vaccine in humans: The first inserts an HIV gene called *gag* into a plasmid DNA vector, and the second inserts *gag* into a modified adenovirus. The company is conducting studies in uninfected individuals, as well as HIV-positive patients undergoing HAART therapy.

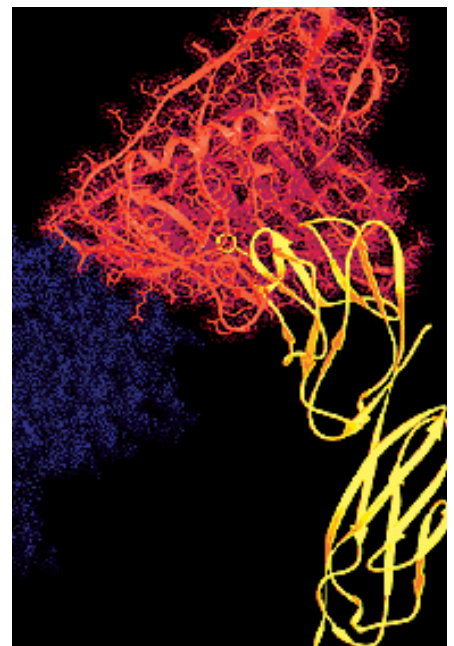
Also, the U.S. and Thai governments announced at the Barcelona conference that they are planning the largest vaccine trial yet, testing two products on more than 16,000 subjects in Thailand. The test will first inoculate subjects with a vaccine designed to stimulate cell-mediated immunity. Developed by the French company Aventis Pasteur, it is a live-virus vaccine that uses the canarypox virus engineered with the genes of several HIV proteins. This will be followed by the administration of



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▶ In 1998, Wayne A. Hendrickson, an HHMI investigator at Columbia University, and colleagues solved the three-dimensional structure of the HIV-1 protein, gp120, that makes first contact with human cells. When this surface protein (red) encounters a lymphocyte that bears the protein CD4 on its surface (yellow), the gp120 docks with the lymphocyte. The virus also must bind to a chemokine receptor, discovered by Dan R. Littman and others, in order to begin infection.

◀ Two groups of HHMI investigators in 1997 independently revealed the structure of a protein fragment, called gp41, from the surface of HIV that penetrates a cell’s membrane, allowing the virus to gain access to the cell’s reproductive machinery. Peter S. Kim, who was an HHMI investigator at the Whitehead Institute for Biomedical Research, headed the first group. He is now at Merck. The late Don C. Wiley and Stephen C. Harrison, both at Children’s Hospital in Boston and Harvard University, were members of the second group.



PETER KWONG AND ERIK MARTINEZ-HACKERT/COLUMBIA UNIVERSITY

AIDSVAX. This is a process known as “prime boost,” in which patients are “primed” with one vaccine, then after a period of time, they are given a “boost” of either the same vaccine or a different one.

Though the researchers behind such efforts are hopeful, they can’t be too confident. HIV-vaccine research efforts, however creative, have all failed to produce broadly useful results. The same characteristics that enable this crafty virus to confound treatment—first and foremost, its ability to mutate—also make it highly adept at eluding an immune-system attack. A universally protective candidate has proved difficult to achieve.

Public health experts insist that while the research continues, the single most effective way to thwart HIV is to return to the basics. And that means prevention through behavior modification (see sidebar, page 16).

NIAID’s Fauci agrees. “The best way to stop HIV, quite simply, is to not allow it to spread from person to person,” he says. “Interrupt the chain of transmission. That’s the way you outsmart this virus.” **H**