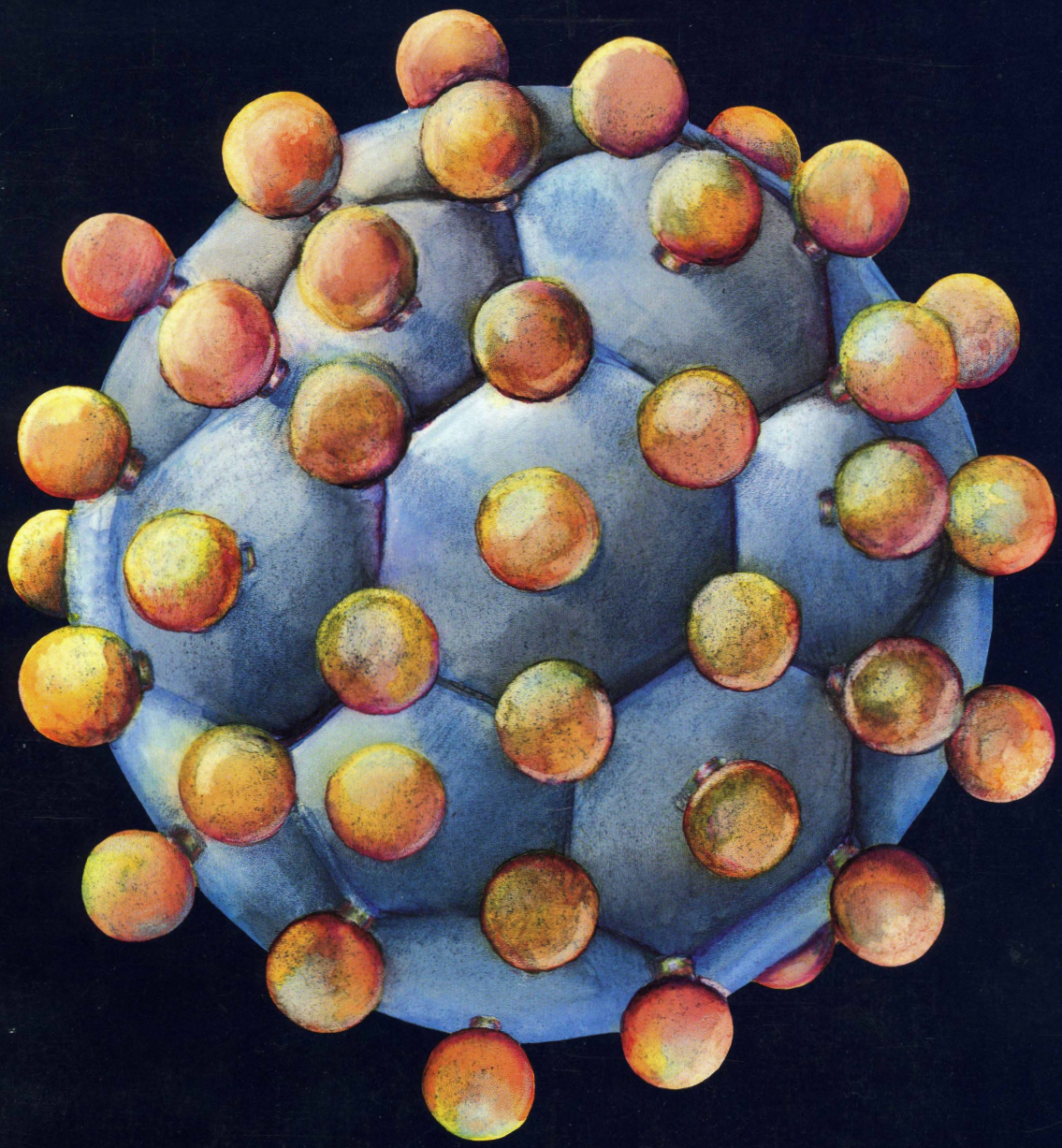
1989

Science vs. AIDS: Defending the Battle Lines

The Rockefeller University



SCIENCE *vs.* AIDS

Defining the Battle Lines

THE ROCKEFELLER UNIVERSITY

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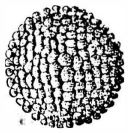

A Report on the Seven Springs International Conference on the Immunological and Infectious Sequelae of AIDS

by LOIS WINGERSON

Sponsored by

THE ROCKEFELLER UNIVERSITY

ABOUT THE AUTHOR

LOIS WINGERSON is an award-winning science journalist who has been a staff writer at *New Scientist*, the British science magazine, and at the New York-area daily newspaper *Newsday*. Her articles have also appeared in *The Economist*, *Discover*, *Science 86*, and *Science Digest*. At present, she is writing a book on gene mapping for publication by E.P. Dutton.

ABOUT THE COVER

An artist's rendition of the surface of the AIDS virus. Illustration by Linda Wilson.

Funding for the conference and this publication has been provided by Katherine Morgan Deane, member of The Rockefeller University Council, and Disque D. Deane, Rockefeller University Trustee.

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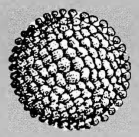
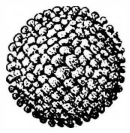


HOW TO READ "SCIENCE *vs.* AIDS"

This publication is produced to be read in two different ways. On the right side of the pamphlet are the highlights of the Seven Springs conference proceedings written in non-technical language. On the left side are easy–to–understand summaries of several key scientific issues pertaining to the AIDS epidemic. This background is intended to provide readers with the basic information necessary to help them comprehend the complex task facing scientists as they work to understand the AIDS virus and develop treatments against it.



PREFACE

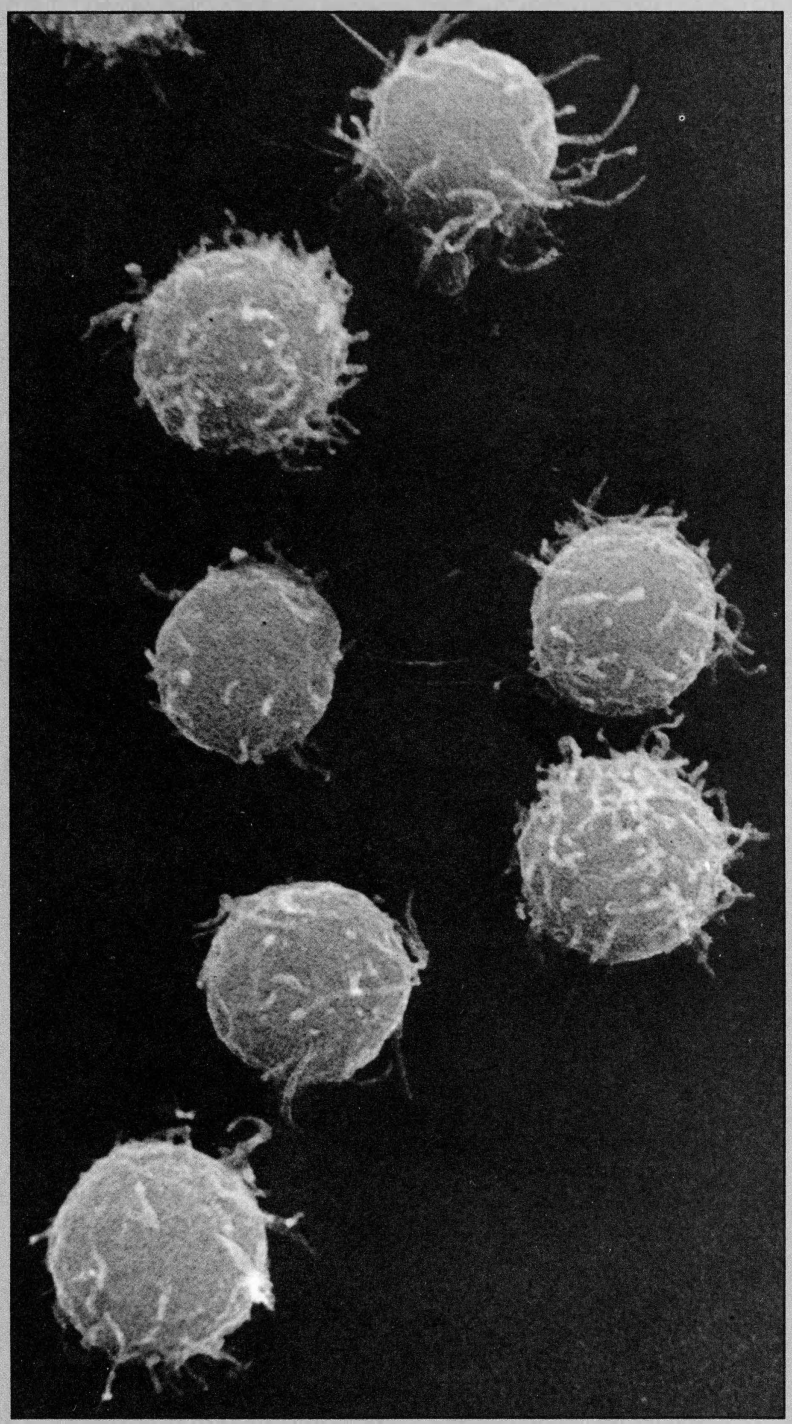
In the spring of 1988, my laboratory colleague Professor Ralph Steinman and I were hosts to a small group of fellow scientists at a meeting at Seven Springs, The Rockefeller University's conference center in Westchester, New York, a quiet enclave some forty-five minutes north of the University's Manhattan campus.

The thirty-one participants, representing a number of different disciplines and institutions, had in common that each is a leader in research that bears directly or potentially on the problem of AIDS. Our object was to spend a couple of days together, free from the intrusions of the laboratory, the clinic, and the press, sharing our ideas and the perspectives derived from our individual investigations in an attempt to assess where we are in understanding this terrible disease, and where our future efforts should be directed.

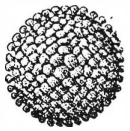
While the meeting was being planned, it was suggested to us that a report of our discussions would be of interest to many people outside the research community. We invited science writer Lois Wingerson to attend the meeting and prepare this publication. (A scientific summary of the meeting by Dr. Steinman and myself appeared in the December 1988 issue of the *Journal of Experimental Medicine*.)

We would like to express our appreciation to Rockefeller University Council member Katherine Morgan Deane, and to Rockefeller University trustee Disque Deane for their generous support of the Seven Springs project.

ZANVIL A. COHN
Henry G. Kunkel Professor
The Rockefeller University



Lymphocytes as seen with a scanning electron microscope, which magnifies actual sizes 1,600 times, and provides a high resolution view of the contours of the lymphocyte surface. However, microscopy does not reveal the chemical and functional differences that exist among lymphocytes. About half the lymphocytes in human blood carry a surface molecule called T4 or CD4. CD4-bearing lymphocytes make molecules called lymphokines (or interleukins) which help generate immunity. It is the malfunction and loss of CD4 helper cells that is so serious in AIDS.



INTRODUCTION

What we already know about the acquired immune deficiency syndrome fills thousands of pages in scientific journals, but it is not nearly enough. There is no cure in sight, and the epidemic continues to spread.

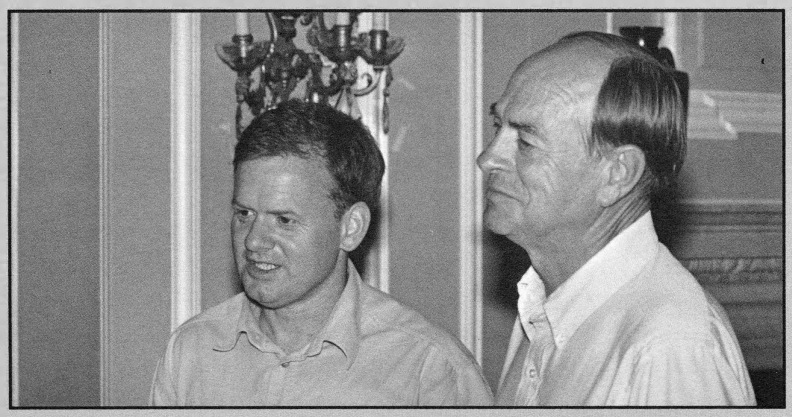
AIDS is still eluding scientists because it is an entirely new challenge—caused by a kind of virus new to medical science, infecting a complex network of human cells that is still poorly understood. Meeting this challenge demands a diverse mixture of scientific specialists, people who have had little reason to pool their expertise in the past. Now there is a most urgent reason, and in May 1988, there was an opportunity.

For two days, a small group of top AIDS researchers met in seclusion at a conference center in Mount Kisco, New York. It was an event unique in the history of AIDS research. There were no press reports, no audience, no formal presentations, no strict agenda; just thirty-two scientists and a slide projector in the small library of an old estate. Their deliberations remain as pertinent now as they were at the time of the conference.

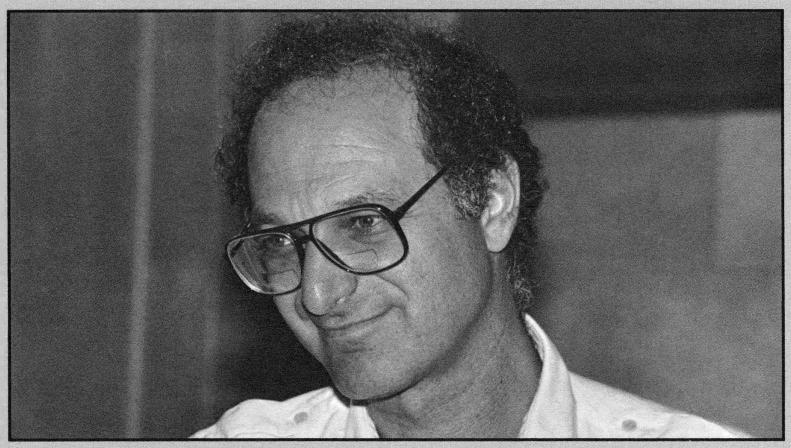
The sponsors from The Rockefeller University had invited the participants to the Seven Springs Conference Center to address a few basic questions: What do we know about AIDS, and what do we still need to learn? Which questions can be answered, and how? Which questions cannot be answered at all?

The decisions came from different vantage points. A few participants were medical researchers who encounter AIDS patients daily. Others had rarely if ever seen an AIDS patient, but spend their days studying cells in laboratory dishes. Some set their focus even deeper, on the molecules inside viruses and cells.

The retrovirologists at the conference had spent years in an esoteric endeavor, studying a class of viruses not known before 1980 to infect human beings. When the AIDS virus joined the class, the work of these scientists was suddenly pivotal. A few people in attendance had not studied AIDS at all, but had

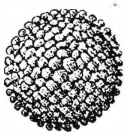


(*left to right*)
Dr. Alan Williams and
Dr. Robert North



Dr. Bernard Fields

(*left to right*)
Dr. Jay A. Levy and
Dr. Malcolm A. Martin



focused on the normal immune system which counteracts diseases inside the human body—and which the AIDS virus invades and destroys.

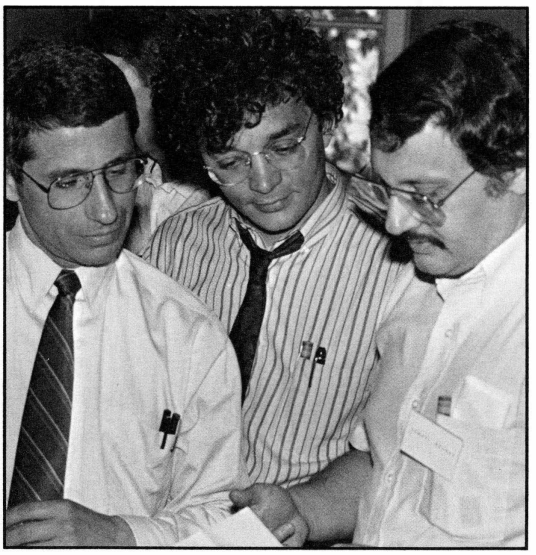
Exactly a month after the Seven Springs meeting, the chairman of a much larger gathering—an international AIDS conference in Stockholm—would tell his audience of 7,000 that the news about AIDS was "even more frightening than we have expected." Indeed, the press reports were grim:

On average, it takes about eight years after initial infection to develop the first signs of AIDS. Eight years after the first reported cases, therefore, the full impact of the disease was just beginning to be evident. It might be even worse than it appeared: In some individuals, according to a new kind of test, the virus seemed to hide out inside cells without even causing an immune response that can be recognized in a blood test. Once infected, a person's odds of survival—speaking very optimistically—may be one percent.

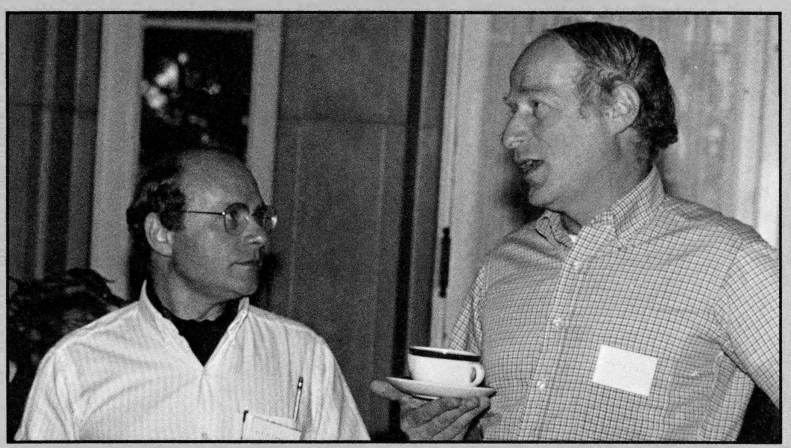
The scientists meeting at Seven Springs knew all this a month earlier, but their consensus for the future seemed less grim than the press reports that would come from Stockholm.

Much does remain to be done before the AIDS crisis is solved. That fact has a mirror image, reflected at the Seven Springs conference: Much can be done.

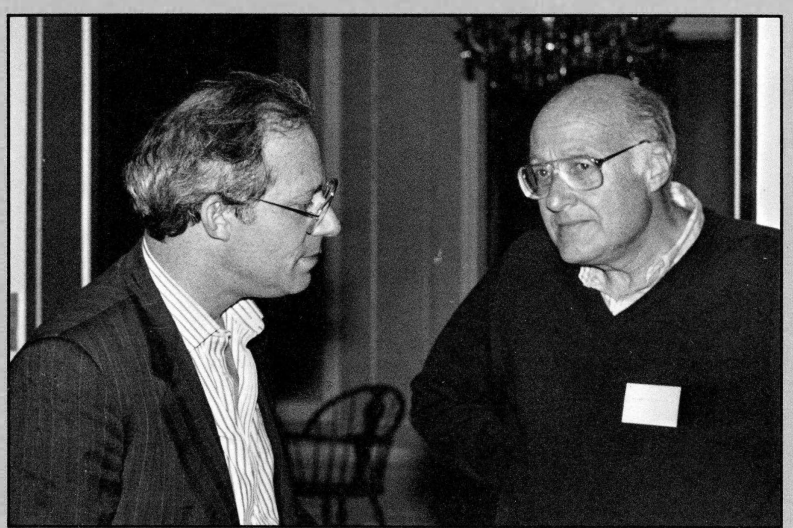
Below left: **Dr. Richard Lerner and Dr. Hilary Koprowski** ***(below right)*** **Dr. Anthony Fauci, Dr. David Klatzman and Dr. Samuel Broder**



(*left to right*)
Dr. Siamon Gordon and Dr. Irving Weissman



(*left to right*)
Dr. Alain Pompidou and Dr. Martin S. Hirsch



(*left to right*)
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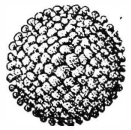

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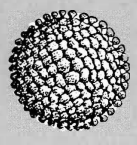

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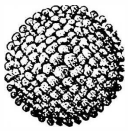

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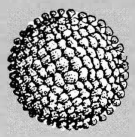

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(Photo left: l. to r.)
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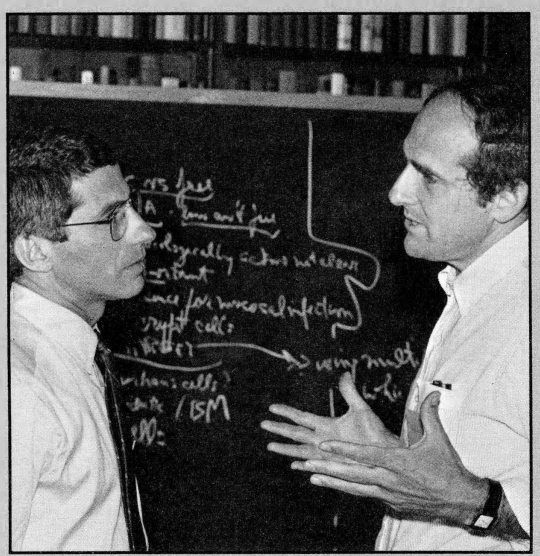
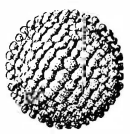
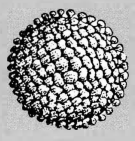


SCIENCE *vs.* AIDS

Defining the Battle Lines

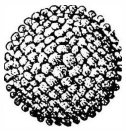




ORIGINS: OBSCURE

It is tantalizing, but probably futile, to ask how AIDS began. The AIDS virus shows similarities to some viruses prevalent in African monkeys, and one theory holds that it spread to a human being from a monkey, perhaps as recently as the second half of this century. But evolutionary studies from Japan suggest that the difference between the AIDS virus and its simian relatives dates back much farther, perhaps thousands of years. Then why did it gain a foothold as a disease probably no earlier than 1960 (to judge from studies of stored blood)?

We may never know. The same question can be asked about the microorganisms that cause bubonic plague, or polio, or rheumatic fever, or the agents that may cause arthritis or multiple sclerosis. And again there is no good explanation as to how these diseases arose, where and why, and why they may wane. The human population has a most unstable truce with viruses, which are in their own ways as inscrutable and changeable as we are.



THE SEVEN SPRINGS INTERNATIONAL CONFERENCE ON THE IMMUNOLOGICAL AND INFECTIOUS SEQUELAE OF AIDS: A SUMMARY

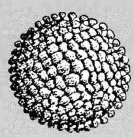
The adversary is incapable of designing strategies. It does not understand malevolence. But for everyone who thinks about the AIDS virus—including the scientists who know it best, gathered at Seven Springs—it is almost irresistible to regard the virus as a deliberately crafty and insidious enemy.

Its name describes its methods: human immunodeficiency virus, or HIV. Actually, there are two forms of the AIDS virus, HIV1 and HIV2, which have some structural differences but similar disastrous effects. At the outset, the conversation at Seven Springs centered on viral guerilla tactics: the ways in which HIV commandeers and disarms the very immune cells that ought to discover and destroy it.

In one sense, scientists already know HIV inside and out. Inside, a core of proteins and molecules of genetic material, RNA. Outside, a coat of glycoprotein, protein wrapped in sugar. (See page 42.) Researchers have no trouble recognizing HIV in the laboratory, using molecular ID tags called antibodies, which an animal's immune system will manufacture in response to HIV's presence in its blood.

In another sense, scientists know almost nothing about HIV. The details of its life history and ordinary affairs remain a mystery. It is as if the FBI had identified a terrorist group and possessed photographs, fingerprints, and a good description—but had no idea whatever of its whereabouts or movements.

HIV gives medical science its first good opportunity to study the interaction of human beings and a class of viruses previously known only to infect animals, the lentiviruses. It's a field of study with many scholars, but a history of only five years.



BRIEF HISTORY OF AN EPIDEMIC

At the outset, what we would come to call AIDS was merely a disturbing, somewhat intriguing, little puzzle. A few otherwise healthy young American men began to succumb to rare infections they had no business catching: Pneumocystis carinii, Mycobacterium avium tuberculosis, diseases of the Third World or, in the United States, of the old and debilitated.

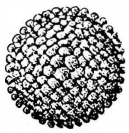
In June 1981, 152 such cases were reported in a medical journal. Nine of ten victims were homosexual or bisexual young men. Fortunately, the first suggested name—GRID, for gay-related immune deficiency—did not take root. After 1981, the number of cases would double every six months and reach far beyond the homosexual community.

Another name gained acceptance: acquired immune deficiency syndrome, or AIDS. The syndrome was defined as:

- progressive immune deficiency;
- disseminated infections such as herpes, candidiasis, Pneumocystis pneumonia;
- perhaps Kaposi's sarcoma, a tumor of tissues lining blood vessels and organs, formerly known mostly among older Italian or Jewish men, or Africans.

1981: The cause of the disease was a matter of speculation. Under consideration were autoimmunity, damage from sperm in anal intercourse, recreational drugs favored by homosexuals or, most likely, a new virus. In general, most governments and the press treated AIDS as a curiosity.

1982: Cases had now been reported from Haiti, Europe, and Africa. An outbreak of Kaposi's sarcoma erupted in Zambia. The diarrhea, fever, and weight loss common among AIDS patients were so prevalent in parts of Africa for other reasons that AIDS was initially very difficult to pinpoint on that continent.



A FREE AGENT?

At its start, the Seven Springs conference grappled with one of the most troublesome unknowns about HIV: whether the virus tends to infect cells while it floats freely in the blood, or whether it always spreads—with one exception—from cell to cell.

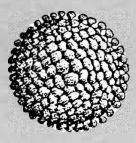
The exception is hemophilia, the inherited bleeding disorder in which blood fails to clot properly. People with hemophilia can contract HIV infections if the virus contaminates preparations of Factor VIII, which they must take to stop bleeding. This blood fraction, prepared from the concentrated plasma of many donors, is filtered finely enough to extract bacteria and whole cells, but not free viruses. The HIV in Factor VIII must be transmitted as a free agent—but was the virus freed by the plasma preparation, or was it free in the donor's plasma in the first place?

Moreover, is such cell-free passage of the virus really an exception? The answer is elusive, because the free virus is elusive. People who are infected but not yet ill are likely to transmit the virus to others, yet it is rare in their body fluids. The only body fluid where HIV is abundant in cell-free form is the cerebrospinal fluid that bathes the spinal cord and the brain—hardly a major route for passage of HIV from one person to another.

Precisely what, then, is the origin of the infection in the semen passed in sexual intercourse, or in the contaminated hypodermic needle which spreads HIV to an intravenous drug abuser? It's hardly an esoteric question, because a critical one follows: To control the spread of AIDS, must we prevent passage of free virus, infected cells, or both? And which cells?

"The definitive experiment has yet to be done," said Malcolm Martin of the National Institute of Allergy and Infectious Diseases (NIAID). "Every meeting I go to we ask the same question. No one has the answer."

Finding it will be difficult. Most AIDS patients have been infected for years before they develop symptoms. Ethically and practically, it's impossible to study the moment of infection, whether it is during intercourse or a needle prick.



1983: Intravenous drug abusers, people who have received blood transfusions since 1978, and people with hemophilia who take injections of the blood fraction Factor VIII, were added to the high-risk groups.

During the same year, Luc Montagnier's team at the Pasteur Institute identified a new virus from several patients with AIDS or with symptoms that precede the disease.

1984: Robert Gallo and coworkers at the National Institutes of Health also identified an AIDS virus (subsequently found to be very similar to Montagnier's virus). They also developed a way to grow the virus in the laboratory, and both the American and French laboratories developed tests that blood banks could use to search for the presence of the virus in donated blood.

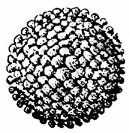
1985: Two simple, grim statistics: By year's end, eighty-three percent of the first 500 AIDS patients in San Francisco had died. Median survival after diagnosis was eleven months.

In April, widespread screening of donated blood began in the United States to exclude units contaminated with the AIDS virus.

1986: Now the virus that causes AIDS had its own monogram: HIV (for human immunodeficiency virus). In clinical trials, the antiviral drug AZT was shown to slow, but not arrest, the immune destruction in AIDS. A study of AIDS in Florida found no evidence to support the fear that AIDS might be spread by mosquitoes.

1987: Concern was rising that the risk of AIDS transmission among American heterosexuals might ultimately be as high as that being documented in Africa. The concern might be overemphasized, but it was not trivial: AIDS was now the leading cause of death among intravenous drug abusers and hemophiliacs, both groups that include many heterosexuals. Thirty percent of America's 1.1 million intravenous drug abusers are female, and half of these women have a history of prostitution.

Meanwhile, human testing of two potential AIDS vaccines and widespread use of AZT were approved for AIDS patients.



With this issue, there arose in the first minutes of the Seven Springs conference the most frustrating aspect of AIDS research: the lack of an ideal animal model. Although a virus that infects some monkeys has many similarities to HIV, and although mice have been genetically engineered to have HIV in every cell of their bodies, direct infection with HIV itself does not cause a disease like AIDS in any species other than human beings.

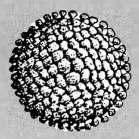
A scientist studying the common cold can ask volunteers to submit to applications of a virus in the interests of science; the consequences are uncomfortable but mundane. Obviously, such experiments are out of the question for an AIDS researcher. What then?

Recently, a virus very similar to HIV has been shown to cause a disease very similar to AIDS in rhesus monkeys, and this is extremely promising as an animal model for AIDS. But to date, most researchers have had to resort to less than optimal methods in addressing basic questions about the AIDS virus. Even with the rhesus monkey model, some questions—such as the precise mode of entry of HIV into the body—may be unanswerable.

Most of what scientists know about the AIDS virus, beyond watching the progress of the illness itself, has been gained from studying human cells persuaded to grow in laboratory dishes. How much of the "knowledge" gained in this artificial manner is irrelevant? (As one conference participant put it: "A human being is not a tissue culture!") Without a good animal model, many theories about HIV will remain nothing more than best guesses.

"We have to be honest with each other," interjected another participant early during the first session. "Instead of kidding ourselves, somebody ought to take down the message that the one thing we do know is that we don't know."

"We do know things about the pathway," countered the session chairman, Bernard Fields of Harvard Medical School. "We know some of the genes. We often don't know why the virus goes in a certain pathway. There are enormous gaps in the complexity of the biology. In so many places, we can't [obtain] rigorous biochemical proof of a mechanism. But we do *not* know nothing!"



1988: Eight years after AIDS was first recognized, it is still synonymous with death.

By the time of the Seven Springs Conference in May, HIV had spread to more than 62,000 Americans, including 39,000 homosexuals, 11,000 drug abusers (and 5,000 people who fall into both categories), as well as 600 hemophiliacs and 1,000 children. Public health experts predict that by the end of 1993 some 450,000 Americans will have AIDS.

How bad an epidemic is AIDS? For comparison, every year in the United States, approximately:

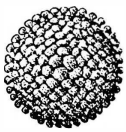
1,000,000 people *are diagnosed with cancer*

900,000 people *have their first heart attack*

150,000 people *develop lung cancer*

50,000 people *die in highway accidents*

Worldwide, more than 81,000 AIDS cases have been reported in 133 countries. An AIDS expert at the World Health Organization says the real incidence is probably twice as high. By 1990, it is estimated, more than a million people will suffer from the disease.



MEANS OF ENTRY

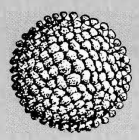
However it gains access to the body, HIV has to enter a human cell so that it can reproduce. It must invade the cell's genetic machinery and use it for its own replication. Which cell does it choose?

Evidently, HIV most often gains access to certain of the white blood cells: the helper T cells, the monocytes, and macrophages. All of these have important functions in the immune system (see p. 30). New studies suggest that macrophages may be HIV's first victims inside the body, the refuge where HIV hides out and multiplies even before provoking an immune reaction.

At the outset, when AIDS first appeared, researchers focused on HIV's tendency to infect and kill a certain subpopulation of the T cells of the immune system. Alan Williams and Don Mason of Oxford University had recently identified a unique molecule called CD4 on the surface of helper T cells, and they used it as a label to distinguish these from other kinds of T cells. As Mikulas Popovic of the National Cancer Institute (NCI) reminded the conference, very early AIDS experiments showed that antibodies which bind to CD4 could prevent the virus from infecting T cells in the laboratory. When this emerged, it seemed obvious to assume CD4 was the lock that HIV could open to enter a T cell.

The revelation that macrophages could also be infected came later, and brought with it serious implications. Cells of the macrophage type, and their blood-borne relatives the monocytes, are everywhere. Macrophages take up long-term residence in the lining of organs such as the lungs, the kidneys, and the brain. In all these places, apparently, they are both housekeepers and nurturers; they produce messenger chemicals and also take up and degrade cellular debris. If the virus could infect a macrophage-type cell, it seemed there was almost no limit to where it might lodge.

Although cells of the macrophage variety do have some CD4 molecules on their surfaces, there's a much simpler (though as yet unproven) way for HIV to enter: endocytosis, the natural process by which macrophages engulf and destroy foreign matter and damaged cells. Does HIV usually infect



AT RISK

Male homosexuals: Infected with HIV via anal intercourse.

Intravenous drug abusers: Infected by sharing syringe needles with someone carrying HIV in his or her bloodstream.

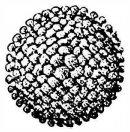
Hemophiliacs: Infected from contaminated injections of Factor VIII, taken to avoid uncontrolled bleeding.

Transfusion recipients: Infected by receiving donor blood contaminated with HIV. Less than two units of donated blood in a thousand are found to be infected by HIV, and these units are discarded. Because some researchers think that the virus can briefly hide out in cells without provoking antibodies, it has been theorized that some 460 people each year might receive infected blood that escapes detection.

Infants of infected mothers: The AIDS virus reaches them in utero, during birth, or perhaps while nursing.

Sexual contacts of AIDS carriers: The virus apparently can be transmitted during ordinary heterosexual intercourse. The actual risk of catching AIDS heterosexually, especially from a woman, is unknown and highly controversial.

There is no evidence to support any suggestion that AIDS can be spread by casual contact, kissing, or insect bites.



macrophages via the CD4 receptor, or is it swallowed in the normal course of events?

That leads to another important unknown. If HIV often infects macrophages by endocytosis, how can it be stopped? Should a macrophage be prevented from doing its regular job?

"We can't yet answer what it is about the macrophage that makes it win or lose against any virus," observed Dr. Fields. "This is a critical question that is a gap in our information."

The latest type of white blood cell to be implicated as a host for HIV is the dendritic cell. A principal function of these cells is to act as accessory to the normal function of CD4-positive cells. If infected, dendritic cells may interfere with T-cell function, or transmit the virus.

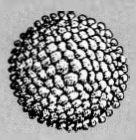
Another troubling puzzle is where, inside the vagina or rectum, the AIDS virus first lodges after being transmitted during sexual intercourse. Can an intact vaginal lining be infected?

The cells on the epithelium, the surface lining, of the vagina and rectum don't possess the CD4 molecule, the lock that HIV can open. Does HIV infect these cells through another opening (in which case many drugs and vaccines proposed to halt the infection are useless)? Or must there be some minor injury during intercourse, which gives the virus access to underlying cells that do bear CD4 on their surfaces?

The evidence is mixed. As Jay Levy of the University of California, San Francisco, pointed out, studies from Africa show that the risk of heterosexual infection appears to be increased in the presence of genital ulcers or of two venereal diseases, chancroid and chlamydia. All of these factors could bring immune cells to the surface, where the virus could reach them.

On the other hand, both Dr. Levy and Michael Oldstone of the Scripps Clinic and Research Foundation displayed pictures showing evidence of HIV infection in certain cells lining the intestine and rectum. But are these really macrophage cells that have taken up residence in the gut, where macrophages are very common? Photographs alone won't resolve the issue; further study is needed.

"Some of these questions you'll never be able to answer in humans," commented Martin Hirsch of Massachusetts Gen-



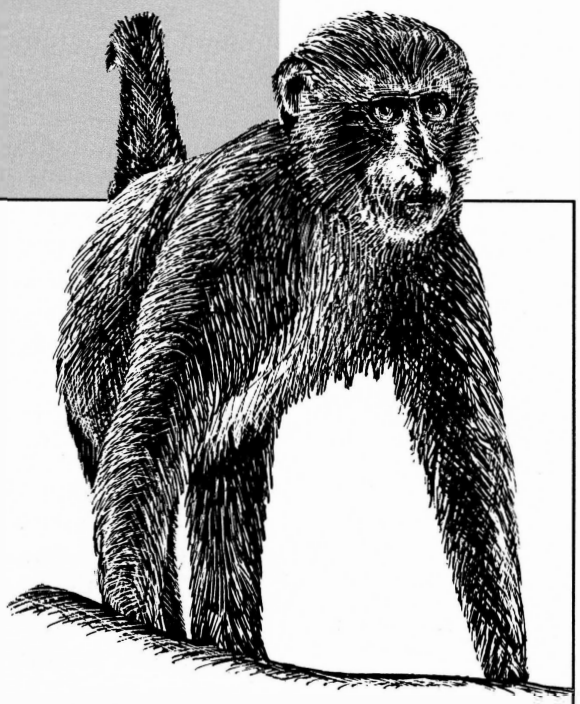
WANTED: ANIMAL MODEL

Acquired immune deficiency syndrome is unique to human beings. This simple fact, more than anything else about AIDS, has slowed efforts to combat the disease.

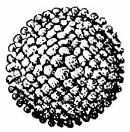
CHIMPANZEES can be infected with HIV, but they do not proceed to develop immune deficiency.

RHESUS MACAQUE MONKEYS can be infected with the simian immunodeficiency virus, SIV, which is closely related to HIV in its genetic constitution and its effects. Monkeys infected with SIV may remain persistently infected or may die of problems that parallel AIDS. The rhesus macaque is probably the best animal model for AIDS available today, and is in great demand. However, it is too soon to say how much of what may be learned about SIV will translate directly to AIDS.

GENETICALLY ENGINEERED MICE, created at the National Institutes of Health, contain copies of the AIDS virus in every cell. They are the only mammals other than humans that get sick from the effects of HIV.



CONGENITALLY IMMUNODEFICIENT MICE have been implanted with fetal human thymus, liver, spleen, and lymph nodes in experiments by Dr. Irving Weissman and Dr. Michael McCune at the Stanford University School of Medicine. As Dr. McCune reported these experiments during the Seven Springs meeting, only four of these mice had been inoculated with HIV, and it was too soon to say whether they might go on to be infected with HIV or develop a disease like AIDS.



eral Hospital. "You can't study the individual [infectious] episode."

"That question, we can't answer in man," agreed Hilary Koprowski of the Wistar Institute. "We can reconstruct skin in tissue culture. We can do a lot of experiments and show, I'm sure, that the virus will get in. But that doesn't mean it occurs in vivo."

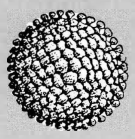
It might be possible, however, to learn why some individuals remain infection-free despite years of regular sexual contact with a HIV-infected partner. There could be elements of the fluid environment in the human vagina or rectum that inactivate the virus. These may vary from person to person. "This area has not been looked at very much," said Dr. Fields. "It's drudgery to pull this all together, but it's probably worth it."

VIRUS FACTORIES

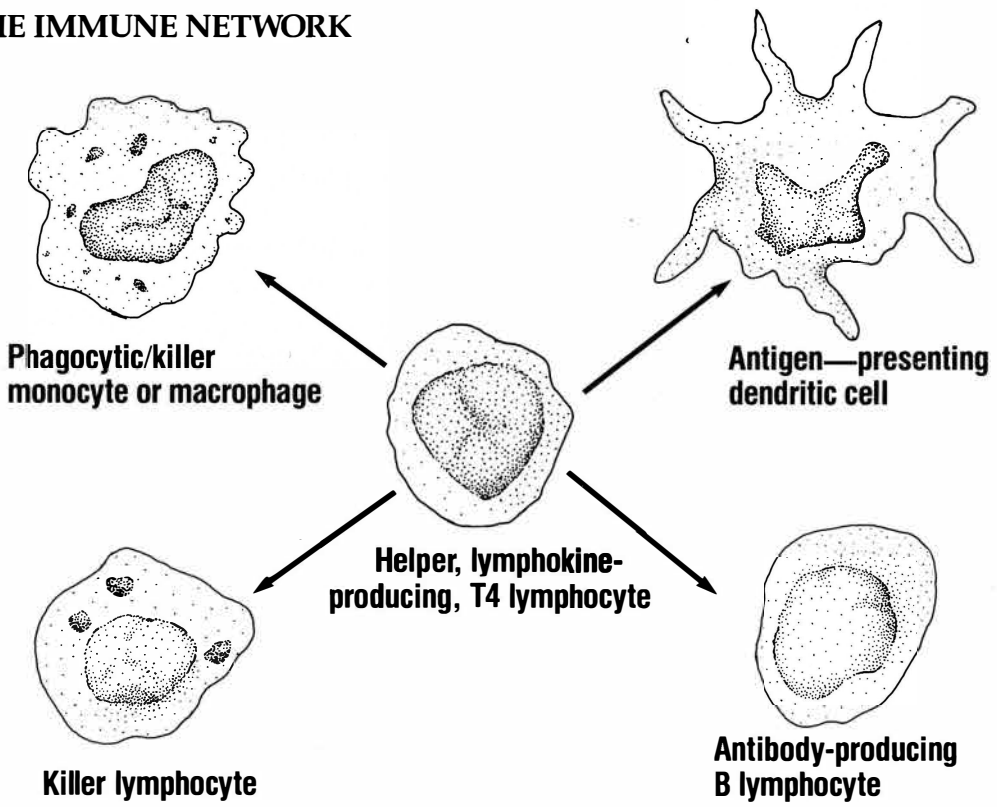
The fate of a cell infected by HIV depends on its cell type. It appears that HIV ultimately kills helper T cells, but it often seems to spare a cell such as a macrophage in order to use it as a factory, to assemble new versions of itself. Much of what is known about HIV comes from studies of T cells; speakers at Seven Springs agreed on an urgent need to repeat these experiments using macrophages and monocytes.

Inside such a cell, HIV probably behaves much like the other retroviruses that have been studied in animals (see p. 28). It allows the cell's enzymes to unzip its coat, and proceeds to use its own enzymes to translate some of its RNA genetic message into DNA. Then the DNA version of the virus's blueprint somehow elbows its way into the cell's own genetic material, where it may sit silently for years.

What rouses it into action later, ironically, is probably the cell's response (in its normal role within the immune system) to the presence of a further infection. In laboratory dishes, HIV can be provoked to replicate when "quiet" infected cells are triggered into action by some immune stimulus. For an AIDS patient, the first sign of the disease is often the appearance of a major infection of another kind.



THE IMMUNE NETWORK

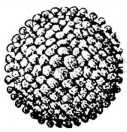


When a healthy person gets an infection, he or she can mount an astonishing number of different kinds of responses to the insult. Here are examples of the reactions that can be set off by a microbial invader such as the measles virus:

Like internal sanitation engineers, the macrophages wipe up the dead infected cells, destroying and breaking up the viruses inside. In the process, the macrophages present viral proteins on their cell surfaces in a form that T cells can recognize. Dendritic cells also can take up antigens in tissues where they are deposited. Their job is to tell the T4 cell that antigen is present, and to induce the T4 cell to grow and make lymphokines.

Activated helper T cells secrete soluble factors, known as lymphokines, which halt the replication of virus and spur the maturation of other lymphocytes into killer and more helper T cells. Cytotoxic T cells kill virus-infected cells. Helper T cells boost the action of phagocytes, as well as of the B cells that create antibodies, soluble Y-shaped structures that recognize foreign proteins such as parts of a virus's coat.

Gradually, after several weeks, a population of memory T cells arises, which preserves the ability to recognize the defeated virus, should its relatives ever return.

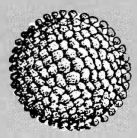


Once the cell is activated, the viral DNA inside it begins to copy itself and to assemble new AIDS viruses. These collect inside the surface of the infected cell and eventually "bud" out, wrapping themselves in part of the cell's glycoprotein surface before they pull away.

In laboratory dishes, escaping viruses may pepper the surface of an infected T cell with holes; it appears to explode. This makes a dramatic photograph, but is it clinically important? Just as easily, the virus can spread directly when an infected cell fuses with an uninfected one—or, in fact, with a large number of other cells—to form "giant cells" or syncytia. Such cell fusion is a neat explanation for some of the clinical phenomena seen in AIDS patients. Whether cell death or cell fusion is more important to the immune devastation of AIDS is still unclear.

What is clear is the resulting devastation to the immune system. Its routine business requires some cells to touch and aggregate, and others to travel long distances through the blood or lymph canals, often homing in on the organs where immune cells are created or wait at rest—the lymph nodes, the spleen and thymus, and the bone marrow. Any one of these cells, infected with HIV, could transmit new viruses to many other cells.

It's the worst nightmare of an intelligence chief: One agent turns, and before long the entire system is corrupted. Nobody knows until it's too late.



AIDS AND SOCIETY

If AIDS presents daunting problems to science, its challenge to society is equally complex—and the reaction has often been distressing.

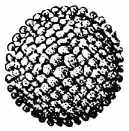
The blind eye: In the United States, the most frequent victims of AIDS are members of outcast groups: homosexuals and drug addicts, and increasingly a third group, prisoners. Compared to the polio epidemic of the 1950s, which spread across a diverse segment of American culture, it was, at least initially, far more difficult to raise official and public interest (not to mention funds) to combat the AIDS crisis.

The cold shoulder: Unlike polio in the 1950s, AIDS appears to be unequivocally fatal (until such time as a cure is found). Many of the unaffected react with fear and hostility. Children with AIDS have been stigmatized by adults; in one case, a young AIDS victim's house was burned down. Many health professionals—doctors, nurses, dentists, even morticians—avoid contact with AIDS victims. Surveys show that many who do not shun them fear for their own lives.

The pointing finger: Society grapples with the competing problems of discrimination and the need to know. To monitor and characterize the epidemic, it is urgent to identify which people have been infected. AIDS testing is already routine in the US military. Many public figures support mandatory AIDS tests for various other groups such as immigrants, prisoners, and marriage applicants. People infected with HIV face two fights: the fight against the virus, and the fight against discrimination.

The underclass: In America, AIDS is most common in the lower socioeconomic classes, and tends to draw health care resources (already scarce) away from poor people who have other problems. As a result of AIDS, public hospitals suffer shortages of antibiotics, beds, nurses. Whole wards are taken over by infants with AIDS born to drug-abusing mothers; not enough foster homes can be found.

The Third World: In parts of Africa and South America, AIDS cripples an already weak health-care system. That weakness contributes to the spread of AIDS in turn, because blood testing is less common than in the United States and Europe, and hypodermic needles are used more than once.



ONE OF A THOUSAND

How this could happen—how HIV can, in time, completely eradicate the immune system—is a baffling issue that arose again and again at Seven Springs. "The kinetics [rates of progress] of the immune destruction is really a black box," said Samuel Broder of the NCI. "Many things about the clinical course are very complicated and difficult to understand, including the initial infection which does not lead to a sudden, fulminant T-cell depletion."

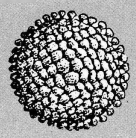
The latent periods—extended episodes of general health despite HIV infection—are characteristic of the disease. The virus may seem to be hiding out, because no one is aware of it, but is it really inactive?

In the laboratory, resting T cells or monocyte/macrophages appear relatively resistant to infection. And once infected, monocytes will not release new viruses without being activated by an immune-system stimulator, noted Anthony Fauci of NIAID. But are these latent cells an artifact of the laboratory?

Dr. Martin raised the question of what is a truly resting cell. The virus itself carries substances that may activate an immune cell to respond as if it had detected a new infection. If such a cell is already heavily infected with HIV, the presence of these substances in themselves may provoke the manufacture and release of new viruses from the cell.

"My own feeling is that we need to differentiate a single cell or a population of cells from the whole individual," remarked Robert Gallo of the NCI. "In the whole human, there is no time when you do not find expression [production of new viruses] in some cell. It may be very small at times, but in a pool of 100,000 cells, you almost always find some expressing the virus."

On average, noted Dr. Fauci, at most one of every 1,000 white blood cells in an AIDS patient appears to be infected by HIV, according to the most sensitive test. Only one of every 10,000 or even 100,000 is actively producing new viruses. Yet even early in the disease, while a patient appears to be well, the population of T cells steadily declines. It's very puzzling.



CLINICAL COURSE

The course of AIDS varies widely from patient to patient. Only one feature seems invariant: fatality.

The incubation period of AIDS—the interval between infection with the virus and onset of serious disease—is about eight years among adults. Someone infected by the virus may notice a flulike episode and a period of weakness and lethargy shortly after infection, but will recover to remain "healthy" for some years.

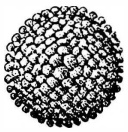
After a brief period, people infected with HIV do develop an immune response to the virus, including the production of antibody molecules that recognize many constituents of the virus including its protein coat. These antibodies allow the presence of the virus to be documented, but they do not stop the disease.

During the relatively long latent period, a part of the patient's immune system begins to deplete rapidly. In particular, certain immune cells known as T4 helper cells, important in marshalling the immune system to fight infections, decline precipitously. The mechanism underlying the loss of T4 cells is still not clear.

At the same time, probably soon after the infection, many patients begin to experience psychological problems such as apathy, lack of coordination, and difficulty in completing ordinary tasks. These problems may be subtle, or may progress to frank dementia with alarming speed. For eighty percent of patients with signs of brain infection, full-blown dementia develops within a year.

Sooner or later the immune depletion also has consequences. Many patients develop Kaposi's sarcoma, a disfiguring and disseminated cancer primarily of the skin. Interestingly, recent studies suggest that the incidence of Kaposi's is declining among Americans with AIDS.

A more significant aspect of the final decline in AIDS is the relentless progression of "opportunistic" infections, attacks of illness due to fungi or bacteria that a healthy immune system could control without trouble. (The most prominent is Pneumocystis carinii pneumonia, a lung infection formerly most common among cancer patients and transplant recipients who are weakened by immunosuppressive medications.) The or-



"Those of us who follow AIDS patients for years see them completely run out of T cells," he said. "We still haven't explained how you ultimately, completely, run out of T cells. I've been in many discussions, and this has never been resolved."

Nothing in medical history appears relevant to the question. The scientists at the conference searched for a good analogy that might provide some explanation: Organ transplant patients treated with immunosuppressive drugs? People with the autoimmune disease myasthenia gravis, who have had the thymus (the organ where T cells arise) removed as a treatment? Bone marrow donors? None of these people completely run out of helper T cells, as an AIDS victim inevitably does.

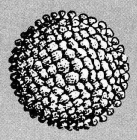
There are a number of possible explanations. Recent studies show that HIV becomes more virulent (more infectious and more deadly to cells) as the disease progresses. It is also possible that the immune system contributes to its own destruction, by killing infected cells.

The fusion of infected cells with uninfected ones is another attractive explanation. Both in lab dishes and in monkeys, giant cell agglomerations called syncytia arise in the presence of HIV. In the laboratory, these syncytia die about a day after they form.

However, the syncytia that appear under laboratory conditions are arising in clones of T cells that have grown up from a single ancestor blood cell and may be only distant descendants of the cell that once traveled in a human's bloodstream. Who can know how they may have been altered in the process of adapting to laboratory conditions? Depending on the conditions in cell culture, noted William Haseltine of Harvard Medical School, scientists can "push" HIV-infected cells to either form syncytia or swell and burst.

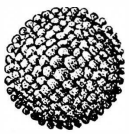
"There's a very complicated, dynamic situation going on," he said. "There are a whole series of factors to consider, which will determine if you see syncytia, cell death, or continuously virus-producing cells."

Medical researchers rarely see large numbers of syncytia in the lymph nodes or spleens of patients. Even if syncytia do form at some stage of the disease, it is not obvious when or where to look for them. Are they really important in the disease?



ganisms that cause these secondary infections often invade the same kind of immune cells infected by the AIDS virus. It is their presence in the body, not that of HIV, that is ultimately fatal.

If AIDS is likened to a guerilla campaign, AIDS viruses would merely be the forces that disarm the defending army. Other agents later overrun the invaded territory, to devastate and destroy it.



HOMING IN ON THE SOURCE

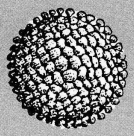
The explanation for the depletion of T4 cells may not lie in the T4 cells at all, but elsewhere in the network—in the cells that nourish them as they develop and interact with them later in their lives. A virus that infects a macrophage may thus be far more deadly than one that kills a T cell.

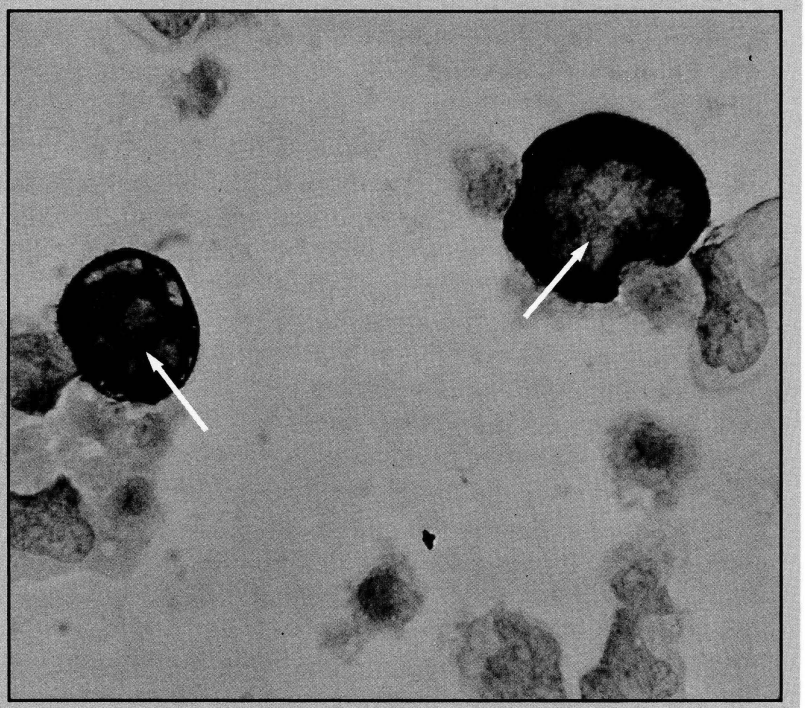
Ralph Steinman of The Rockefeller University pointed out that traveling immune cells, called dendritic cells, only recently recognized to be infectable with HIV, may carry HIV back to regions in the spleen and lymph glands where T lymphocytes are often first stimulated during an immune response. HIV-infected dendritic cells conceivably could malfunction, thus reducing the efficacy of the helper T cell response; or the infected dendritic cells might transmit HIV to the T cells with which they must interact physically to induce immunity.

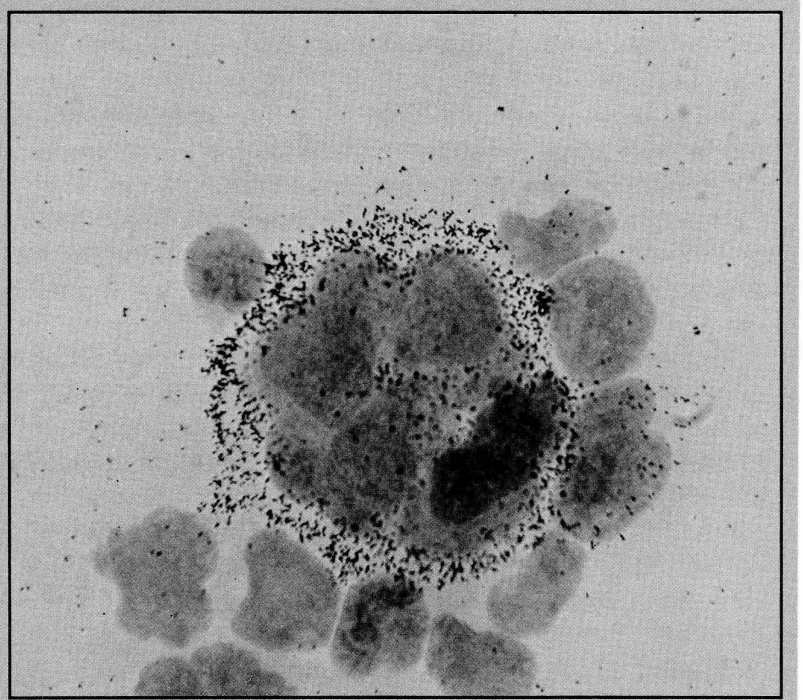
Cells lining blood vessels in many organs, added Siamon Gordon of Oxford University's Sir William Dunn School of Pathology, are members of the macrophage lineage. They may also be infected and then serve as widely distributed reservoirs of HIV.

In the bone marrow, he went on, a form of mature macrophage with many fingerlike projections sits at the center of developing monocytes, apparently nourishing them. They're CD4-positive, presumably they're infectable, and they sit close to other cells which can be infected, Dr. Gordon said, making them "an incredible opportunity for the secondary spread of the virus."

He presented a very persuasive scenario for the means by which an AIDS virus might rapidly spread its descendants across an entire network of immune cells, and it is tempting to take it for granted that this is how the infection works. But the mechanism is still only a theory. Since the first report in 1986 showing that HIV could infect monocytes inside an AIDS patient, there has been very little further direct evidence—clinical evidence—that cells of this class are infected outside the brain.



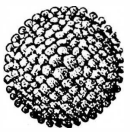




Two views of how scientists identify cells that are producing the HIV virus. The top photo illustrates a method called immunocytochemistry. An antibody (immuno) to a component of the HIV virus is isolated from infected patients, or it is made in mice using the monoclonal antibody approach. The antibody is then added to a preparation of cells (cyto) here shown by the shady oval profiles, each of which is one cell nucleus. The antibody is coupled to an enzyme which then chemically produces a colored product. Two antibody labeled cells, in other words, cells carrying HIV protein, are arrowed.

The bottom photograph illustrates a method called in situ hybridization. In this, molecular biology is used to make a complementary copy of the HIV messenger RNA, which must be made to translate the virus's genetic information into HIV proteins (see p. 42). The copy is modified with radioactive atoms so that it can be used to detect HIV messenger RNA in any test sample. When the radioactive probe finds the message of the virus in the cell, many black spots are produced over the cell.

Photographs supplied by Drs. Harm Bos and Erik Langhoff of The Rockefeller University.



IN THE BRAIN, ANOTHER PUZZLE

Early in the infection, the AIDS virus infiltrates the brain. Even before the immune deficiency arises, said Dr. Broder, many AIDS patients begin to show neurological abnormalities, both in brain scans and in psychological tests.

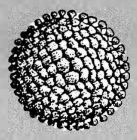
No one knows exactly how HIV reaches the brain. Infected T cells or monocytes could carry it there. Inside the brain, HIV infects macrophage-class cells known as microglia, which normally may perform a nursing function—perhaps nourishing brain cells or clearing away spent chemical signals. Whether or not HIV infects other kinds of brain cells is a matter of debate.

The brain damage that results can sometimes be reversible: AIDS dementia can recede among patients taking the antiviral drug AZT. And the physical damage is minor, he added. Pathologists often remark at autopsy that the visible damage is inappropriately slight in the brains of patients who were severely demented before death.

The mysteries are amenable to study. The products of macrophages of the nervous system, and their effects on the brain, "constitute a key area for research," said Dr. Fields.

Another fertile area for study is the difference between the people infected with HIV who feel healthy and those who are beginning to succumb. For example, Dr. Hirsch reported his studies of asymptomatic AIDS patients from Greece and the United States. When they felt healthy, their white blood cells were able to kill about half of their own HIV-infected cells in a laboratory dish. But when they developed Pneumocystis pneumonia, these responses (as well as other immune functions) dropped dramatically. When they took AZT, the laboratory activity of their white cells revived.

Myron Essex of the Harvard School of Public Health also described his studies of people who are infected with HIV but appear well. Those who did not progress to develop AIDS (or the related syndrome ARC) during the two years of this study had an intriguing property in common: They produced antibodies against a particular small region of the HIV coat protein. Would this explain how people remain healthy during infection? Could it perhaps form a basis for treatment?



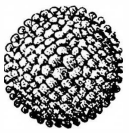
COUNTERATTACKS

Social measures: By 1985, all blood samples in the United States were being tested for evidence of HIV infection. This has led to a great decrease in the risk of transfusion. "Safe sex"—the use of condoms and selectivity in choosing sex partners—can also reduce the risk. In San Francisco, a massive public education project dramatically reduced the proportion of gay men testing positive for HIV infection during the course of four years. Signs of AIDS infection were detected in more than one in five male homosexuals newly tested for the antibody in 1982. The prevalence dropped below one in a hundred gay men tested in 1986.

Vaccines: The AIDS virus itself cannot be used as a vaccine because it is invariably fatal. A weakened or killed virus, as used in some influenza and polio vaccines, may be too risky to use for vaccinating healthy people (although Jonas Salk, developer of a polio vaccine, is trying it as a way to boost the immune response in people who already have AIDS). Two artificial AIDS vaccines are currently under testing in human volunteers in the United States. Both use a virus harmless to humans, which is genetically engineered to contain a small fragment of HIV's protein coat as an immune stimulus.

In March 1987, a French researcher announced that he had injected a small group of volunteers in Zaire, and himself, with a "vaccine" consisting of inactivated virus. They all developed antibodies against the virus, and some also showed an increase in the activity of T lymphocytes. All three proposed vaccines provoke the production of antibodies, but it is too soon to say whether these can prevent the disease.

In England, testing has begun on another sort of vaccine, which incorporates a protein from the virus's core (rather than from its outer coat).



WHY NOT A VACCINE?

People do not succumb to the human immunodeficiency virus simply because their bodies fail to recognize its presence. Very early in the infection, AIDS patients do react to the virus. They naturally produce antibodies, well-characterized proteins that recognize HIV and should lead to its defeat.

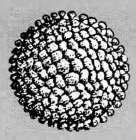
In most cases, these antibodies, which are known as neutralizing antibodies, easily conquer an infection. In fact, many vaccines work by intentionally provoking their creation. Vaccines provide the immune system with an innocuous first introduction to a pathogen (as sometimes do reminders that are known as booster shots). If the agent appears later as an infection, the immune system can respond quickly. It responds first with antibodies, which both initiate an antiviral response and remember the virus for years.

But the first antibodies against HIV are not as good as the name they are given. They fail to neutralize the virus, either in cell culture or, evidently, inside the body. This confounding fact provides one main reason why the best strategy against a viral disease, a vaccine, proves unexpectedly elusive in the case of AIDS.

In 1984, when HIV was isolated, the head of the United States Department of Health and Human Services cheerily predicted there would be an AIDS vaccine in two years. Four years later, in a quick show-of-hands vote at the Seven Springs conference, only about half of those present felt there will be an AIDS vaccine in ten years, and no one would bet on five years.

The AIDS virus presents a daunting challenge for vaccine developers. Its protein coat is heavily masked by complex sugars. It mutates rapidly, giving descendant viruses an opportunity to escape an immune response designed against the parent virus. Furthermore, the virus tends to hide out inside cells where an immune system can't see it under any circumstances.

History provides some discouraging precedents, Dr. Koprowski remarked as the discussion of vaccines began at Seven Springs. Progress against livestock lentiviruses—which



RETROVIRUSES

Nearly 80 years ago, an unusual visitor came to the laboratory of a young cancer researcher, Peyton Rous, at The Rockefeller Institute for Medical Research (now The Rockefeller University). A chicken breeder arrived at the lab, accompanied by his hen, which had a malignant tumor called a sarcoma on its leg.

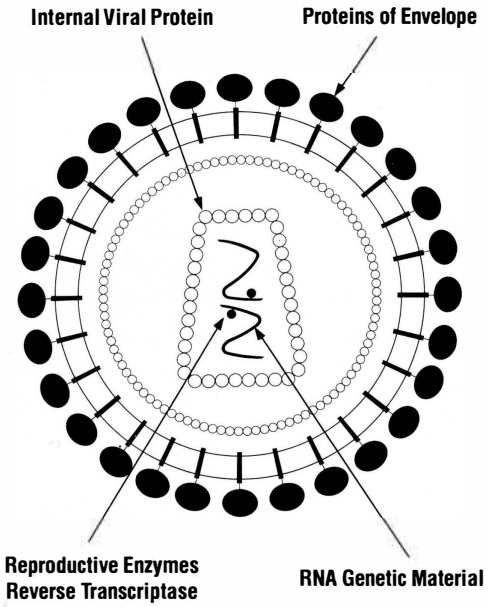
Rous removed the tumor, filtered off the cells, and injected other chickens with the remaining liquid. They developed similar tumors. Rous decided that an agent too small to be seen under the microscopes of 1911, perhaps a virus, was at fault. His idea was greeted with resounding silence at the time. But he was right.

Some viruses that cause human disease, such as the agents that cause chicken pox or hepatitis, are made of the same genetic material that acts as a blueprint for human cells: deoxyribonucleic acid, or DNA. These viruses act by inculcating themselves directly into the cells they infect. Many tumor viruses, such as the Rous sarcoma virus, are made of an alternative kind of genetic material: ribonucleic acid, or RNA.

When human (or chicken) cells divide, they copy their DNA into more DNA. Genetic information in the DNA is transcribed into messenger RNA, which then is translated into protein. The protein carries out the function of the gene. In animals, the flow of information is DNA to RNA (and then to protein) and not the other way around. So how could an RNA virus ever proliferate in an animal cell?

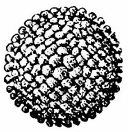
The puzzle was solved only in 1970, when David Baltimore of MIT and Howard Temin of the University of Wisconsin discovered an enzyme, reverse transcriptase, peculiar to RNA viruses. These viruses use it to translate their genetic code "backwards" into DNA. Thus their name: retroviruses.

Retroviruses became a mainstay of biotechnology because they allow scientists to isolate genes, by starting from readily accessible messenger RNA in cells, and also to insert new genes into cells. Many retroviruses, in addition to the chicken Rous sarcoma virus, were found in animals. But to medical science they were nothing more than interesting curiosities until 1980, when Robert Gallo and his coworkers at the NIH reported that a retrovirus could cause a rare leukemia in humans. Then, in 1983, the retrovirus that causes AIDS came to light.



The genetic material of the HIV virus, diagrammed here with wavy lines, encodes reproductive enzymes, internal virus proteins, and other envelope proteins. Their assembly into new virus particles is diagrammed on page 44. Additional HIV genes that play a regulatory role in virus production are diagrammed on page 46.

Every vertebrate species studied for retroviruses has been found to be susceptible to at least one. Some retroviruses are apparently harmless, a number of them cause cancer, and others cause a wide range of noncancerous degenerative diseases such as AIDS.



are in the same class of pathogens as HIV—has been negligible, despite a great deal of money and effort, because of the same properties that are outsmarting the AIDS researchers.

The visna virus of sheep, a very close relative of HIV, mutates so rapidly that (as a population) it slips away from a vaccine undaunted. Another sheep lentivirus that causes a neurodegenerative disease known as caprine arthritis encephalitis provokes so-called neutralizing antibodies that do not neutralize inside the animal at all.

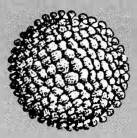
On the other hand, Dr. Koprowski noted, veterinary vaccines against rabies and herpes can protect animals without provoking a trace of neutralizing antibodies. Presumably the vaccine elicits other forms of protection such as T-cell-mediated immunity. It's easy to test such vaccines in animals, by taking the chance they will work. Testing a vaccine against a uniformly fatal disease using human volunteers is another matter entirely.

Nonetheless, some human volunteers are already part of studies directed at developing an AIDS vaccine. In Zaire, Daniel Zagury of the Pierre and Marie Curie University of Paris has injected himself and a small group of healthy volunteers with a prototype vaccine made by genetic engineering. It incorporates a portion of the HIV coat (a protein called gp160) into vaccinia, the virus used to vaccinate against smallpox.

Afterwards, Dr. Zagury and his volunteers developed an antibody response against HIV, and in laboratory tests, the white blood cells of some of those who had received the vaccine were able to kill cells infected with the virus. These responses have lasted for longer than a year. But would they protect any of the volunteers against a real HIV infection? The only way to arrive at an answer would be by infecting vaccinated individuals with live virus in order to determine if the vaccine is protective.

In the United States, the Federal government and six medical centers are recruiting volunteers for different studies using direct injection of viral coat proteins, either gp160 or its subunit gp120, as a vaccine. The question is whether these vaccines will provoke any better response than the neutralizing antibodies that fail to protect AIDS patients in the first place.

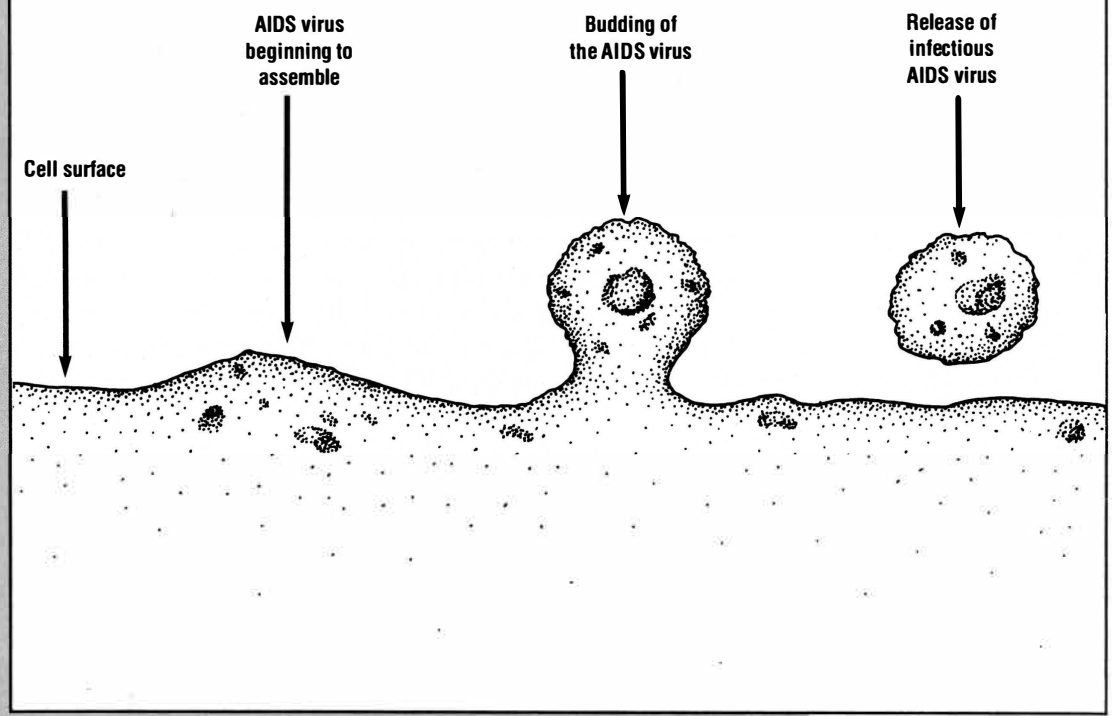
These misnamed neutralizing antibodies bind to part of the viral protein called gp120, which is apparently the protein

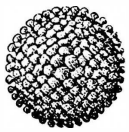


OPERATING PLAN FOR A RETROVIRUS

Like all viruses, retroviruses consist of a genetic core, which contains two single strands of genetic material wrapped in protein, and an outer coat or envelope, a membrane studded with glycoproteins. An infecting retrovirus also carries along its own enzymes. (See p. 42.)

Having infected a cell, a retrovirus uses its own enzymes to reverse transcribe its *own* genetic material into DNA and insert it into the cell's genetic sequence. At some later point, it uses the host's machinery, under the direction of viral signals, to duplicate its genetic code and to transcribe this code into messenger RNA and protein so that it can assemble new copies of itself. Somehow, the genetic material and core proteins of the new viruses congregate near the cell's envelope and bud outwards to form new viruses.





"key" on the virus coat that initially fits the lock of the CD4 protein on an immune cell before the virus opens it and enters. Parts of this gp120 protein represent one of the few sites on the virus coat that are conserved; that is, remain unchanged by mutation.

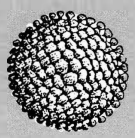
Evidently, conserved sites are preserved by nature because they are integral to the function of the virus. In order to create a workable vaccine, it's desirable to use such conserved regions of a viral coat as a target so that the virus cannot escape the vaccine by mutating.

It's likely, speakers at Seven Springs predicted, that vaccines against this conserved region of gp120 will not be completely successful in themselves. First of all, there are theoretical reasons why they may have untoward effects. In a sense, the neutralizing antibodies that bind to the conserved fragment of gp120 may partly resemble CD4 itself, much as any lock a key will open must resemble any other lock it will also open. Will large numbers of antibodies raised against a viral protein that binds to a T cell interfere with molecules that should do so normally?

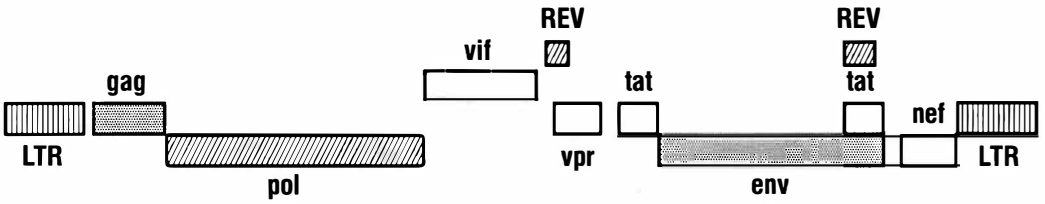
It would take a large number of antibodies to do the job. Using CD4 as the basis for an antiviral strategy, raising antibodies against the virus's recognition site, or "key," requires the molecular equivalent of outnumbering the enemy more than ten to one. It may be impractical to achieve this inside the human body. Finally, there is no proof that these antibodies could prevent transmission of HIV from one infected cell to another.

Recent research points to another site on the virus coat that is more variable, but which may also be vital to the virus's life cycle. Dani Bolognesi of Duke University told the Seven Springs conference about his work with this site, called the N site, which was discovered by injecting animals with gp160 or its subunit gp120. They produced antibodies different from the neutralizing antibodies that have been seen routinely. More of these alternative antibodies were later found, in small amounts, in some AIDS patients.

Antibodies that bind to the N site appear not to prevent the virus from initially binding to a cell, but they do block its later fusion to the cell wall and prevent the formation of syncytia. Researchers studying the N site believe it is involved in a



GENES OF THE AIDS VIRUS

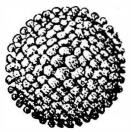


Genetic engineering has provided a detailed genetic knowledge of HIV1 and HIV2. The functions of most of the virus's genes are known, though exactly how they work is not.

HIV contains three genes common to all retroviruses: *env*, *pol*, and *gag*. *Env* encodes the instructions for making HIV's outer envelope. *Gag* encodes the information for the inner protein in its cylindrical core, and *pol* contains the code for the virus's own enzymes involved in such key steps as reverse transcription. (See page 42.)

In addition, HIV's genome contains a number of novel genes (many of them recently renamed). Together, they create a sophisticated system to control the virus's reproduction.

Some of the novel genes, such as NEF (formerly 3′ *orf*), appear to slow down the production of new virus. Other genes seem to enhance the virulence of the AIDS virus. VIF (formerly *sor*) increases its ability to infect new cells. TAT and REV (formerly called *trs/art*) regulate the production of new HIV's. Another gene, VPR, has unknown functions as yet.



structural change whereby the virus anchors firmly to the cell wall after docking onto CD4; perhaps it helps to "turn" the "key" after it is inserted in the lock.

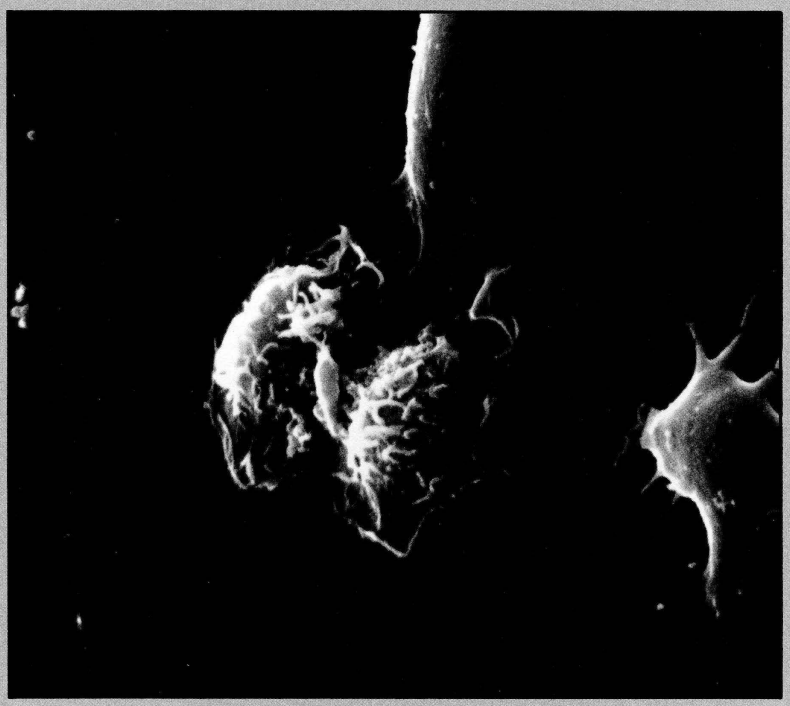
Antibodies directed against this site on the virus are no more likely than antibodies against the other gp120 site to form a useful vaccine in themselves. Dr. Bolognesi has been looking for versions of these antibodies in a laboratory worker accidentally infected with HIV. He found that, after about a year, the HIV isolates inside the patient had begun to mutate in ways that could escape the antibodies directed against the N site.

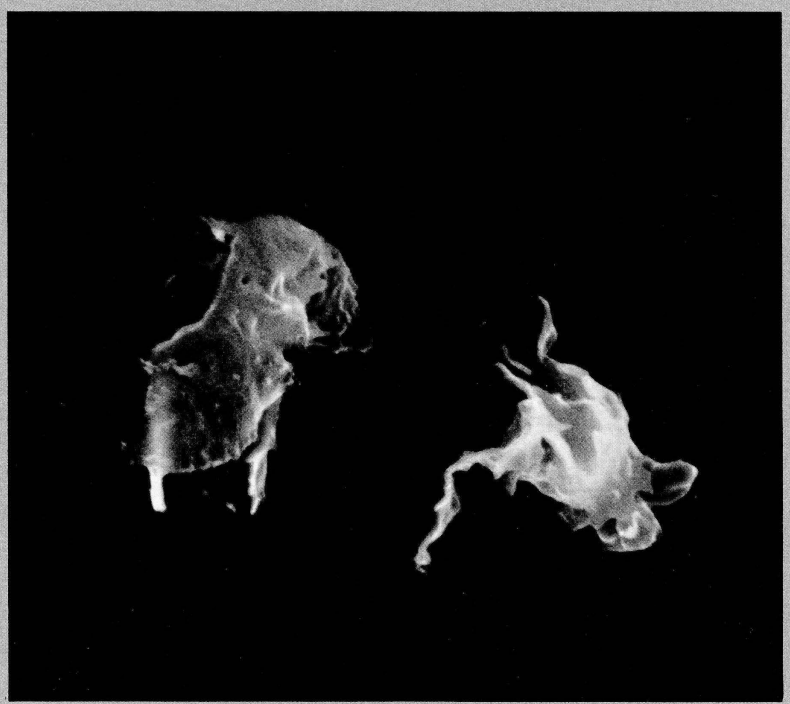
Antibodies are only a small part of the immune response, of course. On one arm of the immune system, white cells that arise from the bone marrow manufacture antibodies; on the other arm, white cells that come from bone marrow and pass through the thymus become T cells which kill viruses.

These two recognition systems don't even "see" a virus in the same way, Jay Berzofsky of the NCI pointed out. T cells never see the virus coat floating free in the bloodstream; they see pieces of it on the surface of another immune cell that has digested and processed the virus. Perhaps, he reasoned, it is possible to bypass antibodies entirely and design a vaccine that will stimulate T cells.

Which fragments of the virus tend to emerge on the surface of an antigen-presenting cell, where the T cells can find them? Dr. Berzofsky addressed the question by asking what structural properties are common to the molecule fragments known to stimulate a reaction from helper T cells. About three out of four such fragments, he has found, are helical, with one face of the helix having an affinity for water and the other face being hydrophobic, or water-repellant. "This structure," he remarked, "would tend to chemically stabilize a protein on the surface of the target that the helper cell must recognize."

Using computer modeling, Dr. Berzofsky has been analyzing the AIDS virus coat for chemical sites that match this two-sided helical pattern. To be useful for a vaccine, he reasoned, such sites should also come from a part of the viral coat known not to mutate frequently, and should not be heavily masked by complex sugars. He found two molecule fragments on HIV's glycoprotein coat that appeared to fit these criteria.



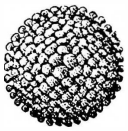


Scanning electron microscope views of two of the other cell types that can be infected with HIV: macrophages (top) and dendritic cells (bottom). Like helper lymphocytes, some macrophages and dendritic cells can express the T4 or CD4 molecule. Recall that T4 is the lock recognized by the gp120 protein key on the virus, permitting infection of the T4-bearing cells.

The macrophage is a phagocyte which actively scavenges, engulfs, or eats (phagocytoses) organisms. In some cases, the macrophage is permissive and is used as a site for intracellular microbial growth. The immune interferon that is produced by T4 helper lymphocytes makes macrophages less permissive or more microbicidal.

The dendritic cell is a sentinel cell which takes up small amounts of foreign materials or antigens. The cell then moves actively to find T cells that recognize antigenic fragments on the dendritic cell surface. This surface consists of many long sheet-like or spiny processes (veils, dendrites) which are used to probe the environment for T cells. Once an antigen-specific T cell is bound, the T cell starts to multiply and produce lymphokines like immune interferon.

Photographs supplied by Drs. Gilla Kaplan and Ralph Steinman of The Rockefeller University.

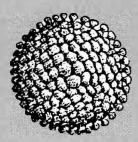


Dr. Berzofsky was able to test a vaccine made from one of these fragments on Dr. Zagury and some of his volunteers in Zaire. In laboratory tests, T cells from Dr. Zagury himself and eight of his healthy volunteers produced a strong immune response against HIV-infected cells after injections with Dr. Berzofsky's trial vaccine. But the upshot of the study was discouraging: Dr. Berzofsky's "new" site that stimulates T cells turned out to be identical to the viral site recognized by neutralizing antibodies. It only underscores, he remarked, how few antigenic sites, or epitopes, this virus gives human researchers to work with.

Other viruses than HIV have the habit of changing their coats slightly to evade vaccination, but many of them also have enough antigenic sites on their surface that multivalent vaccines—designed to seek out a number of sites—can generally provide protection. Given HIV's generally high mutation rate, its own paucity of recognition sites "augurs very ill if you play it out," mused Barry Bloom of the Albert Einstein College of Medicine. If you immunize a patient against one of these few known antigenic sites, he predicted, it's reasonable to expect that inside any patient the virus will eventually mutate at that site, escape the immune response, and you will have gained nothing.

Even if, by magic, researchers could produce a fail-safe vaccine today, remarked Richard Lerner of Scripps, it would be difficult to establish whether or not it could work. For a sexually transmitted disease, where and how would you inoculate? In its venereal form, HIV may spread rampantly before it is visible to immune cells or antibodies in the blood. At any rate, any sensible person, vaccinated or otherwise, will try mightily to avoid catching the AIDS virus. In that circumstance, how long will it take, testing how many volunteers, to decide that a vaccine is effective?

"I go to fifteen to twenty-five AIDS meetings a year," Dr. Koprowski said. "All of them turn around how to improve the vaccine. We should put great pressure on the SIV people [researchers studying a monkey virus closely related to HIV] to move with great speed to test vaccines."



DRUG TREATMENTS

As yet there is no cure for AIDS. The only drug approved by the Food and Drug Administration for AIDS treatment, 3'Azido2', 3'Dideoxythymidine (AZT or zidovudine), slows the progression of the disease and relieves some symptoms for as long as it is taken. But it has serious side effects.

A large number of potential anti-AIDS medicines are in clinical testing, both inside pharmaceutical companies and in multicenter trials sponsored by the National Institutes of Health. About 4,000 patients have been enrolled in the NIH trials since January 1988.

Drugs under testing include:

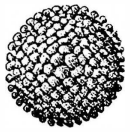
AZT: Approved by the FDA in March 1987, after a study showed only one death among 145 patients taking the drug for seven months, as against nineteen deaths among the 137 people who took a placebo. It is now sold by Burroughs-Wellcome under the brand name Retrovir. AZT ultimately depletes the white blood cell count, which makes it unsatisfactory as a sole treatment. It is currently being tested in combination with other anti-AIDS drugs, and also in individuals who are infected with HIV but have not yet developed symptoms of AIDS.

ddC (dideoxycytidine): Similar to AZT in action, but causing different side effects: neurological symptoms such as weakness and defects in the sense of touch. ddC improves immune cell counts and body weight in patients who have taken it. Being tested in schedules that alternate ddC with AZT, to limit side effects, and also in combination with other drugs such as interferon.

Dextran sulfate: A sugarlike compound which seems to inhibit the virus from binding to cells.

Interferon and other cytokines (products of healthy T cells): Immune-system boosters that may retard the progress of the AIDS-associated cancer, Kaposi's sarcoma.

Ampligen: Another immune booster, it restores the function of some immune cells and is thought to control virus replication.



DRUG APPROACHES

"I can't resist pointing out the irony," remarked Dr. Broder. "Four years ago, vaccines were considered the way to go, and drugs against AIDS were considered inherently impossible."

Today the reverse seems to be the case. About thirty drug candidates are being tested on AIDS patients. One of them, AZT or 3'Azido2', 3'Dideoxythymidine, has been shown to have some effects in retarding the progress of the disease in human beings, even though it is not a cure.

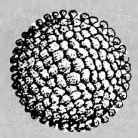
Most of the drugs currently considered for AIDS have been pulled down from the shelf and dusted off—the products of abortive drug development in pharmaceutical companies, which could find no use for them earlier. Better treatments may arise from research specifically directed at interrupting some part of HIV's life cycle. In the near term, Dr. Broder said, the best prospects seem to be drugs that can prevent HIV from translating its genetic blueprint into new virus particles, since this requires a number of events that are used by the virus and not by most normal cellular genes. Another prospect would be to prevent HIV from binding to a cell.

Already, a number of laboratories have succeeded in blocking the virus's entry into cells by flooding a laboratory dish with artificial versions of the CD4 molecule. Another approach would be to link a toxin to a version of the CD4 site, killing any virus that grabs it.

"It would be a difficult job for the virus to develop a strategy to evade this," Dr. Broder pointed out. Those mutants less likely to bind to the drug and be destroyed would also be less likely to lock onto immune cells.

A less specific drug, the polysaccharide dextran sulfate, may act by interfering physically with the binding of the virus. Long used safely as an anticoagulant, it is now under testing against HIV.

About 20 compounds exist that can prevent a virus from creating a DNA version of its own genetic blueprint, which it must do in order to infiltrate the DNA of the cell it has entered. Many of them work more or less like AZT: With a simple chemical modification, such as the addition of an oxygen atom, biochemists create an abnormal version of a DNA build-



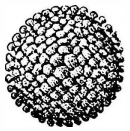
GM—CSF (granulocyte-macrophage colony stimulating factor): An immune system modifier that stimulates production of white blood cells. Tested in combination with AZT.

CD4 agonists: Molecules that mimic the structure of the molecule called CD4 on the surface of immune cells, with which the AIDS virus links to gain entry. Five different research groups have shown that these molecules can significantly block the virus in laboratory dishes. Preliminary studies show that monkeys tolerate the drug without obvious harm. Clinical trials are underway.

Ribavirin: An antiviral drug shown in some studies to prevent the lymph-gland swelling that progresses to AIDS and also the development of Pneumocystis carinii pneumonia. Unfortunately, recent studies show that ribavirin also inhibits the body reactions necessary to convert AZT to its active form, and therefore can hinder the effectiveness of the only government-approved AIDS treatment.

Many of these drugs, and a wide variety of other treatments, are available to AIDS patients on the black market. This makes it very difficult to assess AIDS treatments objectively. Researchers cannot be certain whether or not a patient may be concurrently taking another drug that either improves or hinders the effects of the drug (or placebo) in the official study.

While many researchers are trying to find a way to stop AIDS itself, it is up to medical doctors to try to fight the other illnesses that inevitably arise after AIDS infection. Because the immune system is already weak, they need to find new treatments in addition to the standard antibiotics. For example, Henry Murray of New York Hospital reported at Seven Springs that he and a number of other researchers are testing the use of immune modifiers such as gamma interferon to stimulate AIDS patients' ability to fight opportunistic infections. It is these infections, not the HIV infection itself, that are ultimately fatal.



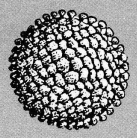
ing block that cannot form the chemical bond needed to extend the DNA molecule. The virus adds AZT to the DNA polymer, and the polymer stops growing.

Because the virus relies on a cell's own enzymes to construct its DNA transcript, it is totally vulnerable to such a strategy. In vitro, AZT can inhibit the replication of HIV at doses ten to twenty fold lower than the doses that impair the survival of human cells themselves.

But such drugs do interfere with cells' own DNA manufacture, and so cause some harm, because they do stop cells from dividing or repairing damage to their genetic material. Studies are now underway to test alternating AZT and ddC as a way to control the virus without disabling side effects such as nerve disorders or anemia.

These drugs are the mainstays for the near future, Dr. Broder said. There are a number of other ways in which HIV may be vulnerable to defeat by drugs:

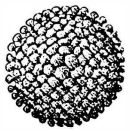
- ☐ Reverse transcriptase: This protein translates HIV's RNA genetic code into the form that can be inserted into the human cell's genes. Because it is unique to viruses, a drug that inactivated it might do no harm whatever to human cells.
- ☐ The TAT gene: In tissue culture, TAT seems to allow HIV to gear up its production of new viruses. The protein through which TAT has its effects has been identified, and ways may be found to block it. TAT is essential for virus production.
- ☐ The REV gene: AIDS viruses use REV to manufacture structural proteins, such as the viral coat. Drugs that block REV could also block the virus's life cycle.
- ☐ Glycosylation inhibitors: These chemicals can prevent the virus from chemically adhering complex sugar molecules to its exterior. Because the dense layer of sugar molecules that coat an AIDS virus appear to be one way it evades recognition by the immune system, finding a way to leave the virus coat "naked" could be a very effective strategy.



PRIORITIES FOR THE FUTURE

Researchers at the Seven Springs Conference raised a number of important priorities for ongoing AIDS research:

- **The need to understand the mechanism by which HIV infection leads to the complete depletion of CD4-positive immune cells;**
- **The need to look for protective immune responses, such as killer cells that can recognize and eliminate macrophages infected by HIV;**
- **The need to search for factors in body fluids that may help to protect against or control HIV infection;**
- **The need to extend the studies that have already been carried out on T cells to macrophages and dendritic cells;**
- **The need to test the effects of the substances produced by brain macrophages and microglia on other brain cells; and**
- **The need to optimize the use of the one animal model that may relate best to AIDS, the rhesus monkey infected with simian immunodeficiency virus (SIV), and to develop additional animal models such as those that involve the maintenance of a human immune system in mice.**



OF THE FUTURE

Eight years on, this is most of what we know about the science of AIDS. If the questions seem to outweigh the statements, consider the epidemic that terrified America in the other half of this century.

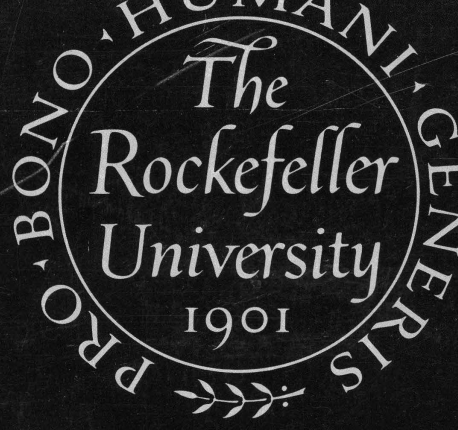
It was 1916. Public hygiene had begun to defeat diseases such as diphtheria and typhoid in American cities and, as they would a half-century later, many people began to regard medical science as all but invincible. Suddenly, for no evident reason, an outbreak of polio swept across New York City, striking people without regard to any kind of distinction.

Despite being a victim of the disease, Franklin Delano Roosevelt won the 1932 presidential election and became a vital symbol of courage during a long period when almost nothing was known about the polio virus. It took nearly 40 years to develop an effective vaccine.

By 1981, the year when a few male homosexuals in New York died of tenuous rare infections, most Americans took for granted that high-tech medicine had made intractable epidemics a thing of the past. Nature brought them up short.

It is now clear that AIDS will have a far greater impact on the world than anyone dreamed in 1981. We may have to accommodate it in the long term, as we accommodate cancer, mental illness, and poverty. But in contrast to the struggle against polio, we have started off knowing much more, and our tools are far better.

Because HIV uniquely strikes at our ability to combat infections, what scientists must learn as they defeat it will unquestionably have a major impact on medical science at large. Because of AIDS, someday we will understand better how to identify and confront other known viral diseases—and, probably, many more not yet linked to known viruses. AIDS research will certainly improve our understanding of the normal immune system, and of other diseases in which it goes wrong.



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