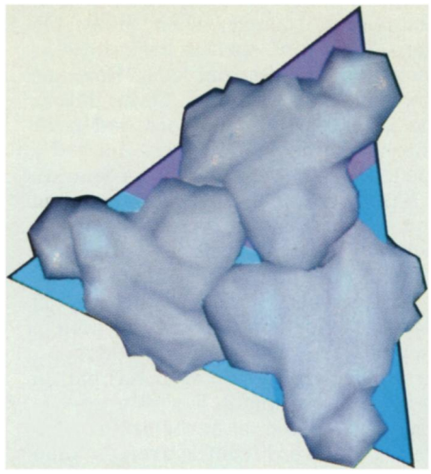
The Shell Game

A common cold virus offers clues to sabotaging AIDS



T.J. O'Donnell/Searle

In this computer model, the p17 protein of the AIDS virus, which makes up the HIV outer capsid, is folded to simulate the shape of one of three proteins that form the picornavirus capsid.

By RON COWEN

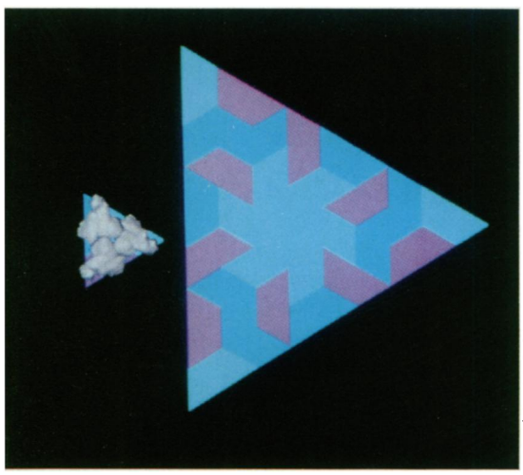
Imagine a fierce army encamped within a fortress sealed so tightly that the soldiers can never get out. Now picture a fortress so riddled with holes that nearly anyone can get in. Either way, the structure would cripple the army's attempt to invade surrounding territory.

That's the strategy some molecular biologists have adopted as they seek to weaken or paralyze the AIDS virus (HIV).

By tinkering with the ability of protein "bricks" to assemble into one of the two fortress-like shells — known as coats or capsids — surrounding the lethal genetic core of HIV, these scientists hope to force the virus to build a defective packaging that disarms its infectious spread. A drug that weakens the bonds between proteins or that stiffens individual protein "bricks," preventing them from stretching or compressing as needed to pack tightly together, might create a leaky fortress. The remaining viral material would then be vulnerable to destruction by the cell. Stiffening the protein bricks might also prevent an already assembled shell from cracking open and releasing the virus' genetic contents into the surrounding cell.

But researchers attempting to design drugs with just the right properties to degrade HIV's two protein coats have found themselves working in the dark, for no one knows the exact three-dimensional structure of these shells. Instead, investigators have turned to a family of smaller viruses for structural clues. Called picornaviruses, these measure about one-thirtieth the size of the AIDS virus and possess a single protein shell rather than two. Yet their protective coats may resemble those of the HIV in the way the proteins are organized.

Moreover, researchers have already crystallized and imaged several picornaviruses, including those that cause polio and the common cold. X-ray diffraction studies of the crystals have revealed that the protein tiles making up these viruses' capsids fit together in a symmetric pattern, in which a regular 12-faceted figure pokes through a 20-faceted one — an arrangement that vaguely resembles a soccer ball. And electron microscopy studies indeed suggest the two capsids of HIV may feature a similar protein arrangement.



T.J. O'Donnell/Searle

Reflecting the nine-fold larger surface area of the AIDS virus, 27 copies of the p17 capsid protein make up a single wedge-shaped "tile" of the HIV outer capsid (right) compared to three proteins for the picornavirus capsid.

In 1985, a research team led by Michael Rossmann at Purdue University in West Lafayette, Ind., became the first to chart in atomic detail the topography of an animal virus — human rhinovirus-14, a picornavirus that causes colds. They discovered that the rhinovirus capsid shows a recurring pattern of three proteins that fit together like bathroom tiles to form a relief map of miniature mountains and canyons.

The canyons slice through the capsid at 12 sites, each representing a vertex where five bound and folded proteins meet, Rossmann found. Moreover, the canyons' tiny passageways — too narrow for antibodies to penetrate — enables protein structures hidden at their bottoms to attach to receptors on the surface of human cells with minimal interference from the body's immune system, thus allowing the virus to enter a new cell. After entry, the rhinovirus need only open its capsid and release its genetic material for the infection to reach its sneezing, sniffling conclusion.

About the same time that Rossmann made his finding, scientists at the Sterling-Winthrop Research Institute in Rensselaer, N.Y., announced they had constructed two compounds that slowed infection of some picornaviruses, including rhinovirus-14. In collaboration with these investigators, Rossmann's team crystallized a solution of the drugs mixed with the viruses, catching the compounds in the act of binding to the picornavirus capsids. The researchers discovered that the small drug molecules slip into the narrow canyons formed by bound proteins and then settle into empty cavities beneath each canyon floor. Filling these cavities stiffens the capsid, preventing the virus from shedding it, Rossmann says. Thus, the virus' genetic blueprint, ordinarily used to commandeer cells, remains locked within the picornavirus. Rossmann reviews these findings in the fall 1989 VIRAL IMMUNOLOGY and the 1989 *Annual Review of Biochemistry* (Annual Reviews Inc., Palo Alto, Calif.).

His group, again working with Sterling-Winthrop researchers, is currently searching for drugs that bind more tightly to the canyon bottoms of the rhinovirus-14 capsid. In addition, they are attempting to use the rhinovirus-14 model to design drugs that might prevent one of HIV's two protein coats from forming properly or disassembling. A separate group, headed by James P.

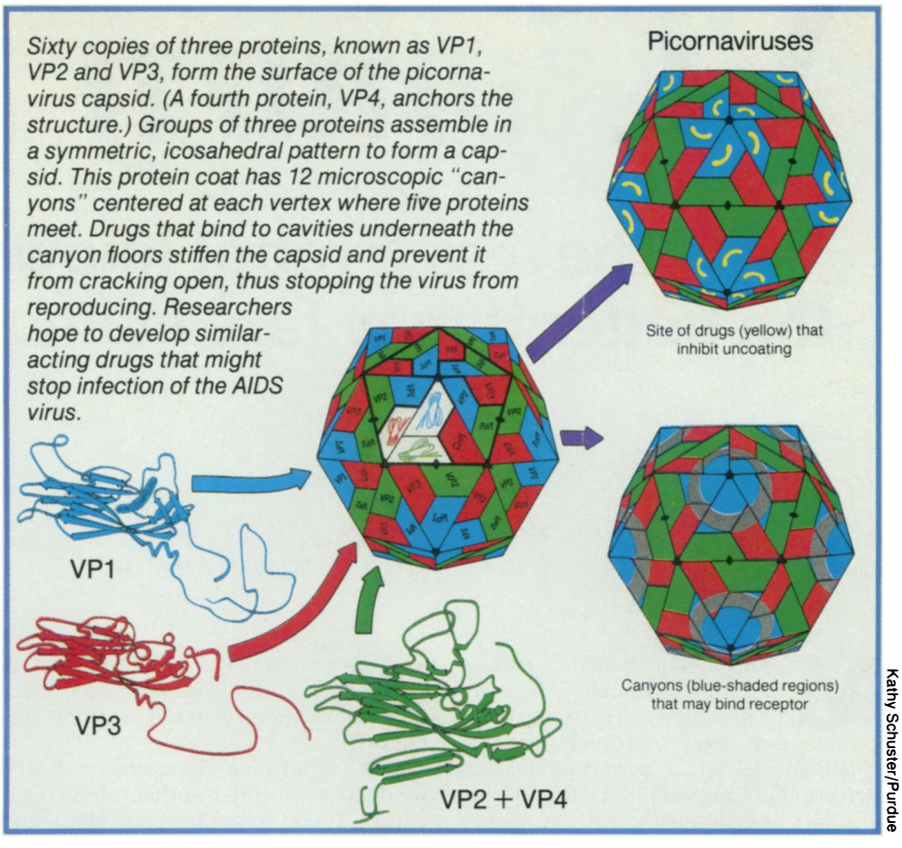
Snyder of Searle Research and Development in Skokie, Ill., is pursuing a similar approach to drug design.

The hills and valleys of HIV's two coats remain unmapped. But proper formation and disassembly of the coats appear critical for infection, and some researchers think both coats may possess a topographic symmetry similar to the rhinovirus-14 capsid. Just as 60 copies of three different proteins form the surface of the rhinovirus capsid, some 2,000 copies of a protein known as p24 come together to form the bullet-shaped capsid surrounding HIV's genetic material. And numerous copies of another protein, called p17, form a second, outer shell just beneath HIV's lipid-membrane envelope.

In the July 1988 PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES (Vol.85, No.13), Rossmann suggests that when copies of p24 come together to form HIV's inner shell, each may adopt a multi-looped, twisted structure similar to that of the three capsid proteins coating the human rhinovirus-14. Such a configuration would cause the p24 capsid to form a symmetrical pattern of canyons and hidden pockets, each a possible target for drugs that might interfere with this capsid's formation or disassembly. And last month, at a meeting on HIV structure held at the National Institute of General Medical Sciences in Bethesda, Md., Snyder outlined a new computer simulation that uses the rhinovirus-14 model to design drugs that might prevent assembly of HIV's p17 outer coat.

The computer model assumes that the p17 coat has the same symmetry as the rhinovirus-14 capsid but covers a larger surface area – about nine times that of human rhinoviruses-14. So Snyder's team – knowing that the rhinovirus-14 capsid has 60 "tiles," each containing three different proteins – models the HIV protein coat using 60 tiles, but with each tile containing 27 copies of the p17 protein. Using a computer to select a particular configuration of the 27 copies of p17 within a tile, the team then assembles all tiles according to the rules of icosahedral symmetry. Each assembly configuration (Snyder isn't sure which one correctly mimics the HIV outer shell) corresponds to a particular pattern for binding neighboring p17 proteins. Thus, each computer-generated shell may suggest a different drug molecule to thwart that assembly. But the end result remains the same: A defective shell may make HIV more susceptible to immune defenses.

Snyder admits his approach relies on trial and error, and he expects the first several drugs suggested by the computer model to react only minimally with the picornavirus family or HIV. It may take years to develop a new and safe anti-AIDS therapy through this process, he adds.



Kathy Schuster/Purdue

However, he says his team has begun synthesizing several computer-designed compounds and will start *in vitro* testing soon to determine whether any show potential against HIV.

Andrew Prongay, a collaborator in Rossmann's laboratory, cautions that the p24 protein might not contain folds and loops similar to the rhinovirus proteins. In that case, he says, the researchers would have to abandon their comparative computer modeling and come up with new methods of interfering with the normal function of the protein coats.

Nonetheless, Prongay, Rossman and their co-workers have begun intensive study of the p24 capsid. At last month's HIV structure conference, he announced that the group had succeeded in crystallizing p24. Prongay is now conducting X-ray studies of the crystalline structure, which he says should put to an end questions about the HIV capsid's structural similarity to rhinovirus capsids.

Krysztof Appelt of Agouron Pharmaceuticals Inc. in La Jolla, Calif., expresses skepticism that current studies of the protein coats will readily yield drugs effective against the AIDS virus. He maintains that other components of the AIDS virus, such as the enzymes reverse transcriptase and HIV protease, are better targets for drug therapy because scientists have already identified their structure.

"The similarities are very, very general between the HIV capsid and that of the rhinovirus," Appelt argues. "I think it would be great to design a drug against the HIV capsid, but with our current knowledge that seems unlikely."

Another research group says it may already have a drug that reduces infection by manipulating HIV's p24 capsid. The investigators say their evidence suggests that hypericin, a compound derived from an herb called St. Johnswort, inactivates HIV and a mouse retrovirus *in vitro* by interfering with the release of genetic material from their cores. Led by David Lavie of the Weizmann Institute of Science in Rehovot, Israel, and Daniel Meruelo of the New York University Medical School in New York City, the team reports its findings in the August 1989 PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES (Vol.86, No.15).

Meruelo told SCIENCE NEWS he believes that hypericin inhibits HIV infection by stiffening the HIV capsid, possibly in a manner similar to the newly developed drugs that inhibit rhinoviruses. He adds that FDA-approved clinical trials of the compound – which was previously tested in humans as an antidepressant – will begin soon.

Can a drug that weakens the capsid's assembly or prevents its disassembly serve as an effective treatment in fighting AIDS? "It would take someone with the wisdom of Solomon to decide that at this time," Meruelo says. But, he adds, "when you have a virus this deadly, any approach is worthwhile." □