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NEWS OF THE WEEK

VIROLOGY:

Active Poliovirus Baked From Scratch

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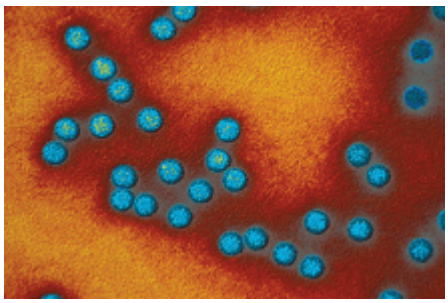
With mail-order DNA and more than 2 years of painstaking work, researchers for the first time have assembled a virus from its chemical code. The lab-built poliovirus killed mice and was almost indistinguishable from the original. Biologists disagree on how difficult it would be to construct far bulkier viruses such as smallpox to create bioweapons.

Scientists hail the research, described online this week by *Science* (www.sciencemag.org/cgi/content/abstract/1072266), as a technical achievement. But in an age when anthrax travels through the mail, few could avoid the paper's obvious implications, both for polio--a disease that once triggered panicky epidemics and is now nearing global eradication--and other viral diseases. "It is a little sobering to see that folks in the chemistry lab can basically create a virus from scratch," says James LeDuc, director of the Division of Viral and Rickettsial Diseases at the Centers for Disease Control and Prevention in Atlanta. Vincent Racaniello, a virologist at Columbia University in New York City, was more blunt. "Poliovirus," he says, "will never be gone."

A genomic runt at just 7741 bases, poliovirus is composed of a single strand of RNA and ranks among the most thoroughly dissected viruses of all time. Once it infects a cell, the RNA translates itself into a large protein, which is then cleaved to produce a cluster of smaller ones. Those proteins attack critical cells such as neurons in the central nervous system.

The researchers--Jeronimo Cello, Aniko Paul, and Eckard Wimmer of the State University of New York, Stony Brook--built an almost perfect replica of the virus, reading the recipe available in a public database of the letters that make up the virus's chemical code. Because RNA is chemically unstable, the scientists converted the RNA sequence to DNA, replacing every uracil base with a thymine. Then they ordered short stretches of carefully arranged bases from one of the many companies that churns out such piecemeal DNA. Cello took about a year to layer these

fragments together to form the first third of the virus. Once he established that these stretches stayed oriented correctly, he hired a DNA synthesis company to assemble the remaining portion, which it did in 2 months. To distinguish the synthetic virus from natural strains, the group inserted 19 markers, minor mutations that weren't expected to alter polio's behavior.



According to plan. Poliovirus reconstructed from its genetic sequence is indistinguishable from the original, shown here.

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DNA in hand, the researchers immersed it in enzymes to convert it back to the RNA at polio's core. The artificial poliovirus acted much like its natural counterpart: It multiplied, and antibodies could block it from entering cells. Mice injected with the synthesized virus became paralyzed after about a week, as did animals infected with normal poliovirus. But the artificial version was less lethal: Between 1000 and 10,000 times more virus was needed to kill an animal. The team suspects that one or more of the 19 markers are hobbling the virus.

The research might throw a wrench into polio eradication plans. "It erodes the underpinning of the whole eradication concept," says Peter Jahrling, a smallpox researcher at the U.S. Army Medical Research Institute of Infectious Diseases in Fort Detrick, Maryland. Last month the World Health Organization (WHO) announced that it had erased the disease from the European continent, and, according to Bruce Aylward, WHO's coordinator of the Global Polio Eradication Initiative in Geneva, "the goal is to stop immunization" once the disease is fully eradicated. But given the possibility of recreating the virus, researchers who favor continued immunization, such as Donald A. Henderson, an adviser to the U.S. government on bioterrorism policies, hope that WHO will reconsider its stance.

Then there's the question of whether one could reconstruct other pathogens whose sequences are publicly available. Smallpox, among the most feared bioterror agents, is far more massive than polio at 185,000 bases and far more complex. LeDuc, for one, doesn't believe that rebuilding it is imminently doable. But given the new results, others aren't so sure. "In principle, yes, [it's] possible to synthesize smallpox," says Vadim Agol, a virologist at the Russian Academy of Medical Sciences in Moscow.

Despite such nightmarish scenarios, scientists have no plan to stop posting new genetic sequences online. Wimmer says that no concerns were raised to him about publishing the paper. As Cello says, "By releasing this you alert the authorities ... [about] what bioterrorists could do."

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REPORTS:

Chemical Synthesis of Poliovirus cDNA: Generation of Infectious Virus in the Absence of Natural Template

Jeronimo Cello, Aniko V. Paul, and Eckard Wimmer

Science 9 August 2002: 1016-1018 **Abstract »** **Full Text »**

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