VIROLOGY

First Genes Isolated From The Deadly 1918 Flu Virus

The great flu pandemic of 1918 was nothing to sneeze at. Although rarely more than a footnote in modern history books, it killed 20 million to 40 million people worldwide, 675,000 in the United States alone. That's 10 times more than the country lost in World War I, which raged at the same time, and almost twice the total number of U.S. deaths caused by the 15-year-long AIDS epidemic. Now, one of the pandemic's victims, a 21-year-old Army private who had been stationed in Fort Jackson, South Carolina, has given modern researchers their first direct look at the culprit.

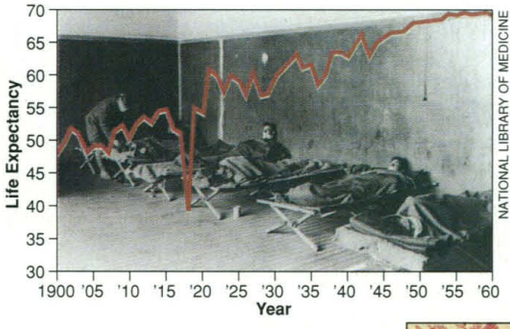
On page 1793, pathologist Jeffery Taubenberger, molecular biologist Ann Reid, and their colleagues at the Armed Forces Institute of Pathology in Washington, D.C., report that they have isolated pieces of five flu genes from the private's lungs. While the genes collected so far portray the killer as a run-of-the-mill swine flu, its deadly secrets may emerge as Taubenberger and Reid collect more of the viral genome and other researchers apply the same methods to other preserved tissue from the 1918 epidemic.

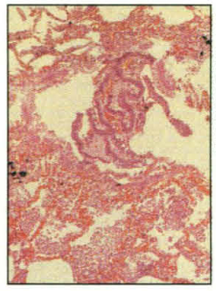
This report "probably won't make as big a splash as cloning that sheep, [but] if we can get a handle on what caused those deaths, that is important," says historian Alfred Crosby, author of the book, *America's Forgotten Pandemic: The Influenza of 1918*. Knowing what a killer flu virus looks like, for instance, could enable health officials to spot particularly dangerous flu strains as they emerge and "help us prepare for what we need to be prepared for," says Nancy Cox, a virologist from the Centers for Disease Control and Prevention (CDC) in Atlanta.

For the current work, Taubenberger and Reid began with several dozen samples of lung tissues taken from soldiers who died of the 1918 flu and preserved in their institute's archives. These, they realized, could be analyzed by modern polymerase chain reaction (PCR) methods, which can amplify extremely small quantities of DNA and RNA, thus providing sufficient amounts of material for sequence analysis. From the beginning, though, the researchers knew success was a long shot.

Typically, the flu virus infects the respiratory system, replicates, and then is shed into the air via the lungs, all in just a few days. Thus, unless one of the soldiers had died very soon after infection, all traces of the virus would probably have vanished by the time an autopsy was performed. What's more, the flu virus stores its genetic information on single strands of RNA, which is much more susceptible to degradation by cellular enzymes than DNA. "So you have to look for little pieces," Taubenberger explains. These problems had already proved insurmountable once before. Several years ago, virologist Robert Webster of St. Jude Children's Research Hospital in Memphis, Tennessee, had failed in an attempt to pull genetic material from archived samples.

To increase their chance of finding viral RNA, the researchers worked for a year refining their PCR procedure. In addition, they carefully examined each of the lung-tissue specimens under a microscope to find ones with indications of viral infections, such as lung air spaces filled with fluid and blood and damaged cells in the bronchial epithelial linings. Of the 28 tissue samples examined, Taubenberger says, the private's "was the only case with all the features of viral pneumonia."





Global killer. The 1918 flu filled hospitals, decreased life expectancy, and *(right)* damaged lungs.

His was also the only specimen in which they could identify flu-virus RNA sequences, including segments of five genes: those encoding hemagglutinin and neuraminidase—proteins that help the virus get into a cell—and three structural proteins. From the hemagglutinin and neuraminidase gene fragments, the researchers were able to confirm earlier conclusions about the virus's source.

Influenza viruses are thought to pass into pigs from birds, usually ducks, that are kept in close proximity to swine in some parts of the world, particularly China. Subsequent genetic changes enable the virus to move from pigs into people. However, there had been some speculation that the 1918 flu virus might have been so deadly because it was an avian virus, and these data indicate that isn't the case.

Because the flu-virus genes from the private closely resemble those of viruses isolated from pigs, the researchers concluded that his virus had been in the pig population for a while. That is also in line with previous antibody studies of blood taken from people who had lived through the 1918 pandemic, indicating that this killer plague was a classical swine flu. "What this says is we had better watch what's happening in the pig populations of the world," Webster says, because it seems that ever-changing flu viruses in those animals are the source of new flu viruses in humans. Currently, both the CDC and the World Health Organization devote most of their flu-surveillance efforts to monitoring for new flu strains in people, not animals.

The work does not support another supposition about the virus, however. Virologists have suggested that the extreme virulence of the 1918 flu virus may have resulted from the same genetic change that enabled an avian flu virus to kill whole flocks of chickens overnight in Mexico 2 years ago (*Science*, 17 March 1995, p. 1594). The deadly mutation altered a segment of the hemagglutinin protein, probably increasing the virus's ability to infect cells. But Taubenberger's group found no such change in the specimen they examined.

Hoping to find more clues to these and other mysteries, such as why young adults, usually the most resistant to flu infections, were the hardest hit in 1918, Taubenberger and Reid are continuing to isolate more of the viral RNA from the private's tissue samples. At the same time, they plan to use PCR to make lots of copies of all the viral RNA bits they recover. From these, they will create a cDNA library so as to have an unlimited supply of that genetic material.

Meanwhile, Taubenberger hopes that his success will inspire others to comb their archives for other possible samples. Already, before learning of the current work, virologist John Oxford of London Hospital Medical College had begun collecting samples from his own institution and from Australia and Prague for a similar analysis. And a Canadian-led team hopes to dig up seven miners who presumably died of the 1918 flu and have been preserved in their graves in the frozen ground near Spitzbergen, Norway.

Webster worries about the slim chance that those bodies will contain live virus. But it may be worth the risk, he says: "We want to know what killed these people. The potential is there for this kind of virus to return."

–Elizabeth Pennisi