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Getting to the Core of Reovirus

HHMI researchers have solved the structure of an important component of the reovirus, a double-stranded RNA virus that bears similarity to pathogens such as rotavirus, a potentially deadly cause of diarrhea in infants.

Using X-ray crystallography, a team led by Stephen C. Harrison, an HHMI investigator at Children's Hospital in Boston and Harvard University, determined the three-dimensional architecture of the core structure of reovirus.

The team, which also included lead author Karin M. Reinisch, a postdoctoral fellow in Harrison's laboratory, and Max L. Nibert at the Institute for Molecular Virology at the University of Wisconsin, Madison, reported their findings in the April 27, 2000, issue of the journal *Nature*.

"This work will provide much insight on two levels," says Aaron Shatkin, director of the Center for Advanced Biotechnology and Medicine at Rutgers University and a professor at the University of Medicine and Dentistry of New Jersey-Robert Wood Johnson Medical School. He notes that understanding the reoviral structure will inform studies of cellular genetic messaging in higher life forms, including humans. Many double-stranded RNA viruses are found in nature, so the structure could serve as a model for these related viruses, says Shatkin.

The reoviral core is an internal component of the virus that remains intact after the virus penetrates the host cell, as in other plant and animal double-stranded RNA viruses. The core synthesizes, modifies, and exports viral messenger RNA (mRNA), which eventually makes its way to the host-cell ribosome where viral proteins are constructed from the mRNA, thus completing the viral takeover of the host cell.

Once they solved the core structure, Harrison's group began to examine its role in viral replication. The researchers found that the core encases the viral genome, which is made up of ten distinct segments of double-stranded RNA. They also showed that the core organizes RNA so that it can easily be transcribed into many copies.

Harrison's team found how the core ensures that mRNA is modified with a methyl guanosine cap in a sequence of reactions discovered many years ago by Shatkin.

"Reovirus makes a cage to trap the end of the messenger RNA as it emerges from the core," explains Harrison. "It makes a hollow that holds the RNA long enough with five possible sites that it can hit, any one of which can do the job in order to make sure the RNA gets capped."

These "hollows" resemble turrets projecting from the core and are the capping machinery. The synthesized mRNAs are capped the instant they emerge from the active site of the polymerase. The guanosine cap is essential for RNA stability and for the ribosome to recognize the viral RNA.

In Harrison's opinion, the structural study of important viral pathogens has revealed a number of fundamental principles in macromolecular and protein assembly.

"I've always been interested in problems of viral structure from both ends," says Harrison. In the case of the reovirus core, he asks, how do you organize a structure like that to be able smoothly and without entanglement to make multiple copies of ten different messenger RNAs, cap them, and get them out?

The reovirus core has also interested Harrison's lab because it may bear similarities to other double-stranded RNA viruses that are important pathogens.

In upcoming studies, Harrison's group will be looking at the relationship between the structure and function of the outer shell proteins that get reoviruses and rotaviruses into cells.

Broadly speaking, viruses use two styles of entry. Enveloped viruses, like influenza and HIV, have bilayer membranes, and get in by fusing their envelope with the cell's membrane. Solving how membrane-less, non-enveloped viruses like reovirus and poliovirus, gain entry has been harder to do, Harrison says.