New Class of Animal Virus Found in Virulent Form of Human Hepatitis

A virus that for 10 years has been associated with certain severe forms of hepatitis turns out to be a novel form of animal virus, which is similar in some respects to certain plant viruses known as viroids; in turn, the new virus and the viroids are similar to group I introns

THE more that biologists learn about the natural world the more they seem to uncover the connectedness of things. Just such a revelation has occurred in what is being hailed as a major advance in understanding the molecular biology of a particularly virulent form of hepatitis.

Three research groups have independently discovered that hepatitis delta virus, which seems to be responsible for an increased morbidity and mortality in certain instances of the disease, shares many structural features with those curious plant pathogens, the viroids and virusoids. This finding follows closely upon the recent demonstration that viroids and virusoids are themselves very similar in many ways to certain types of introns. Such shared similarities invite speculation about a possible close evolutionary relationship between these otherwise disparate structures.

The discovery that hepatitis delta virus is similar to plant viroids and virusoids, in having a small, circular RNA genome, opens a new chapter in animal virology. "There is no counterpart among animal viruses," comments John Gerin of Georgetown University, Washington, D.C. "It is a new class of infectious agent in animals." Gerin suspects that now that hepatitis delta virus has been identified as a viroid-like agent, other similar viruses will soon be discovered in animals. "They might be associated with some of the unusual disease patterns that so far are unexplained."

Hepatitis delta virus was first detected, initially by the presence of a specific antigen, almost 10 years ago in certain Italian carriers of hepatitis B virus by M. Rizzetto and his colleagues. It was soon clear that the delta antigen occurred only in those already infected with hepatitis B virus, implying that the virus itself might be in some way "defective." Patients carrying the delta antigen had a higher incidence of chronic active hepatitis and cirrhosis and were more likely to die than people infected with hepatitis B virus alone. It has since been determined that although the delta virus is found worldwide, the incidence varies greatly between different geographic populations.

After discovering the delta virus, Rizzetto spent some time in Gerin's laboratory at Georgetown University, where the characterization of the virus began. For instance, it is now clear that delta requires the presence of hepatitis B virus for transmission but not replication. Although the delta virus appears to produce its own specific antigen, it coopts the hepatitis B virus surface antigen in assembling its coat. By 1983 it was clear that delta was a small, unusual virus, and Gerin and his colleagues speculated that it might resemble in some ways plant viroids and virusoids. That speculation now appears to have been borne out.

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Gerin and his colleagues at Georgetown University, in conjunction with Michael Houghton and his colleagues at Chiron Corporation, California, report the overall structure and sequence of the delta virus in the 9 October issue of *Nature*. In the same issue A. Kos and his colleagues of the Primate Center, Rijswijk, the Netherlands, describe electron micrographic evidence that the virus is indeed circular. And, in a paper soon to be published in the *Proceedings of the National Academy of Sciences*, John Taylor and his colleagues at Fox Chase Cancer Center, give details of the structure of various forms of the virus in infected cells, which give insights into the mode of replication. The message these several authors bear is persuasive.

What emerges is a set a similarities and differences between the delta virus and the class of agents that include viroids and virusoids. For instance, hepatitis delta is like plant viroids in having an RNA genome formed from a single-stranded, closed circle. It appears to be similar too in its mode of replication, which is by the "rolling circle" process. But, at 1678 bases in its genome, delta is about four times bigger than typical viroids. And viroids do not appear to have protein coats, as delta does. In both these respects, the delta virus is more like the satellite viruses of plants. But satellite viruses require helper viruses for their replication, which is contrary to the situation in viroids and the delta virus.

There is, therefore, a confection of similarities and differences between the delta virus and viroids and their plant-associated cousins in these major structural and functional features. Buried within the sequence data are yet more similarities, some of which are very intriguing indeed.

Houghton and his colleagues identified two short sequences, one of five nucleotides and the other of six, that exist in virtually all viroids and in hepatitis delta virus. In addition, there are two longer regions (nucleotide regions 75 to 94 and 552 to 571) that have a 65% homology with a very characteristic part of the viroid molecule, known as the central conserved region. In their paper Gerin and Houghton and their colleagues comment: "The functional significance of these homologies and of the general similarities with plant agents remains to be established."

It is indeed true that the functional significance of these similarities is likely to remain a mystery for some time, not least because the functional significance of these regions in viroids themselves is largely unknown. But an evolutionary significance may more readily be inferred from them, as Kos and his colleagues are willing to do. They write: "These homologies with plant viruses and the viroid character of the genome of [hepatitis delta virus] encourages speculation about its possible plant origin."

It so happens that during the past year several researchers have noted structural similarities between viroids and the group of introns known as group I.

Group I introns include those in mitochondrial messenger and ribosomal RNA genes, chloroplast transfer RNA genes, and nuclear ribosomal RNA genes. These introns are characterized by the possession of a series of conserved regions, which impose certain secondary and tertiary structure constraints. What is most intriguing about these introns is that at least some of them are able to self-splice, that is to excise themselves and ligate the parental strands in the absence of enzymes, releasing a small circular RNA molecule.

In separate studies Gail Dinter-Gottlieb, of Drexel University, and A. Hadidi, of the Plant Protection Institute, Beltsville, Maryland, noted echoes of the group I intron conserved regions in most viroid structures they examined. In addition, parts of the central conserved region were also to be found in the introns. Both authors suggested some kind of evolutionary relationship between the two groups of RNA molecules, and Dinter-Gottlieb wondered whether viroids may be "escaped introns."

"When we saw Dinter-Gottlieb's paper we immediately looked at the hepatitis delta virus to see if there were any group I intron homologies to be found," Houghton told *Science*. "What we saw wasn't particularly striking, but there are some homologies that are probably statistically significant." The link between the delta virus, the viroids, and the group I introns is therefore complete.

Several interpretations of what this might mean are possible. For instance, perhaps both plant viroids and the hepatitis delta virus (including any other similar agents) are escaped introns. Instead, all three classes of RNA molecule may have derived from an as yet unidentified ancestor molecule. Or perhaps some other relational combination applies. As Houghton says, "It is hazardous to speculate too firmly about it." The guessing game will be settled only by more detailed sequence comparisons. ■

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ADDITIONAL READING

K.-S. Wang *et al.*, "Structure, sequence and expression of the hepatitis delta viral genome," *Nature (London)* **323**, 508 (1986).
A. Kos *et al.*, "The hepatitis delta virus possesses a circular RNA," *ibid.* **323**, 558 (1986).
G. Dinter-Gottlieb, "Viroids and virusoids are related to group I introns," *Proc. Natl. Acad. Sci. U.S.A.* **83**, 6250 (1986).
A. Hadidi, "Relationship of viroids and certain other plant pathogenic nucleic acids to group I and II introns," *Plant Mol. Biol.* 7, 129 (1986).

An Optical Measurement of Berry's Phase

Overlooked for 60 years, a phase factor that can creep into quantum mechanical wave functions has implications in areas ranging from molecular physics to unified field theories

THREE years ago while lecturing in the United States, Michael Berry of the University of Bristol found himself unable to field a question on the behavior of certain types of quantum mechanical wave functions when time-reversal symmetry was violated. Berry went home, spent 2 weeks thinking through the question, and discovered what he called the quantum adiabatic phase, although everyone else now calls it Berry's phase. The quantum adiabatic theorem, which Berry's phase modifies, is almost as old as quantum mechanics itself. "It should have been discovered 50 years ago," says Barry Simon of the California Institute of Technology.

The path dependence is what makes Berry's phase a geometric effect.

The finding triggered a flurry of theoretical papers showing how Berry's phase turns up in contexts ranging from the mature field of molecular physics to the arcane world of the quantum field theories known as gauge theories. A key paper by Simon recast Berry's result, which is geometrical in origin, in the compact language of differential geometry. Simon's treatment not only makes computation less ponderous, but it also underscores the close connection between geometry and modern theoretical physics, where the topological properties of curved surfaces have become of decisive importance.

While the theoretical work has left no doubt as to the existence of Berry's geometrical phase, direct experimental confirmation was lacking until recently. In August, Akira Tomita, who is now at the Raychem Corporation, Menlo Park, California, and Raymond Chiao of the University of California at Berkeley published the first experimental measurement of Berry's phase. In short, as predicted by Chiao and Yong-Shi Wu of the University of Utah, the angle of rotation of linearly polarized laser light on passing through a helical, single-mode optical fiber is a measure of the geometrical phase. It is conceivable, says Chiao, that this kind of optical activity in a fiber will have applications in optical devices, such as wavelength-independent phase shifters and optical rotators. Although highly speculative at the moment, Berry's phase could also be the basis for optical fiber gyroscopes and memory devices.

In June, Guy Delacrétaz and Ludger Wöste of the Swiss Federal Institute for Technology in Lausanne, Edward Grant and Josef Zwanziger, who are now at Purdue University, and Robert Whetten of the University of California at Los Angeles reported a spectroscopic study of triatomic sodium clusters in a supercooled molecular beam. Although the experiment did not measure Berry's phase, it did show that the so-called pseudorotational quantum numbers of a cluster are half integers, which is a direct consequence of Berry's phase. Ordinarily, pseudorotational quantum numbers are integers only.

It turns out that other hints of Berry's phase have appeared in the molecular physics literature for almost 30 years, but no one interpreted them to mean that standard treatments of the quantum adiabatic theorem were incomplete. In both classical and quantum mechanics, adiabatic refers to a change that takes place very slowly in the environment of a physical system. For example, molecular physicists commonly assume that the vibrational and rotational motions of the nuclei are so slow compared to the orbital motion of the electrons that the nuclear movement constitutes an adiabatic change in the environment of the electrons.

According to the quantum adiabatic theorem, which was discussed as early as 1928, if the change is slow enough, the wave function of the physical system will instantaneously adjust to the change. In particular, if the environment returns to its original condition after an adiabatic excursion, the wave function will also return to its original value multiplied by what is called a dynamic phase factor whose value depends on the time duration of the excursion. Except in special