A Virus by Any Other Name . . .

Would still cause AIDS; the assorted AIDS virus isolates are variants of the same virus, but agreement on a name is hard to come by

If any doubt remained, it has now been resolved. Comparison of the genetic material of the viruses that have been isolated and linked to AIDS (acquired immune deficiency syndrome) shows that they are, as expected, variants of the same virus. They are not identical, however, and the differences could affect efforts to develop a vaccine to protect against the disease and also the reliability of the newly developed AIDS tests.

Another issue that has been emphasized by the availability of the sequence data concerns the naming of the AIDS virus. This may be of less practical interest than vaccine development but is nonetheless proving to be a bone of contention.

Currently there are no less than three names for the virus. Luc Montagnier and his colleagues at the Pasteur Institute in Paris call it lymphadenopathy-associated virus (LAV) because they originally isolated it from an individual with lymphadenopathy, a condition characterized by swollen lymph nodes and fever that may be a mild or early form of AIDS. Robert Gallo and his colleagues at the National Cancer Institute (NCI) designated the virus they isolated as human T-cell lymphotropic virus-III (HTLV-III) because they found it to resemble HTLV-I and -II, which cause leukemias or lymphomas in humans. And finally, Jay Levy and his colleagues at the University of California in San Francisco simply called their isolate AIDS-associated retrovirus (ARV). (Retroviruses have RNA as their genetic material and their life cycle includes a step in which the RNA is copied into DNA.)

Although the Pasteur group now prefers the LAV designation, in their original report of the isolation of the virus, which appeared in the 20 May 1983 issue of *Science*, they referred to it as a new member of the HTLV family. They had detected in the patient from whom the virus was isolated antibodies that appeared to react with HTLV-I proteins, a result indicating that HTLV-I and the new viral isolate might be related.

This result turned out to be an artifact, Montagnier says. On further study, he and his colleagues could find no similarity between proteins from HTLV-I and the new virus and concluded that the two viruses were not related after all. The Pasteur group began using the LAV designation in September 1983 when they presented their data at a Cold Spring Harbor meeting on the "Human T-Cell Leukemia/Lymphoma Virus." By then they had obtained three additional isolates, this time from patients with full-blown AIDS, and gave them the designation "immune deficiency–associated virus" (IDAV). According to Montagnier, the designations LAV and IDAV were chosen because the link to AIDS had not yet been firmly established.

For a time at least, Gallo and his colleagues appeared to be following a false trail. In the same *Science* issue in which the Pasteur group described their first LAV isolate, the NCI workers reported a possible connection between AIDS and HTLV-I itself. Gallo had suspected that there might be such a link because HTLV-I and the then still mysterious AIDS agent seemed to be transmitted in a similar manner and they both preferentially infected the same kind of T cell.

> This concentration of differences in the *env* gene may complicate the development of an AIDS vaccine. The envelope protein is the likely target of the protective antibodies.

By the time these reports were published, however, the NCI workers were being tantalized by indications that T cells from many AIDS patients carried another virus, one which was proving hard to grow in cultured cells, a problem that was also hindering the Pasteur workers in their studies of LAV/IDAV.

The Gallo group solved the problem first, publishing in the 4 May 1984 issue of *Science* a series of four papers dealing with the propagation and characterization of the virus, which they designated HTLV-III, and presenting strong evidence for a link between it and AIDS. Levy subsequently published his report of the isolation of ARV in the 24 August 1984 *Science*. By the end of 1984 the work from both sides of the Atlantic had coalesced to the point where there seemed to be little doubt that the investigators were all studying the same virus, a conclusion now confirmed by the sequence comparisons.

They show that LAV and HTLV-III differ in only 1.5 percent of their nucleotides (*1, 2*). The proteins they encode would be identical in about 98 percent of their amino acids. The ARV genome shows more variation, diverging in about 6 percent of its nucleotides from the LAV and HTLV-III genomes and encoding proteins that would be about 90 percent identical to those of the other two viruses. Nevertheless, all the viruses are similar enough to be considered variants of the same virus.

The nucleotide differences are not evenly distributed across the viral genomes. They appear to be concentrated in the *env* gene, which codes for the major protein on the exterior of the viral particle. According to Levy, the *env* genes of ARV and HTLV-III differ by more than 20 percent. In addition, the Gallo group has sequenced another HTLV-III isolate and finds that it differs from the first by about as much as ARV does. The greatest divergence is again in the major exterior protein, according to Flossie Wong-Staal of NCI.

This concentration of differences in the *env* gene may complicate the development of an AIDS vaccine. The envelope protein is the likely target of the protective antibodies that would be generated by a vaccine. If the protein varies too much from one virus to the next, a single vaccine might not be able to confer immunity to all strains.

Moreover, the variability that is now being picked up may indicate that the *env* gene of the AIDS virus is particularly susceptible to mutation, much as are the genes for certain of the influenza virus proteins. In that event, a vaccine might be effective to begin with, but then cease to protect as the virus changed. One question the investigators now want to answer is whether there are conserved sequences in the envelope protein that do not show the variations. If there are and if they prove to be antigenic, then it might be possible to design a vaccine

that will raise antibodies specifically directed against those portions of the protein.

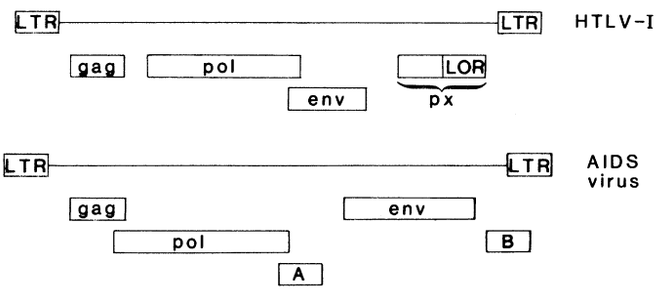
Variation in the *env* gene might also affect the reliability of diagnostic tests for the presence of the AIDS agent. These currently use viral proteins to identify antibodies to the virus in blood samples from infected individuals. The antibodies might escape detection if they were elicited by a viral strain that differed significantly in its proteins from the one used to make the test reagents.

The tests recently approved for use by the Food and Drug Administration miss roughly 5 percent of the individuals who have been infected by the AIDS virus. Preliminary results suggest, Wong-Staal says, that these false negatives are not caused by viral differences but by a lack of immune response in the individual. Needless to say, more work, including more sequence comparisons, will be required to determine the impact of the *env* gene variations on vaccine development and test reliability. The Gallo group is willing to give HTLV-III isolates or clones to researchers who wish to pursue these problems.

The continuing debate over the name of the AIDS virus centers around whether HTLV-III is still an appropriate designation. Gallo and his associates think that it is. Montagnier, Levy, and many others think that it is not. They note, for example, that the biological effects of HTLV-I and -II, which cause T-cell proliferation, are the opposite of the effects of the AIDS virus, which causes T-cell death. As Montagnier puts its, "HTLV-III and LAV are very different from HTLV-I and -II. HTLV-III is not a leukemia virus. I think there is a need for another name."

Nevertheless, Gallo maintains that the HTLV-III designation is appropriate for the AIDS virus, partly because it shares with HTLV-I and -II a strong preference for infecting T cells of the helper class. He points out that he has already changed the original meaning of HTLV from "human T-cell *leukemia* virus" to "human T-cell *lymphotropic* virus" in recognition of the cell-killing propensities of the AIDS agent.

Perhaps the best argument in favor of those who desire another name comes from the analysis of the gene sequences of the AIDS virus isolates, which have now been reported by four groups, including one from Chiron Research Laboratories in Emeryville, California, and another from Genentech, Inc., in San Francisco, in addition to the Gallo and Montagnier groups (*3–6*). All four groups, including that of Wong-Staal, Gallo, and their colleagues, find that the nucleotide sequences of the AIDS virus genome show little resemblance to those of the HTLV-I and -II genomes, except for some short segments primarily in the *pol* gene, which codes for the viral enzyme that copies RNA into DNA. "The AIDS virus is no more closely related to HTLV-I and -II then to Rous sarcoma virus," says Chiron's Paul Luciw.



Genomic organization of HTLV-I and the AIDS virus.

The organization of the protein-coding segments of the AIDS virus also differs from those of HTLV-I and -II. The AIDS virus has a short coding region (box A in the diagram) of as yet undefined function between the *pol* and *env* genes, which is not found in the HTLV's. Moreover, the HTLV's have a long region, designated pX, between the *env* genes and the right-hand long terminal repeats (LTR's). William Haseltine of Harvard's Dana-Farber Cancer Institute and the Gallo group have evidence that the right-hand half of pX, which they have called the LOR (for long open reading frame region), codes for a protein that activates gene expression and may be involved in the malignant transformation caused by HTLV-I and -II.

The AIDS virus genome does not have a comparable pX region, at least according to the Pasteur, Chiron, and Genentech workers, but has a shorter segment with protein-coding capabilities (box B) that extends into the LTR. This segment can encode a protein containing about 200 amino acids, which would make it considerably shorter than the LOR protein, which has a molecular weight of about 40,000.

Haseltine, Wong-Staal, and Gallo also have evidence that HTLV-III makes a protein analogous to the gene-activating LOR products of HTLV-I and -II. In fact, the possibility that all three viruses work by a similar mechanism, even though their final effects are different, is another reason why Gallo thinks that the HTLV-III designation should be maintained.

In any event, the Gallo and Haseltine groups have a different view of the gene organization at the right-hand end of the AIDS virus genome. They propose that the *env* and LOR segments are essentially fused in a single open reading frame that encodes both the envelope protein and the LOR product equivalent. Which of the two possible products will be produced depends on how the messenger RNA that is transcribed from the region is spliced, they suggest. However, according to Daniel Capon, the Genentech group's analysis of the splicing patterns of HTLV-III messenger RNA's does not support such an interpretation, but indicates instead that the open reading frame to the right of the *env* gene encodes a complete protein without *env* sequences. They nonetheless speculate that this protein might have a gene-activating function similar to that of the LOR products of HTLV-I and -II.

As things now stand, Gallo maintains his determination to keep the HTLV-III designation for the AIDS virus. "We never claimed that they [the viral gene sequences] had to be highly homologous." He suggests the possibility of combining names, using HTLV-III/LAV for isolates from Montagnier's laboratory or HTLV-III/ARV for Levy's isolates. This suggestion is unlikely to win many supporters.

Meanwhile, Montagnier proposes that the initials LAV be retained while changing the full name to lymphadenopathy/AIDS virus, now that the connection to the immune deficiency syndrome has been established. "I think that we should keep the name originally given by us because we were the first to isolate it," he asserts. Gallo, incidentally, won the day in an earlier dispute over naming the leukemia viruses because the first HTLV was isolated in his laboratory. Finally, perhaps predictably, Levy is content with the name ARV.

If no one yields, the issue may have to be resolved by an international committee. Harold Varmus of the University of California School of Medicine in San Francisco is currently assembling such a committee in his role as chairman of the Retrovirus Study Group of the International Committee on the Taxonomy of

Viruses. It would include some seven of the regular members of the study group, plus a half-dozen additional participants. Gallo, Montagnier, and Levy are among the proposed members. Varmus plans to solicit naming suggestions and then poll the membership for their preferences. A fourth name, unassociated with any particular group, may be the result.

There are apparently no hard and fast rules for determining viral relatedness. A variety of characteristics, including the size and shape of the viral particles, whether they have RNA or DNA as their genetic material, host range, and biological action, have been used. The ability to determine complete gene sequences, a skill which has been acquired relatively recently, adds a new consideration. According to Varmus, the same genetic principles that have been applied to defining species generally may be applicable to viruses. "Members of a virus 'species' would share genetic characteristics and allow genetic intermingling between members of the same 'species' while resisting the influx of information from other 'species,' " he explains.

Varmus expects that it will be sometime in June before the committee can come to a decision about the name of the AIDS virus. Whether it will be accepted remains to be seen. "These deliberations can be irrelevant to the way people behave," he points out. "Nothing we do is binding. If someone wants to ignore it, he can."—JEAN L. MARX

References

1. L. Ratner, R. C. Gallo, F. Wong-Staal, *Nature (London)* **313**, 636 (1985).
2. M. Alizon and L. Montagnier, *ibid.*, p. 743.
3. R. Sanchez-Pescador *et al.*, *Science* **227**, 484 (1985).
4. L. Ratner *et al.*, *Nature (London)* **313**, 277 (1985).
5. M. A. Muesing *et al.*, *ibid.*, p. 450.
6. S. Wain-Hobson *et al.*, *Cell* **40**, 9 (1985).

Periodic Extinctions and Impacts Challenged

Critics are attacking the evidence that comet showers have caused periodic extinctions; proposed triggers for the showers seem unlikely as well

No one has delivered a knockout punch to the idea put forward more than a year ago that every 28 million years or so a swarm of comets batters Earth and drives as much as 70 percent of animals and plants to extinction (*1*, *2*). But if it has not been knocked out, the hypothesis is certainly falling back under increasing criticism. Many geologists, paleontologists, astronomers, and statisticians who have now had a chance to study the details of the proposals find the geological evidence merely suggestive or even nonexistent and the supposed underlying mechanisms improbable at best (*3*).

Astronomers have just struck a serious blow to one periodic extinction hypothesis by demonstrating that the sun's yo-yo motion through the disk of our galaxy cannot cause the periodicities claimed for extinctions and Earth impacts. In the original proposals, each time Earth bobbed up or down through the central plane of the galactic disk during its 66-million-year oscillation, it would have a greatly increased chance of encountering one of the huge clouds of gas bunched near the plane. Having the mass of hundreds of thousands of suns, the gravitational pull of such a molecular cloud could perturb the comets encircling the solar system and send a billion of them into the inner solar system, a dozen or two of which would hit Earth over a million years or so. The largest impact or the combined effect of the comet shower would then cause the extinctions.

Patrick Thaddeus and Gary Chanan of Columbia University have shown (*4*) to everyone's satisfaction that, contrary to the galactic hypothesis, molecular clouds are simply not bunched tightly enough around the plane of the galactic disk to make much of a difference. The chance of Earth encountering a cloud near the plane is not much greater than at the extremity of Earth's oscillations where the clouds have begun to thin out.

Compiling their own observations and those of others, the Columbia group finds that molecular clouds become half as numerous 85 ± 20 parsecs above or below the plane than they are at the plane (1 parsec = 3.26 light-years). The most distant excursion of Earth from the plane is 72 parsecs. Thaddeus and Chanan then calculated how much that amount of bunching would increase the frequency of encounters near the plane. Not much, they found, and certainly not enough to make the near-plane encounters distinctly more numerous than those away from the plane. As Frank Bash of the University of Texas notes, "There is no real significance to passing through the galactic plane." Thaddeus and Chanan calculate that a record of at least 300 extinction cycles, not the nine that are available, would be required to distinguish the few encounters attributable to plane crossings from the many that can happen at any time.

A remnant of a large impact

A huge impact splattered this glassy microtektite across the globe about 37 million years ago (diameter about 250 micrometers).

As often seems to be the case with periodic extinction hypotheses, there is a way around this difficulty, but it is one that leaves the galactic hypothesis an unlikely explanation of the reported periodicities. Richard Stothers and Michael Rampino of the Goddard Institute for Space Studies in New York, who advanced one of the original galactic models, point out that if the sun's motion perpendicular to the plane were like that of other stars its age—carrying it several hundred rather than 72 parsecs from the plane—the plane-crossing mechanism could work. Molecular cloud specialists agree, but they see little chance that a far-ranging, high-speed sun was suddenly slowed within the past 30 million years by an extremely rare encounter with a molecular cloud. Thaddeus estimates the probability at 1 percent or less. Others find it equally unlikely.

Another mechanism for driving periodic extinctions by creating comet showers, a newcomer invoking an unseen tenth planet to disturb the comet cloud, has been taking its lumps as well. Even as their model first appeared in print (*5*), Daniel Whitmire and John Matese of the