A Versatile Virus

Epstein-Barr virus displays a few new malignant tricks

By KATHLEEN FACKELMANN

Sharon B. Murphy recalls treating a 4-year-old boy whose immune system had been damaged by AIDS. The youngster also suffered from a rare cancer of the smooth muscle cells.

"To have two rare conditions coinciding in a single patient set off an alarm bell," says Murphy, a pediatric oncologist at Northwestern University Medical School in Chicago. After noticing several similar cases, Murphy and some colleagues did a little medical sleuthing.

They knew about an unpublished report linking the Epstein-Barr virus to this same cancer, so they decided to look for the virus in tissue samples collected from their young patients. "And there it was, son of a gun," Murphy says. "It was quite striking and unexpected."

Epstein-Barr virus belongs to the herpesvirus family. About 90 percent of all adults carry this microbe. For people with a healthy immune system, the virus poses no risk and remains a silent, lifelong companion.

Yet for teenagers who haven't contracted this bug in childhood, Epstein-Barr virus can cause the high fevers and malaise of infectious mononucleosis. In addition, this microbe's curriculum vitae contains several disturbing references to a variety of cancers.

Two teams of U.S. researchers now suggest that Epstein-Barr promotes the development of smooth muscle tumors. Their reports are the first to associate this virus with such cancers. The authors describe their findings in the Jan. 5 NEW ENGLAND JOURNAL OF MEDICINE (NEJM).

In a related report in the Jan. 1 CANCER RESEARCH, a British team links Epstein-Barr virus with breast cancer.

The first data connecting the ubiquitous Epstein-Barr virus to smooth muscle tumors surfaced in a preliminary report presented at a March 1993 meeting of the U.S. and Canadian Academy of Pathology in New Orleans.

Pathologist Paul S. Dickman of Children's Hospital of Pittsburgh and his coworkers had observed three puzzling cases of smooth muscle tumors in young children who had received a transplanted liver. (All three had suffered failing livers and needed a donor liver to survive.) Like any other transplant patients, these children required immune-dampening drugs to ward off rejection of their new organ.

Transplant patients can develop lymphoproliferative disease, in which certain white cells divide to form mostly benign tumors. Scientists believe the tumors arise when immunosuppressive drugs stifle the normally vigilant immune system and the previously dormant Epstein-Barr virus flares into an active infection of the white cells. The virus appears to spur these infected cells to proliferate. Most such tumors seem harmless and go away as soon as patients stop taking their powerful drugs. Yet sometimes the growths turn malignant.

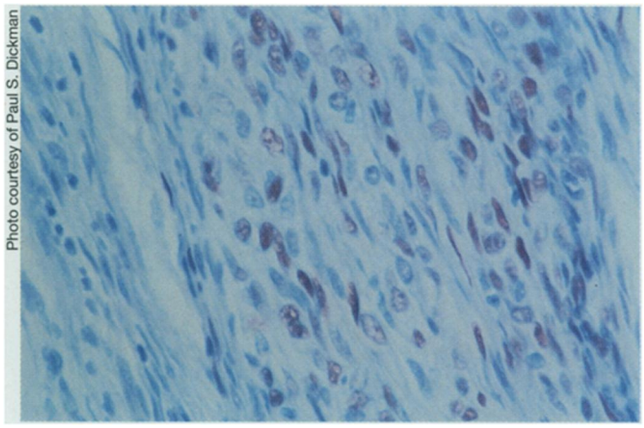

Photo courtesy of Paul S. Dickman

The slide shows a smooth muscle tumor taken from a liver transplant patient. Most cells to the right are stained with a red dye, a sign of Epstein-Barr virus. The cells to the left, which are at the tumor's edge, do not display the telltale mark of this virus.

Knowing that, the Pittsburgh team wondered if the tricky Epstein-Barr virus were behind the smooth muscle tumors they had observed. "So we looked," Dickman says, "and we got lucky." The researchers found Epstein-Barr in tumor samples collected from all three of their young transplant patients.

"It was just ordinary surgical pathology that led to a very nice and interesting finding," Dickman says now.

The Pittsburgh group's report struck a chord with Murphy and her coworkers, who had observed these same smooth muscle tumors in young people with AIDS.

Murphy, Kenneth L. McClain of Baylor College of Medicine in Houston, Charles T. Leach of the University of Texas Health Science Center at San Antonio, and their coworkers had collected tumor samples from five children and one young man with AIDS.

One patient had a relatively harmless smooth muscle tumor called a leiomyoma. Four had the more malignant version of this tumor, a leiomyosarcoma. And one had a leiomyosarcoma and a leiomyoma. The researchers observed these tumors in the lungs, colon, stomach, intestine, and liver of their young patients.

Generally, such tumors strike fewer than 2 children in 10 million. The researchers knew an estimated 5,000 children in the United States have AIDS. Thus the six young patients in their report represented a higher-than-expected frequency of this type of tumor in children with AIDS.

The researchers knew that HIV had crippled the immunity of these children and was undoubtedly responsible for their AIDS. But did HIV cause their tumors? Or did it merely set the stage for the Epstein-Barr virus to infect and cause smooth muscle cells to proliferate?

To find out, the team analyzed the tumors taken from the six patients and looked for signs of HIV and Epstein-Barr.

They could find no evidence of HIV infection in the tumors, yet they found traces of Epstein-Barr in nearly all the tumor cells. In fact, some of the tumors revealed "strikingly high" levels of the virus.

In contrast, they found no evidence of Epstein-Barr virus in normal muscle or in smooth muscle tumors taken from people not infected with the AIDS virus.

"Basically, our data supported the fact that there are high levels of virus in these tumors," Leach says.

Researchers believe the Epstein-Barr virus gains entry to a cell through a particular protein receptor on the cell surface. Tumor samples from the HIV-infected patients had much more of this receptor than did samples from non-HIV-infected patients with smooth muscle tumors.

Finding Epstein-Barr virus at the scene of the crime certainly tainted its reputation. But did this virus cause the tumors? The researchers dug deeper into their case files. In one patient, Murphy and her colleagues found clues that all tumor cells traced back to a single smooth muscle cell infected with the Epstein-Barr virus.

Similarly, the Pittsburgh group found that the tumors of their three patients appeared clonal in origin: All the cells in a given tumor derived from a single muscle cell run amok.

When Epstein-Barr virus penetrates a smooth muscle cell, it leaves behind a characteristic "fingerprint," Murphy explains. Both teams reported that all the smooth muscle tumor cells they studied carried the marauder's mark.

This suggests that viral infection occurs early in the development of a smooth muscle tumor. "Epstein-Barr is way back at the origins of that tumor," Murphy says.

Even more important, the Pittsburgh data indicate that Epstein-Barr virus causes smooth muscle cells to proliferate, notes David Liebowitz of the University of Chicago Medical Center. Liebowitz wrote an editorial in the same issue of NEJM.

Dickman and his colleagues found that the tumor cells contained an Epstein-Barr protein known to spur certain immune cells to divide in laboratory experiments.

Liebowitz suggests that once the Epstein-Barr virus invades a smooth muscle cell, it starts cranking out proteins that signal the cell to grow. That process can lead to a mild proliferation or to an aggressively invasive cancer, he speculates.

So far, all the researchers agree, they lack absolute proof that the virus causes proliferation, which may lead to cancer.

This isn't the first link between Epstein-Barr and cancer. Researchers had previously tied the virus to a nose and throat cancer in China and to Hodgkin's disease, a malignancy of the lymph nodes. And scientists suspect the virus contributes to Burkitt's lymphoma, a cancer seen mainly in the sub-Sahara.

This region of Africa is also known as the breast cancer belt, a curious fact that led a British team to look for Epstein-Barr in breast tumors.

Virologist Beverly E. Griffin of Hammersmith Hospital in London and her colleagues had already shown in the lab that this herpesvirus can infect epithelial cells. Once infected, those cells, which are present in breast tissue, will start to proliferate.

The researchers screened 91 samples of breast cancers taken from patients at Guy's Hospital in London. They also looked at 21 samples of normal breast tissue or of tissue from benign breast tumors.

The team extracted DNA from the samples and looked for Epstein-Barr with polymerase chain reaction, a powerful technique that multiplies pieces of DNA a millionfold. Lo and behold, they discovered evidence of Epstein-Barr viral DNA in 19 (21 percent) of the samples. In contrast, none of the control tissue samples revealed any sign of this potentially sinister virus.

"This is the first demonstration that Epstein-Barr virus can infect smooth muscle cells."

"I was surprised," says coauthor Louise G. Labrecque, a microbiologist at Hammersmith Hospital. Although some clues suggested Epstein-Barr's involvement in this cancer, she considered their chances of finding the link a long shot. Labrecque says that 21 percent may prove a conservative estimate; the team counted weakly positive tumor samples as not having the virus.

The findings lend themselves to several interpretations. One view is that Epstein-Barr is nothing more than an innocent bystander in breast cells that go on to become cancerous.

A second, though highly speculative interpretation, holds that Epstein-Barr virus plays a direct role in the genesis of breast cancer. The scenario unfolds like this, Griffin says: Epstein-Barr virus invades breast epithelial cells and spurs them to divide. Those cells are then left wide open to a second hit, such as a toxin, which causes wild cell growth and cancer.

What about the majority of breast tumors, in which no virus could be found? Epstein-Barr may cause only some breast cancers, she responds.

Alternatively, the virus may act as a hit-and-run carcinogen. Earlier studies by the Hammersmith group show that after it causes cells to proliferate, Epstein-Barr virus can vanish. Yet its effects remain. The epithelial cells continue to divide in the petri dish much longer than expected.

Epstein-Barr may offer researchers an early warning of breast cancer, Griffin says. Her colleagues want to monitor women at high risk of developing breast cancer for signs of an activated Epstein-Barr infection. If they pick up the virus in the bloodstream of a high-risk patient, they'll look more closely for the beginnings of a breast tumor.

Together, these three studies suggest that the familiar Epstein-Barr virus has a more ominous repertoire than previously believed. "It's already associated with nose and throat cancer, lymphomas, and now smooth muscle tumors," Leach says.

In addition, researchers should not discount well-known viruses when confronted with a new tumor, Liebowitz says.

"The present studies teach us that in addition to looking for new agents of disease, we must keep searching for new roles for well-known etiologic agents," he writes in his editorial. "This is the first demonstration that Epstein-Barr virus can infect smooth muscle cells." Liebowitz notes that other studies had shown that the virus can invade epithelial and some types of immune cells.

If researchers can unravel the mechanism by which Epstein-Barr acts in the formation of smooth muscle tumors, they might learn how viruses trigger cell proliferation. "I think this has broader implications in terms of understanding cancer," Leach says.

Finally, such studies point the way to better treatment of virally induced tumors. If researchers can home in on the viral proteins responsible for out-of-control cell growth, drug designers may be able to devise agents to block that process, Liebowitz adds.

Even if the Epstein-Barr virus lies at the root of certain tumors, it almost certainly does not act alone, Griffin says. In smooth muscle tumors, the virus requires an accomplice in the form of a damaged immune system in order to carry out its dirty work. There's no evidence that Epstein-Barr virus wreaks havoc in the smooth muscle cells of healthy people, Liebowitz adds.

As for breast cancer, Griffin believes that Epstein-Barr may kick off the proliferation but that some other agent finishes the task, turning a benignly dividing cell into an aggressor.

Such studies raise the question of just how many cancers start with a virus. No one knows, but Liebowitz believes that the more scientists delve into the matter, the more they'll find. "It may end up that 40 or 50 percent of all human malignancies have some sort of viral contribution to them." □