that tau collects this way when phosphatases, enzymes that remove excess phosphate groups from tau, somehow fail to do their job (*Science*, 10 February 1995, p. 793).

To learn whether trouble at tau's phosphate-binding sites contributes to this problem, Marie Luise Schmidt, a researcher in Trojanowski and Lee's laboratory and the study's lead author, reviewed a set of 59 new antibodies designed by Lee to recognize and bind to these sites. While testing the antibodies on tissue slices from Alzheimer's brains, however, Schmidt found that four of them—especially one called AMY 117—didn't bind to tau at all, but sought out plaques instead.

The team suspected that the purified proteins used to create the antibodies weren't pure—that they contained molecules from amyloid plaques, a glitch they had encountered before. But when Schmidt stained brain slices using both AMY 117 and an antibody to β amyloid, she was astonished to find that the two antibody types gravitated to two different sets of plaques—one of which had never before been glimpsed. The new lesions resemble the amyloid plaques from the outside, but inside they lack the core of β-amyloid protein that traditional staining techniques recognize. So the proteins used to make the antibodies must have been contaminated after all—but with proteins from the AMY plaques, not the amyloid plaques. The existence of the new lesions "couldn't have been suspected without these new antibodies," says Trojanowski.

All 32 of the Alzheimer's brains the team examined exhibited the new lesions, usually close to, but not overlapping, the amyloid plaques. That means Alzheimer's researchers now have an entirely new set of pathological mechanisms to explore. Explains Trojanowski: "It could be that if you sweep away amyloid plaques and tangles and still have these AMY plaques, you would only be getting rid of two-thirds of the symptoms."

The Penn team is currently purifying the AMY 117–binding protein with the goal of cloning the gene encoding it. That could eventually lead to an improved means of diagnosing Alzheimer's based on detecting the protein in the blood or cerebrospinal fluid, and perhaps even to a way of blocking the formation of new plaques.

But will Alzheimer's researchers welcome this new denomination to their increasingly ecumenical field? "Heavens, yes," says neuromolecular biologist Marcelle Morrison-Bogorad, associate director of the Neuroscience and Neuropsychology of Aging program at the National Institute on Aging. "The more angles we have, the closer we'll be to understanding what Alzheimer's actually is."

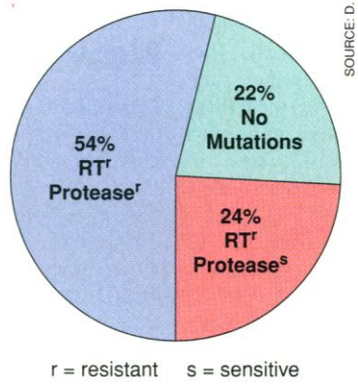
–Wade Roush

AIDS THERAPIES

The Daunting Challenge of Keeping HIV Suppressed

ST. PETERSBURG, FLORIDA—When the media last year trumpeted the great advances being made against HIV, speculating about cures and even the end of AIDS, the researchers' own caveats tended to get drowned out. But at a meeting held here last week, emerging data about treatment failures sounded a discordant note that was hard to miss: While powerful new drug combinations are delaying disease and death, they have serious limitations—and clinicians and patients who ignore these shortcomings do so at their peril.

More than 200 leading AIDS researchers from around the world gathered here from 25 to 28 June for the workshop, which focused on HIV drug resistance, treatment strategies, and the possibility of eradicating the virus from an infected person. One presentation after another reinforced the message that keeping HIV at bay, even with the most potent three-drug cocktails now available, remains a daunting challenge. "Triple combination therapy can fail for a variety of reasons," said John Mellors of the University of Pittsburgh Medical Center, one of the meeting's organizers. As this reality sets in, infected people may end up feeling that their hopes were raised too high last year, he warned. "The pendulum will swing back."



Humble pie. Treatment failures often occur without resistance to reverse transcriptase (RT) or protease inhibitors.

The meeting went into fine detail about why these failures occur. New, more sensitive assays that measure levels of HIV indicate that even the best treatments have a difficult time completely suppressing viral replication, which gives drug-resistant mutants a chance to appear. Less surprisingly, many treatments also fail because patients don't "comply" with therapies that require taking dozens of pills—many of which have serious side effects and dietary restrictions—each day. Although there were encouraging findings about new treatments allowing the immune system to recover if the virus can be suppressed, researchers spelled out just how distant the goal is of completely rebuilding a full range of immune responses in an HIV-damaged body (see sidebar).

Last year's surge in hope was driven by dramatic findings about the wallop delivered by combinations of two drugs that attack HIV's reverse transcriptase (RT) enzyme with one drug from a newer class of compounds directed at the virus's protease enzyme. Several studies showed that such triple combinations—and even some cocktails of RT inhibitors alone—can drive the amount of HIV in a person's blood, the "viral load," down so low that the most sensitive tests could not detect any virus for more than a year in many patients. Researchers warned, however, that just because HIV couldn't be found didn't mean it wasn't there—nor did it mean the virus wasn't replicating.

One of the more disconcerting findings reported here is that, just as researchers feared, the "undetectable" HIV reported last year can routinely be detected with a more sensitive test. A year ago, the most sensitive tests could measure viral levels down to 500 copies of HIV per milliliter of blood. New tests now measure as few as 20 copies per milliliter. Brian Conway, of Vancouver, Canada's BC Centre of Excellence in HIV/AIDS, used such a test in a 151-person study comparing two RT inhibitors to three RT inhibitors. One year after treatment began, 27% of the patients receiving one of the two-drug combos had fewer than 400 copies of HIV. But when the samples were reanalyzed with an assay that went down to 20 copies, only 12% had "unquantifiable" levels. And when Conway ran the test on one sample below 20, he detected the virus three out of 11 times. "A lot of people hear 'unquantifiable,' and they think 'zero,' " said Conway.

John Coffin of Tufts University in Medford, Massachusetts, suggested that researchers consider changing the focus to the number of virally infected cells, which estimates suggest is about 1000 times higher than the HIV copy number detected. So 20 copies, noted Coffin, would equal 20,000 infected cells. "If you say a person has less than 20,000

Recovering From the Ravages of HIV

New anti-HIV treatments are staving off disease and death in thousands of people. But will a patient's immune system actually recover from the ravages of an HIV infection if the virus is kept at bay? On page 112 of this issue, immunologist Brigitte Autran of the Hôpital Pitié-Salpétière in Paris and co-workers show that even severely compromised immune systems can make a significant recovery after state-of-the-art drug treatment has kept HIV levels suppressed for a year. Yet their work also makes it clear that fully rebuilding an HIV-ravaged immune system is a tall order, especially given the limits of treatments available today (see main text).

Autran and colleagues focused on white blood cells, or T lymphocytes, that have a receptor known as CD4 on their surfaces. HIV selectively infects these immune system cells and, both directly and indirectly, leads to their destruction. In time, HIV-infected people are left with so few CD4s that their immune systems can no longer fend off even the wimpiest bacteria, viruses, or fungi. A key benefit widely seen in people receiving potent new treatments is that CD4s dramatically rebound. Yet in all but the healthiest of infected people, CD4s do not return to normal levels. And it's not clear whether patients really regenerate CD4s or just "redistribute" ones that have been sequestered in the lymph nodes and other tissues. This, in turn, determines just how effective the "new" CD4s are at fighting infections.

The Autran group analyzed the CD4s that returned in eight adults taking a powerful combination of three anti-HIV drugs. After 1 year, the drugs had driven down the level of virus in the blood of all of the patients, and CD4 cells had jumped from a mean of 165 cells per cubic millimeter to 327. (The average number in an uninfected person is 900.) But because all CD4s are not created equal, the researchers used other markers on the surfaces of these cells to categorize them as either belonging to the "memory" or "naïve" subset. A memory cell only responds to an invader it has seen before, while naïve cells can launch an immune response—and create memory cells—against newcomers.

During the first 4 months of treatment, returning CD4s were mostly memory cells, and they came back so fast that they were unlikely to be regenerated cells. "The massive increase at the beginning of treatment is redistribution," says Autran. But after that initial phase, the naïve population rose steeply, indicating that new cells were being generated—and providing a more diverse "repertoire" of CD4s able to respond to new invaders, or antigens. "It's a nice, three-color snapshot," says immunologist Donald Mosier of The Scripps Research Institute in La Jolla, California, of the work. "It's the best analysis of which T cells come back after triple-drug therapy that I've seen."

Work by Mark Connors, Clifford Lane, and colleagues at the National Institute of Allergy and Infectious Diseases adds a darker shading to the picture, however. Their study, published in the May issue of *Nature Medicine*, shows that CD4s of HIV-infected people often lack the full range of cell surface proteins that they need to function properly. So even naïve cells, Lane says, may be handicapped.

Scripps's Mosier adds that the chances are "slim to none" that anti-HIV drugs will eventually allow the immune system to completely restock itself with fully functional CD4s, both because of the limits of the drugs and the immune system's ability to fix itself. Says Mosier, "I'd frankly be surprised if you could continue to do this year after year." Autran holds out hope, however. She notes that people not infected by HIV who receive bone marrow transplants lose their CD4s and see them return in the same two-phase pattern her lab observed. "I'm quite optimistic that if we could diminish the level of viral replication enough, we could approach that situation," she says. –J.C.

infected cells, it might give even the most optimistic patient pause," said Coffin.

Considering that many patients have tens of thousands of copies of HIV when they begin therapy, a drop to, say, 400 copies is hardly bad news. The problem, however, is that if HIV is detected, it's replicating and can mutate into resistant strains. Dale Kempf of Abbott Laboratories near Chicago, maker of the protease inhibitor ritonavir, underscored this point. Kempf presented a study that analyzed patients who had failed various ritonavir regimens—including triple drug combos—because viruses resistant to the drug had emerged. He found that the most telling gauge of whether treatment would eventually fail is how low a person's viral load fell. If the therapy knocked HIV down to 200 to 1000 copies per milliliter, viral levels would rise again, on average, 128 days after treatment began. In contrast, for people whose viral levels went below 200—the limit of the assay Abbott used—treatment failed in 199 days. "Very low viral load must be achieved to ensure a durable response, and we don't think 200 is low enough," concluded Kempf.

Douglas Mayers of the Naval Medical Research Institute in Bethesda, Maryland, added yet another eye-opening finding about drug failures, by showing that they often have nothing to do with resistance (see diagram). Mayers's genetic analysis of HIV from 37 patients who failed various RT–protease inhibitor combinations revealed that 22% had no mutations that would make them resistant to any of the drugs. Another 24% were resistant to RTs but not protease inhibitors. "It was a real surprise," says Mayers. "These data suggest that as much as 40% of failure is related to compliance." While the drugs would presumably still work in these patients if they took them, one danger, said Mayers, is that many physicians might see their virus rebounding, assume they had developed resistance, and switch to other drugs. He said failure may also be caused if people have unusually high metabolisms, processing the drugs so quickly that they have too little time to do their work.

Patients who fail one therapy can often switch to other combos of the 11 anti-HIV drugs now on the market. Unfortunately, however, many mutations that confer resistance to one drug will render similar drugs useless. The best hope for people for whom several drugs have failed is new drugs that attack novel HIV targets. But there is a dearth of such drugs in the pipeline, says meeting co-organizer Charles Boucher of University Hospital in Utrecht, the Netherlands. "I'm confident that [these drugs] are not going to be developed in the next 3 to 4 years," says Boucher. "It's going to be a disaster."

Other clinicians at the meeting warned that they already are running out of options for many of their patients who have tried and failed several drug regimens. "I think three-drug therapy is an incredible advance, but our ability to treat people who fail is troubling," said Margaret Fischl of the University of Miami School of Medicine. "We don't have anything to offer them." So in spite of the vast improvement in treatment that the new therapies have brought, there were no trumpets playing victory tunes here.

–Jon Cohen