
HIV and Tuberculosis: A Lethal Convergence

By Jon Cohen

In 2005, researchers in South Africa came face to face with one of their deadliest fears—the emergence of a virulent strain of drug-resistant tuberculosis that quickly killed nearly every person it infected.

More alarming still, the outbreak surfaced in KwaZulu-Natal province, an epicenter of the AIDS pandemic that had hospitals filled with people whose immune systems were already decimated by HIV. As the shocking news made headlines worldwide, Tugela Ferry, South Africa, became ground zero for the lethal convergence of HIV and a new killer, extensively drug-resistant tuberculosis (XDR-TB).

Since the 1940s, antibiotics have cured TB. But the bacterium that causes the disease, *Mycobacterium tuberculosis*, keeps evolving to dodge most drugs thrown at it. In the late 1980s, multidrug-resistant TB (MDR-TB), which requires a more expensive and toxic second-line treatment, became a major health problem.

South Africa has more HIV-infected people than any country in the world and one of the highest rates of TB per capita. Of the 5.4 million HIV-infected people in the country, 250,000 each year also develop active TB, accounting for about one-third of cases of HIV/TB co-infection in the world. It's more dire still in Tugela Ferry, a rural town in KwaZulu-Natal province, where up to 40 percent of adults are infected with HIV and a staggering 80 percent of TB-infected adults also have HIV.

The outbreak of XDR-TB in Tugela Ferry between 2005 and 2006 involved 53 patients at Church of Scotland Hospital. All but one died. Half of these patients only lived 16 days after they were admitted to the hospital—too short a time to even receive the results of the cumbersome test used to diagnose XDR-TB.

Of the 44 patients diagnosed with XDR-TB in the Tugela Ferry outbreak, all were co-infected with HIV. “They were dying like flies,” says Salim S. Abdool Karim, an epidemiologist at the Nelson R. Mandela School of Medicine in nearby Durban. “I initially thought that this

was an isolated outbreak and would burn itself out. I was proven wrong as several hospitals started reporting cases of XDR-TB.” To date, South African researchers have uncovered XDR-TB cases in every province, and there were 468 cases in Tugela Ferry alone by the end of 2008.

“Tugela Ferry was the cataclysmic event that brought this problem to the world’s attention,” says Richard E. Chaisson, an HIV/AIDS and TB specialist at Johns Hopkins University in Baltimore, Maryland. “If it weren’t for that, no one would really be aware of it.”

The tragedy in Tugela Ferry raised critical scientific and clinical questions. How far has XDR-TB spread? Had HIV infection made people more susceptible to XDR-TB, and if so, how? What are the immune responses to *M. tuberculosis* in HIV-infected people? Did the drug resistance develop because people had a TB infection and failed to properly take their medicine, or had they acquired an XDR-TB strain by transmission from others? What enables the lucky few to survive XDR-TB? What’s hampering the development of faster diagnostics, better drugs, and an improved vaccine? What public health measures can slow XDR-TB’s spread in an HIV-infected population?

Perhaps most importantly, Tugela Ferry forced researchers to confront their own shortcomings. “Sorting out exactly how those two pathogens are interacting with each other really has not been studied enough,” says HIV/AIDS immunologist Bruce D. Walker, an HHMI investigator at Massachusetts General Hospital and Harvard Medical School in Boston. “These diseases have always been studied separately, even though clinically they’re completely intertwined. The science has been done independently by different groups that have little interaction with each other.” Walker, with help from HHMI and several other funders, is one of the researchers leading the movement to bridge these gaps.

AN URGENT NEED FOR BETTER TB TESTS

Mycobacterium tuberculosis is transmitted through the air and infects about one-third of the world’s population. It remains latent, causing no harm to 90 percent of those infected. But more than nine million people each year

develop active cases of TB, which typically causes weight loss, night sweats, and lung damage. Nearly two million people die each year from what should be a curable disease, either because they are not properly diagnosed, do not take the right medications, or have drug-resistant strains that defy treatment.

Roughly half a million people worldwide were diagnosed with MDR-TB in 2006; more than 50 countries by the end of last year, including the United States, had reported XDR-TB, according to the World Health Organization. In all likelihood, these numbers seriously underestimate the true scope of the problem. “We have very limited information about the epidemics of XDR-TB and MDR-TB,” says Gerald H. Friedland, an epidemiologist at Yale University who led the study of the Tugela Ferry outbreak. “It’s hard to do battle when you don’t know the size and location of the other army.”

The biggest obstacle faced by epidemiologists and clinicians is that their diagnostic tool kit is rusty, making it difficult to detect tuberculosis infections and drug resistance. “We’re using things we were using 30 to 40 years ago,” says Karim.

Willem Sturm is the interim head of the new HHMI-funded KwaZulu-Natal Research Institute for TB-HIV (K-RITH) in Durban, where he will co-lead a program to develop improved TB diagnostics. “The most important thing we need is a rapid, reliable diagnostic test that not only can tell us whether there’s TB, but at least in KwaZulu-Natal, we need to know what category TB the patient has,” he says. “Is it susceptible, MDR, or XDR?”

To begin the laborious diagnosis of TB, patients first cough up a sputum sample. Lab technicians then stain the sputum with a dye, wash it with an acid, and examine it under a microscope to check for the presence of TB bacteria. The test has a high false-negative rate, and it’s even more misleading in HIV-infected people, who often have little disease in their lungs but life-threatening *M. tuberculosis* infections in their abdomens, brain, and other hard-to-test regions of the body.

If the sputum has detectable TB bacteria under the microscope, it must be grown in culture dishes or special liquid to confirm the diagnosis. Because TB grows slowly, this can take two to six weeks. A third test assesses drug resistance. This test also has serious practical limitations. Again, the first obstacle is time. Technicians must sprinkle the culture dishes with various drugs to determine which

ones can kill the TB growth. This takes several weeks as well. The long process means patients may die from XDR-TB before they receive the results—as happened in Tugela Ferry. This test also is difficult to do in South Africa and many other cash-strapped countries, as it requires special high-tech laboratories that minimize the risk of *M. tuberculosis* escape, as well as elaborate protective gear for the technicians handling the material.

Ultrasensitive tests such as the polymerase chain reaction assay (PCR) that amplify tiny bits of genetic material have had limited success in diagnosing TB. Two diagnostic tests for *M. tuberculosis* DNA are now on the market, “but while they are much faster than culture, they’re not as sensitive,” Chaisson says, adding that something in sputum apparently inhibits the PCR reaction.

Last year, the World Health Organization endorsed two PCR-based tests, called “line probe assays,” for determining drug resistance. They work fast—within 8 hours—but can only find a limited number of TB-drug resistance mutations in a strain of *M. tuberculosis*. Sturm says right now, the high-quality laboratories needed to run line probe assays in KwaZulu-Natal are in short supply. Making matters worse, these assays do not detect XDR-TB—which is tough to diagnose, even with the best culture tests.

William R. Jacobs Jr., an HHMI investigator at Albert Einstein College of Medicine in New York, says a key drawback of these tests is that they can only find what they’re looking for—and identifying drug-resistance mutations in TB is slow, tedious work. Isoniazid, the most commonly prescribed first-line drug for TB, and the best understood of the dozen or so drugs used to treat TB, has been used since 1952. It wasn’t until the early 1990s that researchers—including one team led by Jacobs—reported how isoniazid actually works and subsequently began finding mutations that make the bacteria resistant to the drug. And Jacobs notes that no mutations have yet been found for about 15 percent of *M. tuberculosis* strains that can dodge isoniazid.

Jacobs says the field badly needs simple, fast diagnostics that researchers and public health workers can use anywhere. He plans to study drug resistance and develop novel diagnostics with Sturm at the new K-RITH laboratory. It requires a tremendous amount of work to prove that a mutation causes resistance. “Even if you find a mutation, you’ve got to transfer it from the drug-resistant strain to a drug-sensitive strain,” he says.

“That’s the only way to prove that resistance is bona fide.” That process again requires growing cultures of the mycobacteria, which can take a few months. And strains that develop drug resistance are, by definition, less fit—otherwise the mutant would have occurred naturally—and are notoriously slow growers.

For several years, Jacobs’ lab has worked on developing inexpensive tests that sidestep many of these problems. In an interesting strategy that turns the biological tables on the tubercle bacillus, Jacobs’ lab has been trying to develop highly sensitive diagnostic tools with the help of viruses that grow in healthy bacterial hosts. Jacobs engineered reporter genes into the virus genome that, when expressed, make the host glow if the viruses grow; glowing bacteria in the presence of a drug provide a quick and highly sensitive assay for drug-resistant bacteria. He also hopes his phage system will make it easier for researchers to detect tiny amounts of *M. tuberculosis* in a sputum sample, identifying many infected people now missed by the current diagnostic tests.

PROSPECTS FOR BETTER TREATMENTS

In an ideal situation, doctors would quickly identify both HIV and *M. tuberculosis* infections, assess whether a person harbors drug-resistant pathogens, isolate patients who have MDR- or XDR-TB to reduce the risk that they would infect others, and then prescribe the appropriate drugs. The clinical reality is vastly more complicated and confusing.

Many people who have relatively intact immune systems and no knowledge that they are infected with HIV seek medical care because they’re having night sweats and losing weight. Clinicians frequently do not suspect that those people are infected with HIV, let alone TB. And even if they do tests to confirm that their patient is HIV positive, physicians may still miss underlying TB infection. So those patients may receive antiretroviral drugs for HIV, but no TB medication. “If you don’t treat TB in people with HIV, they die,” says Chaisson.

For patients who test positive for both TB and HIV, many clinicians choose to first bring the TB under control. In 2008, Karim headed a study of more than 600 people that showed that patients waiting to complete TB treatment before getting HIV drugs were 50 percent more likely to die than those being treated for both diseases simultaneously. “At the 2009 Conference on Retroviruses and Opportunistic Infections, this was one of the biggest findings in HIV and TB,” he says. “It will save thousands of lives.”

Multidrug-resistant TB in people with HIV infection adds its own challenges. HIV speeds the course of disease in tuberculosis. It can reactivate a latent infection with *M. tuberculosis* or make it easier for a person to become infected by a new TB strain. Treatment of regular TB requires six months of one pill that combines several drugs. Treatment for MDR-TB requires two years of treatment with different toxic drugs, some of which must be injected. People with XDR-TB have fewer options still, as they typically do not respond to several second-line drugs. The options are reduced even further when patients are infected with both XDR-TB and HIV.

Tugela Ferry laid bare the challenges faced by clinicians combating XDR-TB. *M. tuberculosis* evolves drug-resistance mutations in part because infected people do not properly take their medications. But the surge of XDR-TB at Tugela Ferry appears to have a different engine behind it. Sturm’s lab analyzed the XDR-TB infections in patients in an HIV treatment program at the Church of Scotland Hospital, and a dominant strain popped out, indicating that “primary” or transmitted infection rather than spotty TB treatment was driving the epidemic. What’s more, a careful analysis of 17 patients who were being treated for TB infections and later developed a second MDR- or XDR-TB infection found that all had been infected by a new strain of TB.

“This has substantial implications,” says Karim. TB recommendations have emphasized strategies, such as directly observed therapy, to reduce the chances of missed doses and the development of resistance. There’s also been a growing call to give HIV-infected people isoniazid as a prophylactic to prevent reactivation of latent TB. But if new infections are the main driver of XDR-TB’s spread, the emphasis switches to old-fashioned, public health tactics such as patient isolation, face masks, and ample ventilation.

Since XDR-TB surfaced in Tugela Ferry, clinicians have become more aggressive about identifying patients with XDR-TB, treating them appropriately, and trying to contain its spread. The mortality rate has dropped to 82 percent. “That looks good only in relation to 98 percent, the original report,” cautions Friedland. “The most important thing at this time is to prevent new infections. Once you’ve got it, the outcome for most is not going to be great.”

Sturm says he’s “very optimistic” that within the next year they’ll have more rapid tests for TB diagnosis and drug resistance. “That will radically change the whole

scene,” he says. “It will prevent transmission in health-care facilities, and it will allow us to treat patients immediately with the right combination of drugs. At least with MDR, that makes it much more effective. With XDR, it’s more difficult. We need new drugs.”

Although a few new antibiotics are in TB clinical trials, none are likely to complete testing before 2012. In the meantime, Sturm plans to begin a clinical trial of a combination of two drugs already on the market, meropenem and clavulanate. Test-tube studies recently showed they had remarkable power to kill 13 different XDR-TB strains.

Yet even if any of these drugs pan out, curing a drug-resistant infection likely will still take many months or even years, which gives *M. tuberculosis* ample time to devise ways around the new weapons. “You can’t win easily against TB,” says Karim. “You solve the problem and it creates another one.”

VACCINES

A vaccine is the Holy Grail in TB research, as it might one day send the scourge into obscurity, as with smallpox and polio. But this likely won’t happen anytime soon, according to HHMI’s Walker. For one thing, progress to define the immune responses that protect a person from TB has been slow. “Very little immunology has been done,” he says.

The TB vaccine, Bacillus Calmette-Guérin (BCG), has been in use for 80 years and has been given to about three billion people. The vaccine contains a weakened version of *Mycobacterium bovis*, which causes TB in cattle. But BCG has a checkered history, working in some studies and completely failing in others.

“Nobody knows what protects us from TB,” says Chaisson. “And that’s important. If you’re developing a TB vaccine, you want to know what you’re trying to promote.”

Several TB vaccines are in early human studies, but all aim to fortify the old-fashioned vaccine rather than to trigger novel immune responses. To that end, Walker recently

tried to shed light on one of the most vexing mysteries: How does the immune system contain a TB infection? He looked at HIV-infected people in Durban who also had occult *M. tuberculosis* infections. He hoped that examining the interaction of both pathogens would reveal a clearer picture of the relevant immune responses. “The immune system must be doing something to contain TB,” says Walker. “When it fails, you get reactivation.”

CD4 T cells form the basis of immunologic “memory” to orchestrate an immune response if they have earlier confronted an invader like TB. Walker’s study indicates that CD4s work overtime to contain the TB infection. HIV, in turn, specifically targets and destroys CD4 cells, and he found that people with higher levels of the virus had more impaired TB-specific CD4 responses. A new study will ask whether treating an HIV infection can restore these TB-specific responses and prevent that disease.

Genetic differences in HIV-infected people can determine how their immune systems handle that infection, according to Walker’s work, and he plans to explore whether similar forces control susceptibility to TB infection and disease. These genetic factors, Walker contends, may affect both immunologic memory and more primitive innate immune responses that react to classes of pathogens rather than specific invaders. People who have survived XDR-TB similarly may help clarify whether these lucky few had unusual genes and more robust immune responses.

HIV now infects an estimated 33 million people worldwide, one-third of whom already are infected with *M. tuberculosis*. Most of these infections remain contained, but for how long? And as Tugela Ferry dramatically demonstrates, HIV leaves people much more susceptible to new infections with more dangerous strains of *M. tuberculosis* than humans have ever faced. “Tugela Ferry was a wake-up call,” says Jacobs.



Jon Cohen is a correspondent with *Science* magazine and the author of *Shots in the Dark: The Wayward Search for an AIDS Vaccine*.