

Slippery target. Trypanosomes are parasites that cause several tropical illnesses, including Chagas disease.

Now, after more than a decade of follow-up work, McKerrow and colleagues have received permission from the U.S. Food and Drug Administration (FDA) to launch a phase I safety trial of that compound, dubbed K777, in the United States next year. Palmer, currently director of drug discovery for Biota, a biotech based in Melbourne, Australia, is as pleased as a new parent. “It’s like my baby,” he says, but “someone else has been raising it.”

Along with conditions such as African sleeping sickness and leishmaniasis, Chagas disease is one of the tropical diseases that drug companies have traditionally overlooked, even though there’s no vaccine against the parasite and efforts to curb the insects that carry it have proved hit and miss. But after years of stagnation, research into new treatments for Chagas disease has picked up. Two other drugs, both converted antifungal compounds, are entering phase II trials to determine whether they clear the parasite from chronically infected people. “It’s been decades since there was a clinical trial of a new drug for Chagas disease, so that is very exciting,” says biochemist Frederick Buckner of the University of Washington, Seattle.

Researchers hope it’s just the beginning. Some are scanning chemical libraries, re-evaluating existing drugs, and probing new molecular classes to identify additional compounds that might combat a disease that kills more than 12,000 people every year. “I’m optimistic that there’s going to be a lot of progress in the next decade,” says Peter Hotez, dean of the new National School of Tropical Medicine at Baylor College of Medicine in Houston, Texas. Still, Hotez cautions that this movement “is all relative” because there were so few advances in Chagas disease treatment for decades.

The cost of neglect

Brazilian physician Carlos Chagas first officially described the disease that now bears his name in 1909. Today, the World Health Organization (WHO) estimates that about 10 million people are infected with *T. cruzi*, making Chagas disease a bigger health problem in the Americas than malaria. And in recent years, the illness has appeared outside of the tropics more and more frequently (see sidebar).

Although the news offers no consolation to patients, Chagas disease is predominantly



INFECTIOUS DISEASES

Drug Developers Finally Take Aim At a Neglected Disease

New and recycled compounds target the parasite responsible for Chagas disease, which afflicts millions in Latin America

In the mid-1990s, Jim Palmer, a chemist then at Khepri Pharmaceuticals in the San Francisco area, saw an opportunity to prevent months of effort from going to waste. He had synthesized a set of molecules that block cysteine proteases, a class of protein-snipping enzymes. But the blockers weren’t panning out for chronic conditions such as psoriasis and osteoporosis, which were Khepri’s main targets.

Palmer knew somebody who might be able to use the compounds, however. His friend James McKerrow, a biochemist at the University of California, San Francisco, was hunting for molecules that suppress cruzain, a cysteine protease crucial for the microscopic parasite *Trypanosoma cruzi*, the cause of Chagas disease. Millions of people in Latin America carry this insect-borne pathogen, often unwittingly because the initial symptoms of Chagas disease are usually mild. Yet the parasite continues to breed inside the body, and years or decades after becoming infected, about 20% to 30% of people begin to show signs of serious ill-

ness, most often damage to the heart that can lead to heart failure and death.

Although pharmaceutical companies don’t usually share their intellectual property with outsiders, Palmer—with approval from his superiors—handed over his protease inhibitors to McKerrow and colleagues for testing. One was quite good at killing *T. cruzi* in the lab dish, so Palmer made a more potent version for McKerrow.



Under glass. Blood-sucking assassin bugs, such as this species that lives in Central and South America, spread Chagas disease.

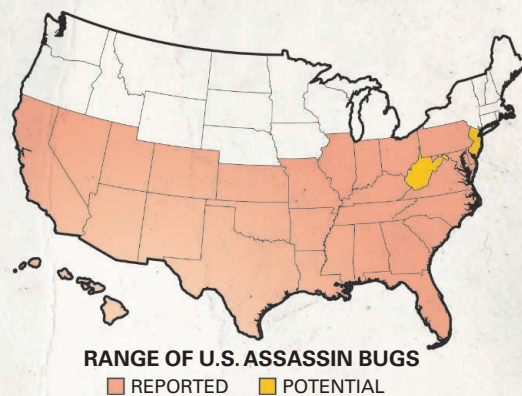
A Tropical Disease Hits the Road

Generally considered one of the many neglected tropical diseases, Chagas disease has been drawing increased attention (see main text), which some attribute to its growth outside of Latin America. Spain and the United States are seeing the highest incidence among developed countries, but there have been cases as far afield as Switzerland and Japan. The main reason for this rise isn't the spread of insects carrying *Trypanosoma cruzi* but rather emigration from Latin America of large numbers of people who are already infected.

Because no one has performed comprehensive testing, the number of infected people living in the United States remains unknown, says medical epidemiologist Caryn Bern of the U.S. Centers for Disease Control and Prevention in Atlanta. To improve on previous projections, she and her colleague Susan Montgomery combined new figures on the prevalence of the disease in different Latin American countries with data on immigration into the United States from those nations. Their results, which they published in 2009 in *Clinical Infectious Diseases*, suggested that about 300,000 immigrants in the United States are likely infected. "It's very much an estimate. We have almost nothing in the way of direct data," Bern says.

Some small-scale surveys have gathered direct data, however, and they point to a fairly high prevalence in areas with large expatriate populations. Cardiologist Sheba Meymandi of the University of California, Los Angeles, School of Medicine and colleagues have run blood tests on patients with heart failure and other cardiac problems, and on Latin American parishioners from several Los Angeles-area churches. The researchers found that about 1% of the approximately 2000 people they tested harbor *T. cruzi*. "One in 100, that's pretty substantial," Meymandi says. Similarly, earlier this year in *PLoS Neglected Tropical Diseases*, a Spanish group testing Latin American immigrants at a clinic in Barcelona reported that 3% were positive for the Chagas parasite.

An unknown is how many infections in the United States result from native assassin bugs. They live from coast to coast, ranging as far north as Illinois and



Pennsylvania, and *T. cruzi* is prevalent in them. One study found that more than 40% of the assassin bugs in Tucson, Arizona, harbored the parasite. However, since the 1950s, the United States has recorded only a few definite cases of parasite transmission. Most researchers think that the risk from these insects is low, in part because homes in the United States are typically not congenial to the bugs. However, Hotez and some other researchers suspect that such transmission is more common than generally accepted. Meymandi notes that she recently diagnosed two teenage patients who were infected in Los Angeles. "It is here; people do acquire it," she says.

Whatever its source, the rising incidence of Chagas disease has already required action by health officials in the United States and other developed nations. Although *T. cruzi* is usually bug-borne, it can also spread through blood transfusions and organ transplants. U.S. blood banks began voluntary testing for the parasite in 2007, and facilities in France and Spain have done the same. And developed countries can expect that the illness's toll will rise. Bern and Montgomery calculated, for example, that if their estimate of the number of infected people in the United States is close to the mark, undiagnosed Chagas disease is responsible for 30,000 to 45,000 cases of cardiomyopathy, or severe heart damage.

—M.L.

an animal disease, says epidemiologist Uriel Kitron of Emory University in Atlanta. The parasite primarily infects wild mammals such as armadillos, opossums, rats, and raccoons, as well as dogs and cats. Spreading the disease are the aptly named assassin bugs, also called cone-nosed or kissing bugs, which feast on blood at night. They usually transmit the parasite to humans through their feces, which people inadvertently rub into a bite or into their eyes or mouth. Like many other neglected tropical diseases, Chagas disease picks on the rural poor. It's prevalent in areas with adobe and thatch homes that offer plenty of crevices where the bugs can conceal themselves during the day.

Chagas disease resembles other neglected tropical diseases in a second way: Current drugs are old, noxious, and impractical. Although not as harsh as the main treatment for African sleeping sickness, which kills 5% of patients who receive it, the only two Chagas medicines, nifurtimox and benznidazole, can trigger serious side effects. Nifurtimox sometimes results in anorexia and nerve damage, whereas benznidazole can spur

skin inflammation that may be so severe it kills. Moreover, a course of the drugs lasts 60 to 90 days, and many patients never finish such a lengthy treatment. "The bar has been set pretty low" for any potential new drugs, McKerrow says.

Both compounds can rid a person of the parasite during the early, acute phase of the disease. The catch is that few people are aware they need treatment at this stage. So for most people, "this infection is for life," says Julio Urbina, director emeritus of the Venezuelan Institute for Scientific Research in Caracas. Whether nifurtimox and benznidazole help the majority of Chagas patients who have chronic infections remains controversial, McKerrow says. A large trial to find the answer for benznidazole is running in several South American countries.

Another motivation for developing new drugs is that parasite transmission continues, despite intensive attempts to curb it. Efforts to control parasite-carrying bugs, involving measures such as spraying infested houses and outbuildings with insecticides, have all but eliminated new cases of the disease in

countries such as Chile, Uruguay, and Brazil. Still, as epidemiologist Ricardo Gürtler of the University of Buenos Aires notes, the problem "hasn't been conquered." He points to the Gran Chaco region of Argentina, Bolivia, and Paraguay, one of the areas hardest hit by Chagas disease, where the insects bounced back after spraying stopped. And in some parts of Latin America, he adds, they have evolved resistance to the insecticides used to control them. WHO estimates that partly because of such setbacks, about 60,000 people still contract Chagas disease every year.

Researchers have also identified a disturbing new way that the parasite can infiltrate the body: through food and drinks, particularly fruit juices. A frightening example came to light in 2007, when more than 100 pupils and staff at a school in Caracas fell ill after they drank guava juice tainted by the bugs' feces.

Reopening the pipeline

If the prospects for new Chagas drugs are looking up, researchers say that part of the credit belongs to the Drugs for Neglected

Diseases initiative (DNDi), a nonprofit organization based in Geneva, Switzerland. DNDi describes its approach as “virtual” because it has no labs and carries out no research. Instead, it serves as a project manager, shepherding promising drug candidates through development by contracting labs, companies, and organizations that can take on different steps in the process, such as screening compounds or running clinical trials. DNDi’s funding comes from a variety of donors, including the Gates Foundation, the European Union, and Doctors Without Borders.

DNDi’s plan calls for at least one new approved drug to treat chronic Chagas disease by 2014. Along with Palmer and McKerrow’s K777, the organization is sponsoring work on E1224, a compound designed by Eisai Pharmaceuticals of Japan. In the body, E1224 transforms into ravuconazole, a molecule currently in phase II trials to determine whether it suppresses fungal infections. Under DNDi’s auspices, a phase II trial of E1224’s potency against chronic Chagas disease will begin later this year in Bolivia. A non-DNDi project led by the Vall d’Hebron Hospital in Spain is testing the approved fungus-killing drug posaconazole against chronic Chagas disease. Merck, the drug’s manufacturer, is also sponsoring a trial on chronic Chagas patients in Argentina.

Why are researchers so keen on antifungal drugs for a parasitic disease? The answer goes back more than a decade, when Urbina and colleagues went searching for existing drugs that could disrupt the synthesis of certain sterols, members of the same chemical family as cholesterol, that the parasite and fungi can’t live without. One reason that posaconazole worked particularly well is that it enters the organs where the parasites take refuge, Urbina says. Studies confirmed that posaconazole and ravuconazole, both of which inhibit a key enzyme required for sterol synthesis, combat *T. cruzi* in animals.

Three new drug candidates might seem like cause for celebration, but Chagas researchers caution against too much optimism. “If you look at the history of drug development, more than 50% of drug candidates fail when they go into people,” McKerrow says. “We need to have the next generation of compounds ready to go.”

Several labs and organizations have started hunting for those compounds. DNDi,

for example, has enlisted researchers at institutions such as Murdoch University in Perth, Australia, and the Federal University of Ouro Preto in Brazil to screen new compounds for antiparasite prowess. One project has focused on oxaboroles, boron-containing molecules that have already shown promise against the parasite that causes African sleeping sickness, says molecular biologist Eric Chatelain, head of DNDi’s Chagas discovery and preclinical program.

Parasitologist Ana Rodriguez of New York University Langone Medical Center and a team of other scientists also recently completed the first high-throughput screen

company Sanofi-Aventis to further develop these potential drugs. One step they need to perform is what’s called drug optimization, tweaking the structures of the molecules to ensure that they can be taken orally, remain stable, and have other favorable qualities.

Posaconazole and ravuconazole suggest that additional compounds that could attack the parasite are already under development for other uses or are even on pharmacy shelves, Buckner says. He and his colleagues have begun to screen FDA-approved drugs for their ability to kill *T. cruzi*. They’ve also been modifying tipifarnib, a cancer treatment that reached phase III clinical trials but wasn’t successful enough to make it to the market; the compound shows promise against *T. cruzi*, Buckner says.

Medicinal chemist Michael Pollastri of Northeastern University in Boston says that the trick to faster discovery of drugs reusable against Chagas involves identifying enzymes that we share with the parasite and that have already been targeted by drug designers. Take the phosphodiesterases, enzymes that control communication within cells. A number of medicines that block human phosphodiesterases, including Viagra, are on the market, and drug researchers have scrutinized many other compounds with similar action that never quite made it through development. By retesting those existing medicines and compounds, scientists might uncover parasite killers, he says.

Many factors could still derail progress toward finding new drugs for Chagas disease. One of the biggest obstacles is the lack of a practical way to determine when a drug is successful in humans, says parasite immunologist Rick Tarleton of the University of Georgia, Athens. “We don’t have an endpoint indicator that says, ‘Ha, that cures people.’” The gold standard—disappearance from the blood of antibodies against the parasite—can take 10 years or more, which makes it unfeasible for drug trials. One of DNDi’s projects aims to find new biomarkers that can more quickly signal when the pathogen has been vanquished.

Despite such concerns, Chagas disease researchers are more upbeat than they have been in years. And yet they are also impatient. “It’s like we are going from zero to 20 [miles per hour],” Hotez says. “It’s not zero to 60.”

—MITCH LESLIE



Warning signs. A poster cautions against the danger from assassin bugs, which hide out in rundown houses during the day and emerge at night to bite the inhabitants.

for Chagas drug candidates, assessing more than 300,000 molecules from a standard National Institutes of Health chemical library. By testing each compound to determine whether it slays the parasite and spares mouse cells, they have tried to weed out obviously toxic molecules. Rodriguez and colleagues eventually whittled the list to seven compounds that killed the parasite in mice.

That was the easy part. “The work really starts now,” Rodriguez says. If they can obtain funding, she and her colleagues hope to partner with the Swiss pharmaceutical