LATIN AMERICA

A Personal Technology Transfer Effort in DNA Diagnostics

In 1987, Eva Harris took a fateful detour from what seemed to be a clearly mapped path to a conventional research career. A 22-year-old with a freshly minted undergraduate degree in biochemistry from Harvard University, she was set to enter the biology graduate program at the University of California (UC), Berkeley. She had even secured a prestigious National Science Foundation predoctoral fellowship. But at the last moment, she decided that straight and narrow path was not for her. She turned down the NSF fellowship, asked Berkeley to defer her admission for a year, spent 6 weeks in Spain learning Spanish, and headed for Nicaragua to see if she could put her biochemistry skills to work in a developing country. "I was driven to find some way I could make science relevant and meaningful for me," says Harris.

That detour led directly to a distinctly unconventional—and hectic—double career. While completing her Ph.D. in yeast genetics at Berkeley and beginning a postdoc at UC San Francisco, Harris has been the prime mover behind a unique effort to bring the polymerase chain reaction (PCR) and other modern DNA-based techniques to some of the poorest countries in Latin America.

As a direct result of Harris's work, scientists in Nicaragua and Ecuador are beginning to use DNA diagnostics for the first time to identify the organisms that cause diseases such as leishmaniasis and tuberculosis. If these initial steps can be followed through, the rapid DNA techniques should greatly speed the diagnosis and tracking of diseases. Public health officials in these countries now have to wait many weeks while pathogens are cultured and sent to foreign labs to be typed. The key now will be whether the new techniques can be integrated into the public health systems.

Harris' successes so far, however, have put her in great demand. Next month Harris, now a postdoc with parasitologist Nina Agabian at UC San Francisco, will be teaching a DNA diagnostics workshop at an international biotechnology conference in Havana, Cuba; she has courses planned for next year in Honduras and Ecuador; and she has been invited to teach in Brazil, Bolivia, and Argentina. Harris has also attracted the interest of the American Society for Biochemistry and Molecular Biology, which supported a recent course she gave in Ecuador. For several years, the ASBMB has run a program to bring modern technologies to Latin America, but it made few inroads into the least developed countries before Harris's Ecuador course, says San Diego State University biochemist Stephen Dahms, chair of the ASBMB's Latin America subcommittee. "Eva has allowed us to have a rapid and broad impact," he says. Adds Richard Cash of the Harvard School of Public Health, who directs the Applied Diarrheal Disease Research Project funded by the U.S. Agency for International Development: "She has introduced a state-of-the-art technology, and the people there are using it to solve local problems. And she did this on an absolute shoestring."

Quick start. Harris, an animated woman whose conversation is a cascade of enthusiasm, didn't have such grand plans when she first set foot in Nicaragua in 1988. Her sponsor then was a Berkeley-based organization called Tecnica (now defunct) that sent technical volunteers—mostly computer scientists—to Nicaragua and South Africa, usually for 2 weeks. But Harris was a biochemist who wanted to stay for several months. "They didn't really have any place to put me," she says. "They literally just dropped me off at the Ministry of Health in Managua. There were roosters running everywhere. I barely spoke Spanish; nothing had prepared me for this. It was the most frightening experience of my life."

KAREN R. PREUSS

Double life. While getting a Ph.D. in yeast genetics, Eva Harris brought PCR to some Central American countries for the first time.

Within a few weeks, however, Harris was teaching a daily course in technical English, giving a weekly seminar on biochemistry, troubleshooting a test for endotoxins in blood plasma for a nearby plasma factory, and helping to work out a technique for identifying different strains of *Leishmania* based on the migration of proteins in gels. Nicaragua has several *Leishmania* strains that cause conditions ranging from self-limiting skin lesions to deadly destruction of internal organs, and a rapid and reliable way to differentiate the strains was needed to trace their spread and determine which patients should be treated with the toxic heavy-metal therapies needed to kill the parasite. "She is a motor; she is always moving and doing things and proposing things," says Alejandro Belli, director of the department of parasitology at Nicaragua's National Center of Reference Diagnostics, where Harris worked on *Leishmania* typing.

After a busy 3 months, Harris left Nicaragua to begin her graduate work at Berkeley, but with the conviction that she would return. She says she told her prospective thesis advisers, "I'm going to go for a month each year to work in Nicaragua, and if you don't like that, I can go [to another lab]." Jeremy Thorner agreed to her terms, and she took up a project in his lab using yeast genetics to study the calcium-binding protein, calmodulin.

She didn't just confine her technology-transfer work to 1 month a year, however. When she wasn't experimenting with yeast she was combing the literature for better assay methods for *Leishmania*. The protein assay Harris had worked on during her first visit had not been successful, but her literature search turned up a promising alternative: A researcher at Yale University, Diane McMahon-Pratt, had monoclonal antibodies to *Leishmania*. Harris got her to donate some to try in Nicaragua.

Harris returned to Nicaragua the following summer, armed with the *Leishmania* antibodies. She soon discovered, however, that the antibody test didn't differentiate among the Nicaraguan forms of the parasite. Mulling over that setback late one night in the lab, Harris, Belli, and a few colleagues wondered whether PCR, then a brand-new technique, might help. But none of them—including Harris—had any experience with it.

Back at Berkeley, Harris sought the advice of Cristian Orrego, the Chilean-born director of the DNA lab at Berkeley's Museum of Comparative Zoology, who had taught PCR in Peru. He taught the technique to Harris, and together they planned a 5-day intensive lab course that included among its experiments PCR identification of *Leishmania* parasites.

On course. Harris began calling companies to scare up support, securing donations of equipment and reagents from Gibco/BRL and Amersham, and a $5000 grant from the New England Biolabs Foundation. By the summer of 1991, she was ready to introduce PCR to Nicaragua. She instructed 10 scien-

tists from the Ministry of Health and 10 chosen from Nicaraguan universities in the technique, assisted by Belli, to whom she had taught PCR just the week before. Only one scientist taking the course had ever worked with DNA before. Harris kept things as low-tech as possible. The students made their own reagents rather than relying on kits, and instead of using expensive thermocyclers to repeatedly heat and chill the samples, they manually moved them back and forth between water baths of different temperatures. To avoid the common PCR gremlin of DNA contamination, Harris designated separate work areas and equipment for the preparation of DNA samples and the performance of the PCR reactions.

"Nobody could do it better than she does it," says Stanford University parasitologist Gary Schoolnik, who met Harris when she taught a workshop at Stanford. "She can make use of some of the relatively undeveloped laboratories that she has encountered and find ways of solving problems that most people wouldn't even try to." By the end of the week in that first course, the students were already using PCR to differentiate strains of *Leishmania*, something that had never been accomplished by any means in Nicaraguan labs. "I was on a high for months," says Harris. "To see that this stuff was actually applicable and appropriate just blew my mind."

After the course, Belli's group continued the *Leishmania* work, developing a technique for diagnosing the disease directly from skin biopsies. That accomplishment won them a grant from the European Economic Community to participate in epidemiological studies of *Leishmania* in South America. Says Orrego: "I think you would have met with skepticism if you said that in a year you would have a group of people doing DNA biochemistry for public health there."

Harris planned a second Nicaraguan course for the following July and began working on assays for other important disease organisms, including the TB bacterium, *Mycobacterium tuberculosis*; *Shigella* and enterotoxogenic *Escherichia coli*, which cause diarrhea; and *Plasmodium falciparum*, the malaria parasite. Cholera was sweeping Latin America at the time, and she also included an assay for *Vibrio cholera* DNA in drinking water.

Despite all the work Harris was putting into the courses, she completed her thesis in April 1993. UCSF's Agabian invited her to join her lab to study the basic biology of *Leishmania* while she continued to take her courses to more countries. "I felt I could really make a contribution by being supportive of her taking an alternative path," says Agabian.

Ecuador was the next country in line. Last April, still working on a shoestring, with only $6500 in funding from the ASBMB and some equipment donations, Harris put on a course in Quito for 20 scientists from 13 institutions in eight Ecuadoran cities.

Scientists from that course formed four teams that are now working on pilot studies to apply PCR to endemic health problems in Ecuador. One group is doing molecular detection of tuberculosis in the mountain region of Zumbahua where the incidence of TB is very high, another is working on detecting *Leishmania* from skin lesions, and two are detecting and typing dengue virus. Harris will teach a second-phase course next year in which the groups will evaluate their pilot studies and write grant proposals to get their projects funded.

"She has introduced a state-of-the-art technology ... on an absolute shoestring."
–Richard Cash

Follow-up. Is Harris's effort likely to achieve long-term success? "There is no question in my mind that for public health purposes ... [DNA diagnostics] can be extremely useful," says Stanford's Schoolnik, who has worked for years on diarrheal diseases in Mexico. The question that has not yet been answered, he says, is whether the methods are sustainable in developing countries without continuous outside support. "That would require that the country have the capacity to produce the reagents, because they are costly otherwise, and that there be a critical mass of expertise not only in how the test is done, but in how to use the test to ask the right question in a field situation." Such expertise will only come with faithful follow-up, says Ron Guderian, an American who has spent 20 years doing research on tropical epidemiology at Hospital Vozandes in Quito. Unless that support is provided, he says, "all that [will have] been acquired is false hope."

But follow-up is what Harris is famous for. Belli says that having her as a contact in the United States and a source of advice, reagents, and journal articles has been key to his success in developing *Leishmania* assays. "It is not the courses themselves; it is the constant follow-up of collaboration which makes this [technology] transfer possible," says Belli. Adds Orrego, "She stays with the people. She continues to work with them, and therefore there is growth after the course." Harris, who plans to continue with her own basic research career in the United States, may be leading a double life for some time to come.

–Marcia Barinaga

FUNDING REVERSAL

Cancer Researcher Returns Grant

Ever since researchers spliced together genes from different organisms more than 20 years ago, researchers and ethicists have worried about where genetic engineering may lead. But few have gone as far as molecular biologist John Fagan. Last week, Fagan announced he is returning more than $600,000 in grant money to the National Cancer Institute (NCI) because he no longer wants to be a part of genetic research.

Fagan, chair of the chemistry department at Maharishi International University in Fairfield, Iowa, called for a 50-year moratorium on releasing genetically engineered organisms into the environment, pending further research, in a 17 November news conference in Washington, D.C. He also expressed concern about potential future manipulations of germ-line cells in both animals and humans. "There are people out there who think favorably of the idea [of] ... potential eugenic applications," he said.

Fagan, age 46, has enjoyed 9 years of continuous funding from NCI; his latest grant renewal, for research into cancer susceptibility genes, came in September. He is also in the fourth year of a 5-year NCI Research Career Development Award. Although his former lab chiefs won't comment, two former colleagues call him a "competent" if not world-burning scientist.

Fagan is giving up his research to focus on what he considers to be a more fruitful activity: research on "traditional" medicine, specifically Indian Ayurvedic medicine, which he thinks holds more promise for disease prevention than does gene-splicing. A longtime practitioner of Transcendental Meditation (TM)—the type advocated by Maharishi Mahesh Yogi, founder of his university—Fagan's career change has been germinating for some time. After getting his Ph.D. at Cornell University in 1977, he was a postdoc and later a senior staff fellow at the National Institutes of Health—where, says a former colleague, he also taught an informal course on TM. He moved to Iowa in 1984. Two years ago, he says, a "deluge" of media coverage convinced him that scientists had begun "to promote [genetic engineering] research in an unrealistic way."

The biomedical establishment is taking Fagan's defection in stride. NCI Director Samuel Broder issued a statement saying that his decision "could be in the best interests of all parties if he has lost enthusiasm for his own research."

–Constance Holden