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An Asian Odyssey *Travels of a Berkeley Lab Microbiologist*

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“May you live in interesting times,” goes the (apocryphal) Chinese curse—but interesting times can also bring benefits. For Tamas Torok, a microbiologist in the Life Sciences Division whose search for extremophiles and other novel organisms has led him to investigate such inhospitable environments as the below-freezing depths of Lake Baikal, the geysers and mudpots of the Kamchatka Peninsula, and the Chernobyl exclusion zone—not to mention areas more challenging to humans than to microbes, like war-torn Kosovo—the summer of 2007 was interesting indeed.



The South Caucasus, the Silk Road, and the Russian Far East all figured in Tamas Torok's travels during the summer of 2007. Inset: Vladimir Dmitriev, of the Russian Academy of Sciences' Institute of Biochemistry and Physiology of Microorganisms, left, and Torok collect samples of extremophiles from hot springs in the Kamchatka Peninsula. (All photos by Tamas Torok except inset by H. Smelov)

In the course of a few short months Torok first managed to bring together an unprecedented consortium of scientists from Georgia, Armenia, and Azerbaijan, three countries who have often been up in arms against one another.

Then, via long detours, Torok moved on to Uzbekistan, whose desert environments—which include the bed of the vanishing Aral Sea—have yielded scores of novel microorganisms with prospects for new biotechnology applications.

Next, in Tajikistan, he and his colleagues tried to answer the grim question of why the occurrence of anthrax in that country is orders of magnitude higher than elsewhere among the former republics of the Soviet Union.

Before the summer ended, Torok had visited Moscow on his way to revisit Kamchatka, where a gigantic landslide had abruptly buried many of the sites of his former research on the wild peninsula.

“It was a busy summer,” he admits. Characteristically, however, Torok insists that the most interesting aspects of his travels were cultural and human. Many of the connections he established promise lasting value to science.

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In the South Caucasus

The first of Torok's visits, in June, resulted in an event of historic significance. The South Caucasus Cooperative Research Program (SCCRP) aims to assist research among scientists in the U.S. and Georgia, Azerbaijan, and Armenia. His trip was sponsored by the U.S. Civilian Research and Development Foundation (CRDF), established by the National Science Foundation as a nonprofit, public/private partnership promoting international scientific collaboration through support to scientists in the former Soviet Union. Torok's task was to help establish a joint network of biotechnology resources among the three countries, which boast unique microbial resources in the form of culture collections they hope to make accessible to one another and to the world's research community.

"For example, Armenia is notable for its numerous isolated villages, many of them engaged in sheep and goat herding and consequently specializing in dairy foods and other fermented products; each has a different microbiota that can't be found anywhere else," Torok explains. "Azerbaijan has been a huge petroleum-producing region since the middle of the 19th century, where pollution has resulted in organisms that are very good at biodegrading oil. Georgia was under the ocean for most of the history of the planet, and one result is salty soil that has given rise to microorganisms uniquely adapted to high salinity."

Torok visited institutes in Baku, Azerbaijan and in Yerevan, Armenia and found that their valuable collections were well cared-for but were held in facilities that suffered from a host of handicaps, ranging from simple lack of funds for adequate freezers in Azerbaijan to abandoned buildings and the emigration of trained staff in Armenia.



At left, an ancient Georgian watchtower and river town frame Azerbaijanis fishing in the oil-polluted waters of the Caspian Sea near Baku. At right, prehistoric rock carvings predate a chapel of Armenia's ancient Orthodox Apostolic Church. Inset: Evrik Afrikan (left), former director and scientific adviser to the State Microbial Depository Center of Armenia's National Academy of Sciences, talks with Giorgi Kvesitadze, director of the Durmishidze Institute of Biochemistry and Biotechnology in Tbilisi, Georgia. (All photos by Tamas Torok)

Torok's host and translator in Armenia was Evrik Afrikan, former director of the Armenian Institute of Microbiology, whose work had inspired Torok decades earlier, when he was a student in Berlin. Torok found Afrikan, now 84, in robust health despite never having given up alcohol or tobacco. From him Torok learned of the natural and political disasters that had battered the nation: devastating earthquakes, war with Azerbaijan, economic embargo by Turkey. The causes are partly rooted in religious strife. Armenia's neighbors are characterized by combinations of Orthodox Christianity, Islam, secularism, and other religious preferences. Unique among them, most of Armenia's inhabitants belong to the Orthodox Apostolic Church, established decades before Constantine founded the Roman Empire in the East.

In Tbilisi, capital of Georgia, Torok called a meeting of representatives from all the institutions participating in the SCCR microbiology project. It was the first time most members of the Azerbaijani and Armenian teams, apart from their leaders, had ever met. "Because of religious differences, wars, uncertain ceasefires, occupations, there had been no connection between these two groups," Torok says. "It was the first opportunity for them to compare problems and opportunities and make coordinated plans."

Much of the meeting was given over to training sessions conducted by Torok. "These researchers are well trained classically, but have not had an opportunity to learn modern methods of molecular biology or to

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practice with modern equipment.” Other results included agreement on goals and methods for systematic collection of microbes from specific environments, efforts to rationalize the exchange of materials in the context of international agreements, and new efforts to publicize their resources and expertise, including participation in meetings of the World Federation for Culture Collections.

“I also made sure we included computer guys in the project so we could build a website and a portal for information,” Torok says. As a result, until communications capacity and reliability in the region can be improved, a website for the SCCRP will be housed on a Berkeley Lab server and mirrored in each of the participating institutes, whose own websites will also be updated. Through the SCCRP portal researchers worldwide will eventually be able to search strains of unique organisms and make arrangements for their use in any of five United Nations languages.

Torok’s trip to Georgia included visits to other institutions, including the Eliava Institute of Bacteriophage Microbiology and Virology. Bacteriophages are viruses that attack bacteria, offering an alternative method of combating drug-resistant infections; with over 70 years of research in the field, the Eliava Institute has gathered unique experience but has languished since the collapse of the Soviet Union. Here Torok had come upon a technology that could be of great benefit to one of humankind’s fastest growing problems, a research and development project, he says, that is “crying out for international scientific and financial support.”

Along the Silk Road

In July Torok returned to Central Asia under the auspices of the US Department of Energy’s Global Initiatives for Proliferation Prevention and the International Science and Technology Center (ISTC) in the Russian Federation. Torok’s first stop, after changing planes in Frankfurt and Istanbul, was the Institute of Microbiology of the National Academy of Sciences in Tashkent, capital of Uzbekistan.

“DOE has supported the collection of hundreds of novel bacterial strains in Uzbekistan, whose wild mountain and desert geography includes a number of extreme environments,” he explains. Many of the organisms in the Institute’s collection are specially adapted to salt and aridity; of the 800 specimens collected under DOE sponsorship, Torok says, “About 150 of them are most interesting in terms of their extremophilic features and the natural products they express.”

While in Uzbekistan Torok visited two ancient cities, Bukhara and Samarkand—the latter already a thriving center when Alexander the Great conquered it in 329 BCE—whose prosperity owes much to their locations along the Great Silk Road between China and the West. Exquisite architecture, Bukhara’s in patterned brick (perfectly preserved in the desert air), Samarkand’s in blue tile, kindles reminiscence of *The 1,001 Nights*.



Along the Silk Road: in Bukhara, mosques, shrines, and minarets are formed of extraordinary brickwork, a striking contrast to Samarkand’s renowned decorative tilework.

(All photos by Tamas Torok)

Central Asia has been less romantic in recent decades, however. The Aral Sea, straddling the border of Uzbekistan and Kazakhstan, was once the world’s fourth-largest inland body of water but has shrunk disastrously since the rivers that fed it were diverted to irrigate cotton. An infamous Soviet bioweapons laboratory, once isolated on Vozrozhdeniye Island in the Sea, has since become connected to the mainland;

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attempts by the Uzbekis, supported by the U.S., to clean up weaponized pathogens including anthrax, bubonic plague, and salmonella have been minimally successful and pose a formidable challenge to the country's Center for Prophylaxis and Quarantine, as well as to the Institute of Epidemiology and Microbial Infectious Diseases, another Uzbeki institution that seeks to control infections such as those spread by nomadic animals and humans wandering across the country's remote, extensive borders.

Of all the establishments he visited, Torok found Tashkent's Institute of Microbiology to be "the least unfortunate among them." The situation seems unlikely to improve, however. Since the U.S. was critical of the dictatorial government's suppression of protests in 2005, many U.S. programs have been forced to terminate. At present, remaining programs are scheduled to end by December 2008; if this happens, impacts on the country's scientific programs, and possible impacts on international health, will be severe.

Torok's next stop was Tajikistan, its capital only 300 kilometers from Tashkent as the crow flies—except that airplanes don't fly that way, because of tensions between the countries. Instead, the trip requires a five-hour flight to Istanbul and another five-hour flight back. "It's like flying from San Francisco to Portland by way of Washington, D.C., with an overnight," Torok says.

After a two-day trip, Torok was in Dushanbe to meet colleagues at the Central Asian Institute for Foot and Mouth Disease, where he is the volunteer principal investigator in an ongoing ISTC project to map the spread of anthrax among animals and humans.

"There are ten to a hundred times more outbreaks of anthrax in Tajikistan than elsewhere in the former Soviet Union, and a suspicion that some of these were caused by genetically engineered *Bacillus anthracis*," says Torok. "Some of the Tadjik scientists had wanted to fight fire with fire and use a genetically engineered vaccine, but I cautioned that if some *B. anthracis* was genetically engineered, we needed evidence of that and specific knowledge of its genetic character first."

Torok had previously invited two visiting scientists to work with him in his laboratory at Berkeley Lab, and together they'd concluded that the crucial first step in knowing what they were up against was to map the strains, "to see where they came from and where they've spread." The country is more than 90-percent mountainous, with a struggling public health system, little understanding of prophylaxis, and people so poor they feel they can't afford to vaccinate their animals, or even to dispose of the infected ones. "If an animal is sick, they eat it," Torok says. "They figure if they just cut off the head, the meat will be okay."

In cooperation with the World Health Organization's World Anthrax Center at Louisiana State University and the University of Hohenheim in Germany, Torok consulted with graduate student Manuchehr Rahmatdzhonov at the Institute in Tajikistan, helping him design a research program that would include extracting and sequencing DNA from specified strains of *Bacillus anthracis*.

Are the virulent *B. anthracis* strains actually the products of genetic engineering? While there's no clear answer yet, Torok suggests that the very design of the Institute hints at former involvement in biological warfare. "The laboratories are buildings inside buildings, equipped with powerful ventilation systems, and they can be sealed off from the outside world by huge steel doors." In Soviet times the Institute had a staff of 1200; now it employs 88 people, most of them only half or quarter time.

Elsewhere the situation was even worse. The Anti-Plague Station, once part of a vital network to protect against the spread of plague in the USSR, is "a shadow of its former self," partly occupied by squatters, with its collection of plague bacteria (*Yersinia pestis*) having been destroyed during the country's savage 1992-1997 civil war. Other Tajik institutions are better organized but desperately short of crucial modern equipment.

To Russia's Farthest East

After a day of talks in Moscow, Torok flew on to the Kamchatka Peninsula to revisit a region where he has prospected for many years. During his week-long stay he collected many new microbial samples of particular interest to DOE's biofuels initiatives, such as extremophiles capable of biodegrading the woody parts of plants, and others from sites producing large amounts of hydrogen and methane. But the trip was bittersweet, for more than one reason.

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“This was most likely the last sampling expedition I will lead to the Russian Far East,” he says, because funding for the Kamchatka project will soon end. Of more immediate concern was the devastation wrought by an immense landslide in the Valley of the Geysers on June 3.

An entire mountainside in the center of the valley gave way, releasing some 12 to 15 million cubic meters of mud and boulders that dammed the river, smothered and drowned a number of geysers and fumaroles, erased helicopter landing pads, and rolled huge rocks to within inches of the cabin used by visiting researchers. Yet by early August Torok could already see signs that some, if not all, the geysers that had given the valley its name would return as the floodwaters receded, and that elsewhere new geysers would erupt.



Gennady Karpov (left center), of the Russian Academy of Sciences' Institute of Volcanology and Seismology, shows Torok the damage done by the enormous landslide that buried many unique features in the Valley of Geysers. A veil of microorganisms in a hot spring opens on a view of Kronotsky State Park's Udon Crater. Inset shows the Institute of Volcanology's field station, built with the assistance of the Department of Energy's Global Initiatives for Proliferation Prevention.

(All photos by Tamas Torok except that of Karpov and Torok by H. Smelov)

Despite the devastation, another rich bioprospecting site not far away, the vast Uzon Caldera, was unaffected. There Torok did most of his sample gathering on this trip. While in the Uzon Caldera Torok met with colleagues from several centers and institutes with whom he has worked to build onsite research facilities, and to form a consortium that will continue the exploration of the area's unique resources.

Says Torok, “I am most certain that the highly professional and collegial atmosphere will not only lead to the successful completion of the current project, but will also result in future joint proposals and projects. The additional contacts were a welcome plus. The bioprospecting expedition was highly productive. The new field facility will serve the international scientific community for decades to come.”

Additional information

More on Tamas Torok's research programs is at http://www.lbl.gov/lifesciences/labs/torok_lab.html

More about Georgia, Armenia, Azerbaijan, Uzbekistan, and Tajikistan (from Wikipedia) is at http://en.wikipedia.org/wiki/Georgia_%28country%29; <http://en.wikipedia.org/wiki/Armenia>; <http://en.wikipedia.org/wiki/Azerbaijan>; <http://en.wikipedia.org/wiki/Uzbekistan>; and <http://en.wikipedia.org/wiki/Tajikistan>

More about the Aral Sea and Vozrozhdeniye Island is at <http://enrin.grida.no/aral/aralsea/english/arsea/arsea.htm> and <http://www.globalsecurity.org/wmd/world/russia/vozrozhdenly.htm>

More about Torok's previous trips to Kamchatka, Kosovo, and Siberia is at <http://www.lbl.gov/Science-Articles/Archive/LSD-return-to-Kamchatka.html>; <http://www.lbl.gov/Publications/Currents/Archive/Oct-18-2002.html#Kosovo>; and <http://www.lbl.gov/Publications/Currents/Archive/Nov-20-1998.html#RTFToC1>

More about the June 3, 2007 Valley of Geysers landslide is at <http://www.nature.com/news/2007/070604/full/news070604-13.html>