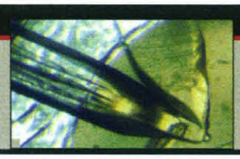


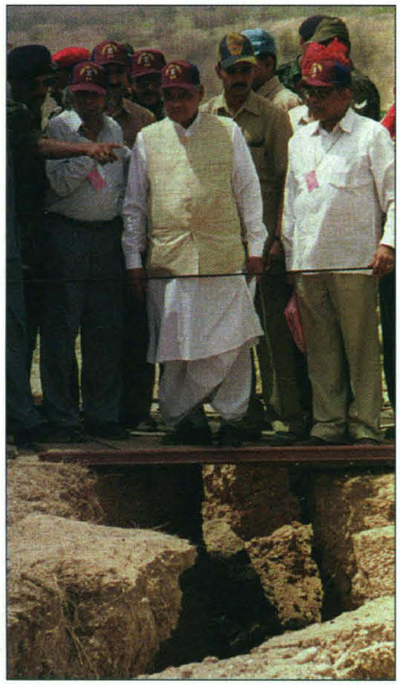
PAGE 497
Dim view of space crystal research

498
A new strategy for French research

INTERNATIONAL COLLABORATION

Indian Scientists Shaken by Bomb Test Aftershocks

NEW DELHI, INDIA—Indian and Pakistani scientists are beginning to pay a price for last May's atom bomb tests—a price that many believe is unfairly penalizing civilian science. Although the U.S. government took steps last week to limit the impact of economic sanctions, individual agencies began suspending all interactions with scientists from a long list of Indian and Pakistani research institutions, denying entry to some and questioning the status of others already in the country, restricting the exchange of lab materials, and canceling ongoing projects. Japan and Sweden have also taken punitive steps, leaving scientists from the subcontinent worrying that the stigma of the bomb tests may affect them for months or years to come.



No admittance. U.S. travel was denied to Indian crystallographer and weapons expert Rajagopala Chidambaram (far right, at Pokharan test site).

AP PHOTO/AJIT KUMAR

About a half-dozen Indian scientists say they have been prevented in recent weeks from participating in international events in the United States. The list includes Rajagopala Chidambaram, chair of India's Atomic Energy Commission and vice president of the International Union of Crystallographers, who was scheduled to speak at last week's meeting of the union in Arlington, Virginia. Other Indian and Pakistani scientists in U.S. research programs, meanwhile, have come "under review," according to a spokesperson for the Department of Energy, and one Indian scientist has already been dropped from a collaboration at DOE's Argonne National Laboratory near Chicago, Illinois. Some exchanges of laboratory materials appear to have been affected. Even the Centers for Disease Control and Prevention (CDC) in Atlanta has cited the bomb test sanctions in refusing to send out a rabies virus clone.

Under laws designed to prevent the spread of nuclear weapons and missile technology, the United States Commerce Department is blocking the shipment of goods to India and Pakistan that could have a military use. Meanwhile, a multiagency working group at the State Department is deciding on a case-by-case basis whether joint scientific and technical projects should go forward. That has left the bureaucracy struggling to formulate general guidelines, and many agencies seem uncertain about the new rules.

U.S. diplomatic officials, for example, offered competing accounts of why Chidambaram, who is involved in India's atom bomb program, was unable to get a visa to attend the crystallographers' meeting. A State Department official in Washington, D.C., says that Chidambaram simply withdrew his request for a visa. But a U.S. embassy spokesperson here says, "Visa procedures [are] under review as a result of the nuclear tests." Chidambaram says that U.S. officials in New Delhi held his application for 10 days, then returned his passport and fees on 9 July.

Caught in the net. CDC refused to share rabies virus sample with Indian researchers.

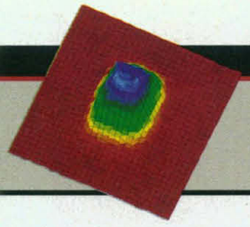
F.A. MURPHY/CDC

Less prominent Indian researchers have also been affected by the chill. A young civilian astronomer who asked to remain anonymous says he was prevented from taking a job in a private remote-sensing center in Virginia because his visa has been withheld for 2 months. "I had sold all my household belongings and was ready to fly out within a week," he says. He wonders why civilian scientists should be "made to suffer just because a nation has to be punished."

Crystallographer Krishan Lal of the National Physical Laboratory in New Delhi says he was denied a visa to visit the United States, even though he works at a civilian research facility. This violates the principles of the International Council of Scientific Unions, says Lal. The union's U.S. representative, the National Academy of Sciences, apparently agrees: Earlier this month, it sent a letter of appeal on Chidambaram's behalf to the U.S. embassy in New Delhi.

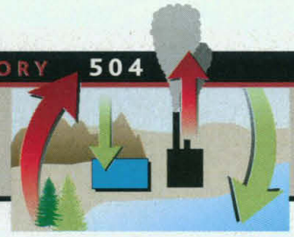
At least one U.S. scientific agency—DOE, which runs the U.S. nuclear weapons program—is also taking a hard line. Recently departed DOE Secretary Federico Peña issued a memorandum on 16 June suspending all DOE laboratory collaborations and visits involving "Indian and Pakistani foreign nationals from nuclear institutes and related entities." DOE has drawn up a list of more than 65 Indian and Pakistani research institutions covered by the suspension, including all of India's civilian nuclear centers and the Indian Space Research Organization. The rules can be waived for individual projects and scientists only by appeal to the secretary. Meanwhile, DOE spokesperson Carmen MacDougall says the agency is reviewing "half a dozen" foreign scientists now at DOE labs to see whether they should return home.

Equally alarming to some Indian scientists are the problems they now face in acquiring supplies and equipment. Scientists at the Indian Institute of Science in Bangalore are still waiting for CDC to ship rabies complementary DNA clones it promised more than 3 months ago. "We were requested to interrupt all biologic shipments until diplomatic resolutions occur," a CDC official wrote curtly to Indian officials last month.



501
A super atom clock

LEAD STORY 504
Ins and outs of carbon dioxide



507
"... probably the strongest risk factor for whether you get Alzheimer's late in life"

Sweden has gone even further. It has ended a $500,000 collaboration with India started last December in the fields of environment, energy, and food processing. And Mohan Gopal Kulkarni, a polymer chemist at the National Chemical Laboratory in Pune, says his collaborative project on biodegradable polymers with Y. Tokiwa of the National Institute of Biosciences & Human Technology in Tsukuba, Japan, has been delayed indefinitely "because of the recent nuclear tests."

Despite these setbacks, most Indian officials seem confident that their country can ride out the storm. Raghunath Anant Mashelkar, secretary of the department of scientific and industrial research, feels that "the new round of sanctions can easily be brushed aside, as India has literally grown up in this atmosphere of technology denials." But some are not so sure. They warn that Indian science will suffer as a result of this international isolation, and that the repercussions from the nuclear blast will be felt even if official sanctions are lifted soon. One engineer worries that civilian research will pay a heavy price for what he calls "the romantic indulgences of a few nuclear scientists." —PALLAVA BAGLA

With reporting by Eliot Marshall.

ANIMAL CLONING

Cloned Mice Provide Company for Dolly

Dolly, the cloned sheep, can no longer be considered a fluke. As the first—and, at the time, only—mammal cloned from an adult cell, she was greeted first with awe and, later, with doubts (*Science*, 19 December 1997, p. 2038; 30 January, pp. 635 and 647). Dolly was up against the dogma that DNA from mature cells could not start over and guide an egg's development into a complex, multicellular organism. Before that dogma could be overturned, skeptics argued, the cloning experiment that yielded the lamb needed to be replicated. Now, they've gotten their wish.

In this week's issue of *Nature*, Ryuzo Yanagimachi and his team at the John A. Burns School of Medicine at the University of Hawaii, Honolulu, provide the first scientific report confirming that cloning from adult cells is not only possible but repeatable. In it, they describe experiments that have so far yielded more than 50 cloned mice. Two other reports in the same issue describe DNA analyses proving that Dolly and the ewe she was cloned from are indeed genetically identical, as would be expected of clones. And in Japan, two calf clones born 5 July but not yet fully described in the scientific literature (*Science*, 10 July, p. 151) have apparently passed similar tests. "[Cloning] is a real phenomenon," comments Richard Schultz, a developmental biologist at the University of Pennsylvania, Philadelphia.

These achievements may reignite the ethical frenzy that followed the first reports of Dolly, primarily because of worries that the technology will be applied to humans. And they will certainly spur renewed vigor among companies vying to apply these technologies. For example, they might be used to clone herds of cattle that produce therapeutic proteins in their milk. "We intend to commercialize [the mouse technology] on a broad range of animals," says Laith Reynolds, CEO of ProBio America, a Honolulu-based company that is now supporting the mouse work in Hawaii.

To clone mice, Yanagimachi, working with Teruhiko Wakayama of the University of Tokyo in Japan, devised a variation of the technique used by Ian Wilmut of the Roslin Institute in Scotland and Keith Campbell of PPL Therapeutics to create Dolly. The idea is to get nuclei from adult cells into eggs whose own nuclei have been removed. The resulting cells can then be triggered to develop into embryos, which can be implanted in foster mothers. But while the Roslin team got the adult cells to fuse with enucleated eggs by subjecting them to an electrical pulse, Wakayama uses a very fine needle to take up the donor cell nucleus, which he very gently and quickly injects into an enucleated egg. "He is very careful to make sure as much of the donor cytoplasm is gone as possible," says Schultz. That cytoplasm could contain factors that might thwart proper development.

The Hawaii group also took a different approach to initiating egg development. In the Roslin team's case, the same electrical pulse that fused the cells prompted the egg's activation. Wakayama first lets the cells sit for up to 6 hours to give the egg cell time to alter the donated DNA so that its developmental genes can be expressed again. Then, the Honolulu team triggers development of the eggs by putting them into a culture medium containing strontium, which stimulates the release of calcium from the eggs' internal stores—the same signal that tells fertilized eggs it is time to start dividing.

For some reason, the Honolulu team's strategy worked best with cumulus cells, which surround an egg as it matures. Over the past year, the group has used them to create some 50 clones, confirming their clonal origins by comparing the DNA of the newborn mice to that of the animals that provided the nuclei. The cloned mice seem normal: The group has cloned some clones and mated others, creating healthy young in both cases. All told, "it's a very compelling paper," says Michael McClure, a cell biologist at the National Institute of Child Health and Human Development in Bethesda, Maryland.

PROBIO AMERICA INC.

Clones' clan. Two cloned mice stand by their white surrogate mother (lower tier), with the egg and nucleus donors above (top right and left, respectively).

Just as compelling are the results of two DNA analyses, conducted independently by the PPL-Roslin team and a group from the Hannah Research Institute in Aye, Scotland, and the University of Leicester in the United Kingdom, to evaluate Dolly's origins. Some researchers thought that the limited DNA analysis Wilmot's team originally performed to show that Dolly is not an offspring of its surrogate mother was not convincing. So, the two groups made a more detailed comparison of DNAs from Dolly, from the cultured udder cells used as nuclear donors, and from the ewe that provided the cells.

Wilmut's team, working with a local company called Rosgen, analyzed 10 microsatellites—short stretches of DNA known to vary between unrelated individuals. "They all had identical patterns," comments Robert Wall, a geneticist at the U.S. Department of Agriculture in Beltsville, Maryland. The second team compared DNA fingerprints—patterns