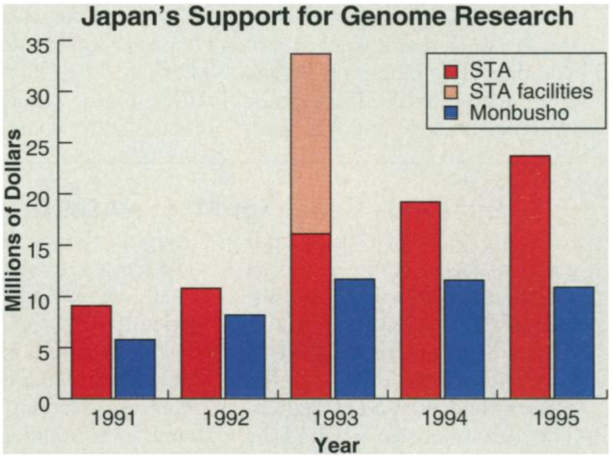
Is Japan a Step Behind on Genome?

Grants to four teams will allow Japanese researchers to begin sequencing selected regions of the human genome, but the government has delayed large-scale efforts to match those in other countries

TOKYO—Like human genome researchers throughout the world, scientists in Japan are convinced the time has come for large-scale sequencing. But many here are worried that the time may pass before they get their effort into high gear. This month the Japanese government is set to announce funding that will allow four research groups with links to overseas teams to start sequencing efforts (see box). But the size of these teams is small, and their work will be confined to limited stretches of selected chromosomes. In addition, *Science* has learned that a proposal to launch a larger scale effort on a par with those being contemplated in the West (*Science*, 2 June, p. 1270) is likely to be scaled back to insignificance. Together, say researchers, those decisions could leave Japan with an embarrassingly small contribution to the worldwide sequencing effort and hamper related research throughout the life sciences and medicine.

While genome scientists are disappointed by the fate of the larger proposal, they are pleased to be getting at least some sequencing under way. The projects are being funded by the Japan Information Center of Science and Technology, an arm of the Science and Technology Agency (STA), and jointly could receive up to $10 million a year for the next 3 years. (STA shares responsibility for human genome efforts with Monbusho, the Ministry of Education, Science, Sports, and Culture). Still, scientists worry that the amount is too small for Japan to keep pace with overseas efforts. "The scale [of the groups] just can't be compared to the groups in the U.S. or England," says Yusuke Nakamura, director of the Human Genome Center at the University of Tokyo's Institute of Medical Science and one of the grant winners.

Ironically, those foreign scientists seem more sanguine about Japan's prospects than do their colleagues at home. In the United States, for example, the National Institutes of Health (NIH) is still reviewing applications for funding new sequencing groups, and the Department of Energy is considering new directions for its own sequencing efforts. Groups in the United Kingdom are also awaiting decisions later this year on funding. To some scientists overseas, the 1 October date for expanding the four Japanese projects looks pretty good. "I'd certainly like to have our [proposal to NIH] funded now," says David Patterson, president and senior scientist at the private Eleanor Roosevelt Institute in Denver.



SOURCE: STA/MONBUSHO

Going up. The two major Japanese agencies supporting human genome research are putting more money into the program.

FOUR TEAMS GET SEQUENCING GRANTS

Group Leader/Affiliation	Lab Site for Grant	Target Area	Suspected Function
Hidetoshi Inoko, School of Medicine, Tokai University	School of Medicine, Tokai University	The HLA region on chromosome 6	Immune response
Yusuke Nakamura, Human Genome Center, University of Tokyo	Cancer Institute of the Japanese Foundation for Cancer Res.	Band 21.3 on chromosome 3	Tumor suppressor genes
Yoshiyuki Sakaki, Human Genome Center, University of Tokyo	Kitazato University	Down syndrome region of chromosome 21	Down syndrome
Nobuyoshi Shimizu, Keio University School of Medicine	Keio University School of Medicine	Down syndrome region of chromosome 21 and immunoglobulin-λ gene cluster and cat's eye syndrome regions on chromosome 22	Down syndrome, immune response

SOURCE: STA

While Japan's timing may be right, the scale of the effort may not be. Nakamura notes that the four teams will typically have no more than a dozen scientists and technicians. Although there are similarly sized groups overseas—Patterson expects his sequencing team to number 15 or so at full strength—most efforts will involve much larger teams churning out sequencing data as fast as they can. Bruce Roe, who heads a sequencing group at the University of Oklahoma, says he currently has 35 people at work on regions of chromosome 22. John Sulston, director of the Sanger Center in Cambridge, U.K., hopes for enough funding for three 50-person sequencing teams.

Japan has nothing on that scale planned, and the STA money won't go as far as it would overseas. "Sequencing machines cost twice as much, supplies cost twice as much, and labor costs are very high," Nakamura says. The funding is based on an estimated cost of about $2 per base. In contrast, Roe puts his group's per-base cost at 50 cents to $1, depending on the level of accuracy achieved.

Japan's sequencers are confident of making important contributions in the areas they are targeting. But, as Nobuyoshi Shimizu, a molecular biologist at Keio University who is leading another sequencing group, puts it: "If, in a few years, someone tallies up how many bases have been sequenced and finds Japan has only done a few percent, it's not going to look good." Nakamura estimates the total output of Japan's four sequencing groups is likely to be in the range of two to three megabases per year. "Japan should be doing at least 10 to 20 megabases per year," he says. Yoshiyuki Sakaki, a molecular biologist at Tokyo's Human Genome Center who next April will become principal investigator of Monbusho's Human Genome Program, sums up the message: "We should have a large-scale, high-performance sequencing facility in this country."

Hassle over hiring

Sakaki would like to set up just such a facility and has asked Monbusho for permanent funding for four new research groups to focus on sequencing, sequencing technology, informatics to support sequencing, and functional analysis. Budget negotiations within Monbusho and between Monbusho and the Ministry of Finance are at "a delicate stage," says Koichiro Ohsaka, director of Monbusho's Grant-in-Aid Planning Office, who declined to predict the outcome.

Teamwork Takes Practice

TOKYO—With 3 billion base pairs, the human genome would seem to offer something for everyone. But in reality, researchers in Japan and around the world face a stiff challenge in trying to avoid costly duplication in carrying out their work.

For Nobuyoshi Shimizu, a molecular biologist at the Keio University School of Medicine who is targeting a stretch of chromosome 22, being a team player means fitting in with the efforts of larger British and U.S. groups. Bruce Roe, who heads a team at the University of Oklahoma, says his group is handling the upper half of the chromosome, with the lower half split between a group at Washington University in St. Louis headed by Robert Waterston and a group at the Sanger Center in Cambridge, U.K. But both groups welcome help from other labs. "There's enough for all of us to do," says Roe. Shimizu's group has just completed what is believed to be the world's largest complete human contig, a map depicting the relative order of a linked library of small overlapping clones. It covers a 1.2-megabase stretch containing the immunoglobulin-λ gene cluster. Because the genomic segment lies within the larger area Roe plans to sequence, he and Shimizu have agreed to start at opposite ends and meet in the middle. Roe is making similar arrangements with other U.S. and European groups.

Scientists working on other chromosomes have not yet drawn up a game plan. Yoshiyuki Sakaki, a molecular biologist at the University of Tokyo's Human Genome Center, and David Patterson, president of the Eleanor Roosevelt Institute in Denver, are informally coordinating the work of six or seven groups on different parts of chromosome 21, with Sakaki planning to sequence part of the Down syndrome region. The chromosome 21 community has a history of close collaboration, Sakaki notes, adding that researchers will plot their strategy during next month's meeting in Minneapolis of the American Society of Human Genetics.

Shimizu is also interested in the Down syndrome region. He and colleagues at Keio and at the Geneva University Medical School recently identified a gene in the critical region that could be involved in some of the features that characterize the disability. His sequencing group intends to tackle a 1-megabase region containing that gene. Coordination of different groups is also just getting started on chromosome 6, the focus of sequencing efforts by Hidetoshi Inoko, professor of molecular life science at Tokai University's School of Medicine. Stephan Beck, head of DNA research at the Imperial Cancer Research Fund in London, says a survey of the community indicates a lot of overlap in regions targeted for sequencing, particularly in the human leukocyte antigens region, which he says is "the best studied region of the human genome" because of its role in the immune response. Beck, who is about to move to the Sanger Center to head that institute's chromosome 6 sequencing effort, says it is up to the individual groups to "decide whether to cooperate or compete," but that he and Inoko will try their best to avoid duplicating the efforts of others.

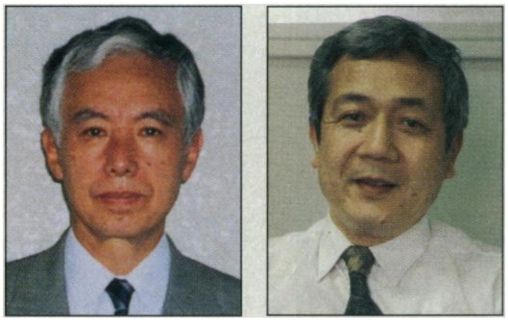
The lone wolf among the Japanese sequencers is Yusuke Nakamura, a molecular biologist and head of the University of Tokyo's Human Genome Center. He believes that he has picked a region of band 21.3 of chromosome 3, the site of suspected tumor suppressor genes, that no one else is working on. A former surgeon, Nakamura is one of the discoverers of a gene on chromosome 5q21 associated with colon cancer (*Science*, 9 August 1991, p. 665).

–D.N.

But one thing is already clear. Whatever budget is finally approved, it won't include funding for technicians. Sakaki says he proposed a special scheme to get around Monbusho's rules against hiring technicians at national universities, but Ohsaka says it's not acceptable. "There is a ceiling on the number of [national university] employees," he says. "It is very difficult to exceed this number and increase staff sizes." As a result, any sequencing group will conform to the existing model, with one professor, one associate professor, and one or two assistant professors. In Nakamura's view, that limitation guts the whole proposal. "Such a group could only do 40 to 50 kilobases a year," he says. "This is not large-scale DNA sequencing."

The STA program is an attempt to circumvent Monbusho's staffing restrictions. Makoto Furunishi, deputy director of STA's life science division, says the agency recognized the need and saw itself as a partial solution. "Monbusho and the Ministry of Health and Welfare aren't set up to support this type of program," he says. "STA has more flexibility." Technicians can be hired under STA grants, and grant sizes can be set to suit a project's needs.

One thing STA grants can't overcome, however, is bureaucratic boundaries. Restrictions on the use of national university facilities will prevent Sakaki and Nakamura from setting up their STA-funded sequencing groups at the University of Tokyo. Instead, Sakaki's group will be based at the private Kitazato University, where he is an associate professor, and Nakamura's group will be housed at the Cancer Institute of the private Japanese Foundation for Cancer Research, where Nakamura had once worked. The groups based at Keio and Tokai universities are not affected because they are located at private institutions.



Opposing views. Kenichi Matsubara *(left)* believes the government is on track, while Yoshiyuki Sakaki says large-scale sequencing should begin at once.

But even at STA, Furunishi says it is difficult to increase spending rapidly for a new program. The sequencing grants, initially lasting 6 months, will probably be extended if the groups are making progress and if other groups are brought in. There has even been talk about setting up a new institute, perhaps modeled on the Sanger Center, that would focus on mapping and sequencing the genomes of model organisms as well as the human genome. But Furunishi advises caution. "In several years we'll look at [genome sequencing] progress and decide," he says.

With overseas groups proposing to complete the sequencing of the human genome by 2001, many Japanese scientists fear such a delay could be fatal. But others are more optimistic. Kenichi Matsubara, a professor at Osaka University and currently the lead researcher for Monbusho's Human Genome Program, says that although Japan's budget process makes it hard to shift resources quickly, it allows established programs to grow rapidly. As proof, he points to spending levels, which have grown from $15 million in 1991 from the two agencies to $35 million this year, not including salaries. Overall, says Matsubara, "the current level of funding and activity [in Japan] is not something to be disappointed about."

Matsubara's view gets some support from Oklahoma's Roe, who says it is a misconception that completing the sequencing of the human genome will mean the end of sequencing. "If we get the whole thing done by some massive effort, what we'll have is the *Encyclopaedia Britannica* with a lot of spelling mistakes," Roe says. Sequencing teams will still be needed to correct those errors, he adds, a task that will stretch "to perpetuity."

Sanger's Sulston agrees that "2 to 3 years is not too late to start" a sequencing effort. In addition to the human genome, researchers would also like to sequence the mouse genome because of its importance to medical and biological research. But Sulston agrees with Shimizu that further delays "would not look good."

The big picture

Appearances are important to Japanese scientists, who still feel the sting of a 1989 charge of scientific freeloading by Nobel laureate James Watson, then head of NIH's genome program. In a letter to Matsubara, Watson indirectly threatened to try to block Japanese access to U.S. genome databases if Japan didn't increase its contributions to the worldwide effort, including the fledgling Human Genome Organization.

Scientists here also worry that a failure to do their share of sequencing will hinder progress on other projects. "I'm very concerned about what [a low level of genome funding] will mean for life sciences and medical research in general," says Sakaki. He sees a large-scale sequencing effort as a necessary first step in analyzing the function of particular genes and, eventually, developing new therapies. Without that initial commitment of resources, he says, the government may not be willing to fund later research and development activities.

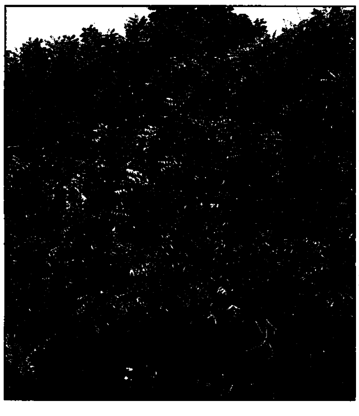
Matsubara is optimistic that decision-makers at Monbusho and STA will find a way to increase support in the future. "They know the importance [of the human genome program]," he says. But Nakamura disagrees, arguing that "the Human Genome Project is seen as a small part of molecular biology or medical research. If the budget stays the same, we have little hope to contribute very significantly to the project."

–Dennis Normile

AGRIBUSINESS

Patents on Native Technology Challenged

The neem tree of India, known as the "blessed tree" and the "curer of all ailments," is at the center of a patent battle that pits advocates of indigenous agriculture against a major U.S. company. Valued in India for its use as a medicine, a spermicide, and as fuel and timber, the neem tree caught the attention of W. R. Grace & Co. of New York because its seeds contain a potent natural pesticide, azadirachtin. Grace patented a method of extracting azadirachtin and stabilizing it. But now the company faces a legal challenge from Jeremy Rifkin, a longtime foe of the patenting of genes and animals. Rifkin claims that Indian farmers have used neem tree seeds as a pesticide for centuries, making this application obvious and unpatentable.



AGRICULTURAL RESEARCH SERVICE/USDA

Pesticide factory. Seeds of India's neem tree yield patented product.

Rifkin's nonprofit group, the Foundation on Economic Trends, has recruited 200 other organizations from 35 countries to join a campaign against the patent, issued in June 1992. Rifkin was scheduled to file a formal petition with the U.S. Patent and Trademark Office (PTO) on 14 September, calling for a revocation of the patent. Rifkin, a consummate coalition-builder (*Science*, 26 May, p. 1126), says the petition is the opening shot in what he hopes will be a widening battle over intellectual ownership of "native technologies." Rifkin argues that "the biological resources that have been discovered by natives ought to be maintained in the open."

While Grace's patent may be under challenge, azadirachtin's effectiveness as a natural pesticide is not. In 1985, U.S. timber importer Robert Larson won approval from the Environmental Protection Agency (EPA) to use the chemical as a nonagricultural biopesticide, and in 1988 he sold the product rights to Grace, which has been marketing it since 1992 as Margosan-O (*Science*, 28 February 1992, p. 1070). In March 1994, the EPA registered Grace's NEEMIX as the first neem product cleared in the United States for use on food crops. Grace entered into a partnership with the Indian company PJ Margo Private Limited in 1993, becoming the minority owner of a factory in Karnataka, India, where seed extract is processed and stabilized for long-term storage. In the past, Indian villagers have not been concerned about storage because they soak the seeds overnight in water or alcohol and place the emulsions on crops the next day.

The Rifkin petition, brought by a coalition of scientific, business, trade, farm, environmental, and cultural organizations, is expected to trigger an automatic re-examination of the patent by the PTO. The agency must respond 1 month after a petition is filed, and petitioners may appeal the decision in federal court.

"We believe we have an airtight case," Rifkin says. The law says an invention may be patented only if it is different from the "prior art" or knowledge a person in the field would have. Grace argues that its patent claim is novel because it applies to "neither the extraction nor the processing of the extract, but to a unique formulation" of azadirachtin developed "to insure its shelf stability." However, according to Rifkin, the petitioners will present "several hundred" journal articles as evidence that Indian scientists had described the extraction method before the patent was issued. And Rifkin says Indian companies were already using the chemical in stable solutions. "The Patent Office was absolutely wrong in granting this patent," he says.

Rifkin also claims the patent will hurt Indian farmers because they may have to stop using their own technology or pay steep royalties. Grace disagrees: "No individual or company is prohibited from the historical or traditional uses of neem extracts," a company statement says. The statement maintains that Grace cannot gain exclusive use of the neem tree extract, because 22 different companies, including three Indian companies, own 40 different patents on neem-related procedures.

Some independent experts in patent law also question the soundness of Rifkin's legal arguments and the significance of the patent for the Indian farmer. John Barton, a Stanford law school professor, says, "It may actually help some Indians by creating an industry for neem seeds." He notes that because the patent covers only one particular method of extraction, it shouldn't directly affect the Indians already using neem seeds.

Rifkin remains confident of his case, however—and of its value as a rallying point for those who oppose patents based on the genetic resources of developing countries.

–Lori Wolfgang