MEETING MICROBIAL GENOMES III

DNA Sequences Provide Grist for Microbiologists

CHANTILLY, VIRGINIA—At the Microbial Genomes III meeting held here from 29 January to 1 February, biologists shifted their focus from sequencing technology to the exciting new biological insights microbial sequences are providing.

Bacterial Partners for Filaria

When Barton Slatko signed on with the Filarial Genome Project, he expected to be determining the genetic makeup of a major human parasite. Filaria worms infect some 120 million people worldwide, causing the tropical diseases African river blindness and elephantiasis. But the work of Slatko, a molecular biologist at New England BioLabs Inc., in Beverly, Massachusetts, took an unexpected turn after the project's team found that some filaria worms are infected by an odd bacterium called *Wolbachia*.

The bacterium is best known for infecting insects, where they are transmitted from generation to generation through the germ cells. Depending on the species, it can make the host infertile or alter the sex ratio of the offspring, such that only one sex survives. In contrast, in filaria the *Wolbachia* seem key to fertility, thereby making the worms susceptible to antibiotics that attack the bacteria. The bacteria "seem to have a pretty interesting role in terms of therapy," Slatko reported. So he and his colleagues have decided to sequence a *Wolbachia* genome, hoping it will lead to new ways to control filarial infections.

Because of the importance of these worms as both human and animal parasites—they also cause widespread infection in livestock—the World Health Organization set up a consortium in 1994 to map the genome of the filarial worm *Brugia malayi* and look for genes that might provide targets for new antifilarial drugs and vaccines. But as consortium members began analyzing the worm's genetic material, "we kept finding bacterial sequences," says David Guiliano, a collaborator from the University of Edinburgh, Scotland. After confirming that the microbial DNA was not contamination, the researchers found earlier reports suggesting that the nematodes might harbor some unknown kind of bacteria.

Starting 25 years ago, a few researchers had noticed "dense bodies"—possibly bacteria—in the embryonic and adult tissues of several species. One report even suggested that antibiotics seem to have antiworm effects. But until recently, "that work was not followed up on," says Guiliano.

Then in 1995, Claudio Bandi from the University of Milan, Italy, and his team reported that they had detected ribosomal DNA sequences indicating that the worms are infected by *Wolbachia* bacteria. Last year, Bandi, Edinburgh's Mark Blaxter, and their colleagues confirmed these data: They surveyed 10 filarial worms, finding *Wolbachia* DNA in nine of them. Based on their analyses, they concluded that the bacteria had long ago parasitized the nematodes.

Weak spot. Sequencing a bacterium that infects filaria worms (*above*) may lead to ways to thwart the roughly 6-centimeter worms, which cause the swollen limbs of elephantiasis.

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Although *Wolbachia* take a toll on insect fertility, "there's no evidence that they are parasites" in nematodes, says Guiliano. Instead, the bacteria seem to contribute to the nematode's reproductive success. Evidence for that comes from Bernhard Fleischer and his colleagues at the Bernhard Nocht Institute for Tropical Medicine in Hamburg, Germany. They reported in the January *Journal of Clinical Investigation* that the antibiotic tetracycline kills the bacteria living in the reproductive tissue of nematodes found in mice. The treatment resulted in smaller, infertile nematodes, but it had no effect on another nematode species that lacked signs of bacterial infection. "If you kill the *Wolbachia*, you will [eventually] kill the [infected] nematodes" by preventing their reproduction, explains Scott O'Neill, a vector biologist at Yale University in New Haven, Connecticut.

And that's good news for filarial researchers, as it may provide a new way to try to interrupt the worm's life cycle. "They had been banging away at the nematodes without turning up much new, and then *Wolbachia* came on the scene," O'Neill points out. Now, "the worm [research] groups are all latching onto [it]." The sequence may lead to better anti-*Wolbachia* drugs, which in turn may be more effective in killing the worms.

The New England BioLabs's effort is likely to come out with the first *Wolbachia* sequences, as Slatko expects to be finished within 6 months. Filaria researchers can hardly wait, says Alan Scott, a molecular parasitologist at Johns Hopkins University in Baltimore who is part of the Filarial Genome Consortium. Anticipation of the sequencing results is creating "quite a bit of excitement."

—ELIZABETH PENNISI

Super-microbe Decoded

If any bacterium can be considered a supermicrobe, it is the one called *Deinococcus radiodurans*. Not only does this soil bacterium live on Antarctic granite, but it can survive doses of radiation a thousand times the lethal level for humans. After each assault, it simply stitches together its shattered chromosomes. At the meeting, biologists announced that they have completed sequencing the *Deinococcus* genome and uncovered one more surprise. Despite its super powers, *Deinococcus* is really a microbial Clark Kent, genetically much like other bacteria.

The 3-million-base genome, decoded by molecular biologist Owen White and his team at The Institute for Genomic Research (TIGR) in Rockville, Maryland, indicates that *Deinococcus* owes its extreme radiation resistance to the same repertoire of mechanisms for repairing DNA found in other organisms. It just has more of them than most other life-forms. The genome also suggests how the microbe evolved its radiation resistance, an ability that could help cope with humanity's own messes. Other results presented at the meeting suggest that genetic engineers may be able to equip this hardy organism with genes that could enable it to degrade toxins and clean up metals at radioactive waste sites.

The TIGR researchers started work on the *Deinococcus* genome in 1995, but the going was slow because it has a high proportion of guanine and cytosine bases, as well as many repetitive regions. Despite

Mighty microbe. The pink color of *Deinococcus radiodurans* belies its toughness.

that, "TIGR has done a good job of it," says Richard Alm, a microbiologist at Astra Research Center in Boston, Massachusetts. Now, the team's analysis is revealing the surprisingly conventional nature of the microbe's radiation defenses.

One element of these defenses is an enzyme called MutT. Radiation damages cells in part by generating reactive forms of oxygen that oxidize key cellular compounds, including some of the nucleotide building blocks of DNA. These oxidized nucleotides can cause faulty DNA replication, but MutT protects against such mutations by helping rid the cell of the oxidized nucleotides. Most organisms have a single MutT gene, but with 20 MutT-like genes, *Deinococcus* is capable of "removing a whole lot of oxidative products," White said.

The sequence also offers a clue to how *Deinococcus* repairs the numerous breaks that radiation induces in its DNA. This repair requires that the microbe put the fragments back together in the right order. In 1995, Michael Daly and Kenneth Minton, molecular microbiologists at the Uniformed Services University of the Health Sciences, Bethesda, Maryland, had suggested that the microbe, which usually carries multiple copies of each of its three chromosomes, rebuilds the copies in parallel. This would be possible, they proposed, if the chromosome copies were aligned. The nearby fragments of one copy could thus provide information missing from the other, and complete chromosomes could be assembled (*Science*, 24 November 1995, p. 1318).

But the genome seems to support a different picture, which first began to emerge when Daly did experiments with artificial DNA inserted into *Deinococcus* chromosomes. He showed that the fragments form circles before they build reconstituted chromosomes. When White and his colleagues realized that the *Deinococcus* genome contains some several hundred repeating stretches of DNA, they wondered whether these repeats were involved in making circles during repair.

One possibility is that early in repair, a repeat at the end of a fragment loops around and links to one at the opposite end, forming a circle and thereby protecting the broken ends of the DNA. Alternatively, these stretches of repetitive DNA may help keep DNA fragments from copies of the same chromosome close by, enabling them to link up more readily and eventually reconstitute a full chromosome. But, Daly cautions, "it's early days," and the roles of both repeats and the circles are still unclear.

Why the organism would have evolved this Phoenixlike ability to rebuild itself after radiation exposure has been a mystery, but its lifestyle and its genome offer some clues. Desiccation damages DNA, and an analysis of the genome by Daly, Minton, and Kira Makarova of the National Center for Biotechnology Information hints that *Deinococcus* gained some of its all-round toughness by adapting to harsh, dry environments.

The researchers showed that *Deinococcus* has genes for at least three desiccation-resistant proteins that thus far have been found only in plants. It also has other genes usually seen in eukaryotes and not in bacteria. "It seems whole families [of genes] were transferred [from other species]," Makarova suggested, although more sequenced genomes will be needed to confirm this.

While some researchers mine the *Deinococcus* sequence for clues to the microbe's capabilities, others have been looking for ways to exploit its hardiness. At the meeting, Minton described the success of Daly's group in providing *Deinococcus* with genes that allow it to detoxify a common environmental contaminant—mercury. Daly and Hassam Brim in his lab showed that neither the mercury-altering genes nor another set of newly added genes for breaking down the chemical toluene interfere with the microbe's resistance to radiation. The engineered *Deinococcus* might thus serve to detoxify sites doubly contaminated with chemicals and radioactive wastes. "If we can take mixed waste and reduce it to a [pure] radiation waste, then we can handle it," explains Marvin Frazier, a microbiologist with the U.S. Department of Energy.

White is eager to see his supermicrobe go to work. "It's able to withstand a lot of environmental insults," he emphasizes. "This is going to really be an industrially relevant organism."

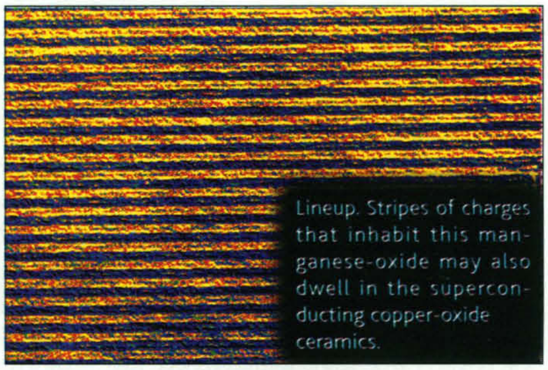
–E.P.

PHYSICS

Could Charge Stripes Be a Key to Superconductivity?

Seen in ceramic superconductors, stripes are confounding many theorists but exciting a few as a possible clue to how these materials work

Takashi Imai wears his stripes reluctantly. Like hundreds of other physicists around the world, Imai has been struggling to understand how an odd family of ceramics manages to conduct electricity without resistance at unprecedentedly high temperatures. Along with most other physicists studying high-temperature superconductivity, he was inclined to discount an idea that has hovered at the fringes of the field for several years—that charges percolate back and forth through these materials in a series of stripes, a few atoms wide, creating a landscape that helps current flow effortlessly through the material in all directions.



Lineup. Stripes of charges that inhabit this manganese-oxide may also dwell in the superconducting copper-oxide ceramics.

"I was very skeptical [of the stripe theory]," says Imai, a young assistant professor at the Massachusetts Institute of Technology. The evidence for the stripes was patchy, and the theory "looked too simple to be true." But that was before last summer, when Imai and his students began running a set of experiments for months on end that showed clear hints that charges were indeed running in defined lanes. By November, the group knew they were onto something big and resolved to double check every detail. "We kept running experiments 24 hours a day, 7 days a week. I skipped Thanksgiving and Christmas to keep taking data," he says, adding casually, "This is a competitive field."

Competitive is an understatement. Deciphering the mystery of high-temperature superconductivity has been the prime ob-

CREDITS: (TOP) TIGR; (BOTTOM) S. MORI *ET AL.*, *NATURE* 392, 473 (1998).