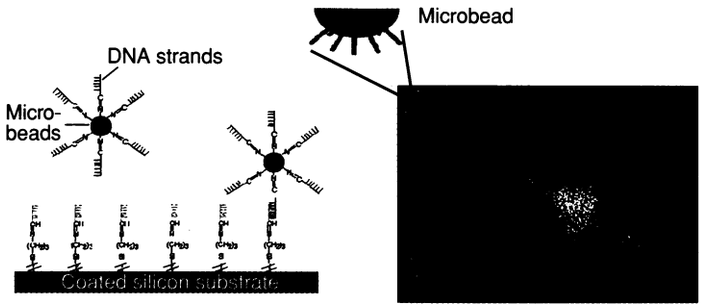
The San Diego researchers were looking for a way to help the right peg find its hole, and they settled on DNA. The chemical bases that make up DNA—cytosine, guanine, adenine, and thymine—will bind to each other only in particular pairings: C with G and A with T. Hence, a single strand made up of the bases ATTTGC will bind strongly with its complementary strand, TAAACG, and not with any other sequence. The researchers set out to exploit this selectivity by attaching short complementary strands of DNA to the pegs and substrate to help the devices find their correct positions.

In their first experiment, the team coated a substrate with a particular short strand of DNA. They then covered parts of the substrate with a mask and exposed it to ultraviolet light. The light chemically altered the DNA in exposed areas so that it could no longer bind to complementary strands. The researchers then coated some microbeads—which acted as dummy devices—with strands of DNA complementary to those on the substrate. When a fluid carrying the coated beads was splashed over the substrate, the beads successfully bound only to those areas that had not been exposed to UV light. One drawback of the technique is that it worked only for small devices, several hundred micrometers across, that would flow easily and not block other devices.

In a second experiment, designed to show that several varying kinds of "devices" could be deposited at once, the group used masks to deposit four different types of DNA strands onto a substrate and then attached complementary strands to four different fluorescent molecules. When the labeled molecules were splashed onto the substrate, the pattern of fluorescence showed that they had bound only to the appropriate regions of complementary DNA. In a real system, this would mean that four completely different types of devices could be attached to many selected sites on a chip.



Nature's glue. DNA strands bind beads and substrate together.

ESENER *ET AL.*

The researchers realize, however, that just providing the glue is not going to be enough. They are now looking for more active ways to guide the devices to their correct positions. One possibility is to add extra chemical groups to the DNA on the devices to give them an electric charge, then create electric fields on the substrate to attract the charged devices to "landing sites." The team is also investigating other techniques, such as creating currents in the fluid that would sweep the tiny devices to the right places.

An even bigger challenge will be creating an electrical connection between the devices and their host semiconductor. The team is looking at the possibility of putting the DNA glue on the top of devices and bonding them, upside down, onto a dummy substrate. Once all the devices are in position, the dummy could be flipped over and pressed down on the real substrate. The substrate might be coated with molten solder, which would add an electrical bond to the mass marriage.

–Sunny Bains

Sunny Bains is a science writer based in the San Francisco Bay area.

BIOTECHNOLOGY

Weighing DNA for Fast Genetic Diagnosis

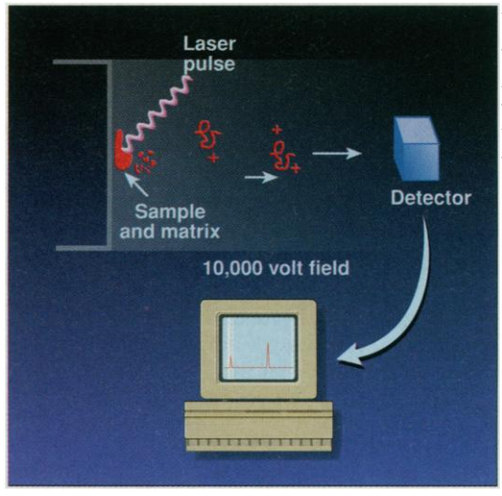
The modern doctor's little black bag, already overflowing with high-tech diagnostic devices, may soon have to make room for another advance. To diagnose a disease, judge future risks, or design a treatment, doctors will one day want to know which disease-related genes a patient carries. And they will want this diagnostic verdict to be as fast and accurate as a cholesterol or blood chemistry test today. As Charles Cantor, director of Boston University's Center for Advanced Biotechnology, puts it: "You need a detection system that can identify the gene sequences that you are looking for with high specificity, quickly, and in large volumes. The best analytical tool for doing this," he adds, "is mass spectrometry."

Borrowed from chemistry, this technology is a sharp departure from current methods, which identify a gene sequence by allowing it to bind to a matching probe, either on a gel or a chip. Instead, a mass spectrometer vaporizes the DNA and accelerates the molecules through a vacuum chamber with the help of an electric field. Tiny differences in the time it takes the DNA fragments to reach the detector reveal small differences in their mass, and hence their sequence.

The basic technique used for biomolecules is one with an unwieldy name, matrix-assisted laser desorption/ionization–time-of-flight mass spectrometry, but a harmonious acronym, MALDI-TOF. It is now a decade old, but recent improvements have made it a hot commodity among companies hoping to commercialize DNA analysis. "With today's technology, MALDI-TOF can analyze hundreds of DNA samples ... in a matter of a few minutes," says Daniel P. Little, who directs mass-spectrometry development at Sequenom Inc., a San Diego–based company hoping to be generating diagnostic products within 6 months.

The standard way to distinguish different variants of a gene is to chop the DNA into fragments, separate them on a gel, and apply probes labeled with fluorescence or radioactivity, which bind to fragments with a particular sequence and light them up. But the process is slow and the gels can be hard to interpret. Newer techniques embed an array of different DNA probes on a single chip, allowing researchers to test for many gene variants at once. These so-called DNA chips can screen DNA quickly. But, as Cantor explains, the probes sometimes bind to sequences they don't completely match, which can limit the chips' accuracy.

Mass spectrometry may combine the DNA chip's speed with exquisite accuracy. The technique has long offered chemists a fast way to sort small molecules that vaporize naturally



SOURCE: SEQUENOM INC.

All in the timing. A mass spectrometer sizes up DNA by vaporizing and ionizing it, accelerating the molecules, and recording their arrival times at a detector.

or can be coaxed into a vapor with bursts of energy from a laser or ion beam. But vaporizing large biomolecules while keeping them intact once seemed impossible. A decade ago, however, Franz Hillenkamp and colleagues at Westfälische Wilhelms University in Münster, Germany, found a way to do so with proteins: Cocrystallize them with certain small molecules, collectively called matrices. When a nanosecond laser pulse vaporizes the matrix, the resulting puff of material gently lifts the ionized biomolecule as well.

DNA was a tougher problem. But in 1993, Christopher Becker, then at SRI International in Palo Alto, California, and now at GeneTrace Systems in Menlo Park, California, found a simple matrix compound, 3-hydroxypicolinic acid, that worked with DNA sequences 20 to 25 bases long. By trial and error, MALDI practitioners have come up with several new matrices that work with DNA fragments as long as 100 bases.

The latest MALDI-TOF machines allow the cloud of matrix molecules to dissipate before applying an electric field. The field accelerates the charged DNA fragments toward a detector, and the differences in time of flight can reveal mass differences as small as 0.03%. If the DNA sequences from a gene have the same length—as they do if they have been produced by the polymerase chain reaction—any departure from the mass of the normal sequence reflects a mutation that has deleted or added bases or substituted others that have a different mass. "The results are an absolute indicator of the presence or absence of specific DNA sequences," says Sequenom's Little. MALDI-TOF can distinguish gene variants that differ by as little as a single base pair, and it can also analyze microsatellites—stretches of two-, three-, or four-nucleotide repeats often used as markers for locating disease-causing genes.

Besides offering unmatched precision, MALDI-TOF is inherently fast. The DNA forms a vapor and flies to the detector in fractions of a second; even repeating the process several times with the same sample to boost the sensitivity takes as little as 2 seconds. By preparing the samples in a grid and having the laser scan each spot in turn, a MALDI-TOF instrument can analyze 100 samples or more in a matter of minutes.

The combination of speed and accuracy could give the technique a role in genome sequencing as well as diagnosis. Standard, Sanger-type DNA sequencing generates many partial copies of a DNA sequence, each one starting at one end of the sequence and ending with a different one of the constituent bases. To determine the original sequence, biologists need to know the final base on each partial copy, together with the copy's length. Doing so now requires reading hundreds of bands on gels. But by sending the mixture through a mass spectrometer, biologists could quickly read off the fragments' lengths and—from the mass differences between successive fragments—the final base on each one. Investigators at both GeneTrace and Sequenom have published sequences determined with MALDI-TOF, the latest one, from Sequenom, appearing in the April *Nature Biotechnology*.

For practical gene sequencing, however, MALDI-TOF would have to work with DNA fragments much longer than the current 100-base capacity. Becker reportedly has discovered a new proprietary matrix that he expects will extend MALDI-TOF's reach to 1000-base sequences. "If you can really do upward of 1000 bases using this technique, and if it is indeed faster and cheaper, then this would be a big breakthrough for high-throughput sequencing," says Jeffrey Polish, who works in Mark Johnston's sequencing laboratory at Washington University in St. Louis.

In the meantime, the technology has no shortage of applications. Sequenom has shown, for example, that it can discriminate among 30 of the mutations that cause cystic fibrosis and pick up polymorphisms in the apolipoprotein E gene, which have been linked to familial hyperlipidemias, heart disease, and Alzheimer's disease. GeneTrace has developed a mass spectrometry–based system that can analyze which genes are being expressed in cells by identifying expressed sequence tags, short stretches of DNA copied from the messenger RNA made by active genes. Knowing which genes are active in a tissue can help pharmaceutical companies determine which ones are good drug targets.

With MALDI-TOF instruments running about $125,000 each—less than a standard clinical chemistry analyzer—these systems may also end up in large diagnostic labs. "Diagnostics at the level of the gene is something that we know is valuable, but is difficult, slow, and expensive today," says David Cooper, chief scientific officer at Nichols Institute Reference Laboratories, a division of Quest Diagnostics, one of the big 3 national reference laboratories. MALDI-TOF, he says, could be just the right medicine.

–Joseph Alper

Joseph Alper is a free-lance writer in Boulder, Colorado.

MATERIALS SCIENCE

Making a Bigger Chill With Magnets

LOS ANGELES—Refrigerator magnets are best known for holding shopping lists and old postcards onto refrigerator doors. But in a few years, much more powerful magnets could be the key to keeping food cold in so-called magnetocaloric refrigerators, which would be more energy efficient and less polluting than standard models. Now a new class of magnetocaloric materials, announced here last week at a meeting of the American Physical Society, could make these magnetic refrigerators more practical and versatile.

The magnetocaloric effect works when strong magnetic fields align quantum-mechanical "spins" of electrons within atoms. This transition reduces one aspect of the randomness, or entropy, of the atoms. But according to laws of thermodynamics, some other aspect of randomness has to increase in compensation, so the atoms increase the randomness of their velocities—vibrating and heating up. Once this heat is carried away by a coolant such as water, the field is removed and the effect works in reverse, chilling the material and cooling a refrigerator. To date, the peak performance has been with the element gadolinium.

By adding various amounts of silicon and germanium to gadolinium's crystal lattice, Vitalij Pecharsky and Karl Gschneidner of the Ames Laboratory at Iowa State University discovered a new class of materials that can chill two to six times further in a single magnetic cycle, meaning that the refrigerators could operate with weaker magnetic fields or less material. Depending on the germanium-to-silicon ratio, the new materials also operate from about room temperature all the way down to –253 degrees Celsius. The cold end of the range would allow magnetocaloric freezers to liquefy hydrogen or natural gas for use in clean-burning power plants or future automobiles.

To come up with the new compounds, the team followed up on hints that magnetocaloric materials containing gadolinium and either silicon or germanium—but not both—prefer a different range of temperatures than gadolinium alone. "We're not trying to come up with exotic new compounds out of the pure blue sky," says Gschneidner. The surprise, he says, was that the magnetocaloric effect turned out to be far larger when both germanium and silicon were added to the material.

"These new materials give you a lot more flexibility in designing magnetocaloric [refrigerators]," says Carl Zimm, a senior scientist in magnetic refrigeration at Astronautics Corporation of America in Madison, Wisconsin. The team is still working on making enough of the material to try it out in Zimm's prototype gadolinium-based refrigerator, which has been running for about a year. The test should take place "within a couple of months," says Gschneidner.

–James Glanz