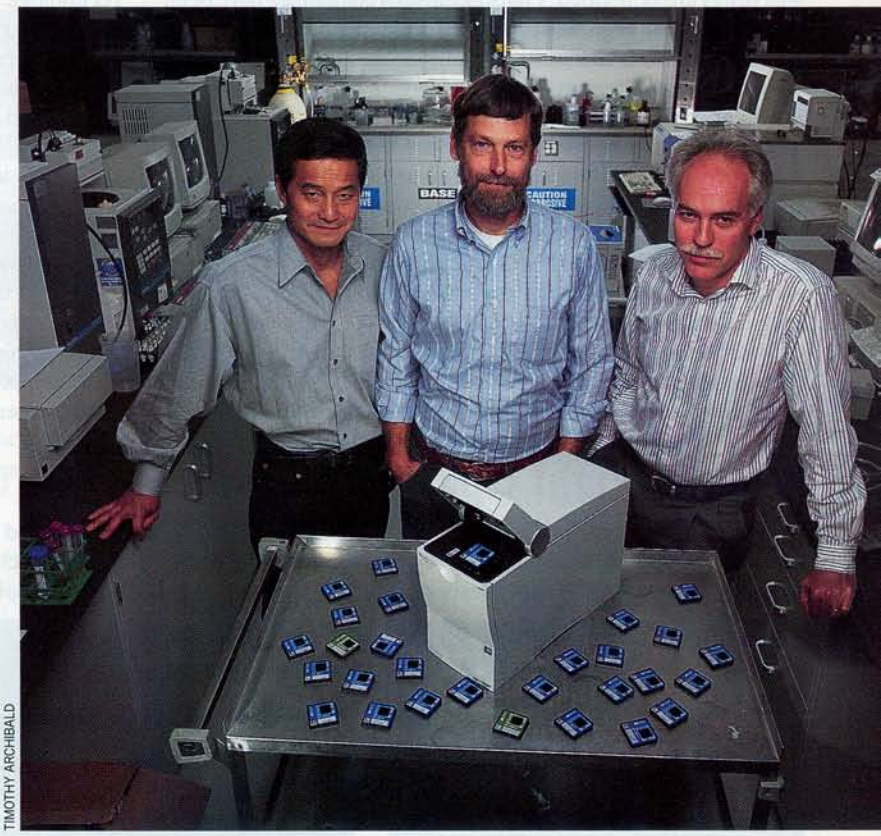
GOOD-BYE, Test Tubes HELLO, Labs-on-a-Chip

Biotech experiments and germ-warfare tests are getting done faster and cheaper in chips with tiny mazes of valves and reaction chambers.

BY STUART F. BROWN



TIMOTHY ARCHIBALD

Much of that lab gear in back is replaced by a machine with a chip from Caliper Technologies, founded by (from left) Calvin Chow, Wally Parce, and Michael Knapp.

Like music fans sliding CDs into stereos, scientists in biochemistry and pharmaceuticals labs have recently been loading little square thingies called LabChips into novel, toaster-sized machines. They're free to take a 20-minute coffee break while the machines, which replace a whole array of conventional equipment, automatically sort through samples of DNA fragments that have been placed in tiny wells on the chips. When the analysis is complete, the lengths of the fragments and their respective quantities are displayed on a computer screen. Already tested in prototype and now in production, the machines have slashed the time and effort required to perform an important quality-control check in the exploding field of biotechnology.

Another compact new instrument may soon be a standby in the chaos of a battlefield. Suspecting that an enemy may be unleashing deadly anthrax, say, or plague or smallpox, a medical corpsman will be able to open a portable device no larger than a suitcase, similar to two prototypes already delivered to the U.S. Army. He will then pour material gathered by a helicopter-borne air-sampling device into small plastic cartridges inside the instrument, and press a button to start a biochemical analysis. If the troops are lucky, the screen on the portable lab's laptop computer will report a half-hour later that fears of a biological warfare attack are unwarranted and that no protective measures are necessary. The battery-powered instrument, which can be carried by hand, gives answers that now require a mess of equipment cramming the back of a Humvee, as well as a trailer-mounted electric generator.

Welcome to the nascent world of microfluidics, where laboratory benches cluttered with chemistry paraphernalia are being replaced by a new breed of devices built around chips and other elements—some small as a fingernail—that are the counterpart of a PC's microprocessor. These labs-on-a-chip contain tiny mazes of channels, valves, and chambers through which minute amounts of liquid chemicals can be pumped and reactions monitored. Using no moving parts and requiring little or no assembly, labs-on-a-chip can measure and dispense volumes as small as one picoliter, an incredible fifty-millionth of a drop of water.

That's a spectacular advance, because chemicals used in drug research cost as much as several hundred thousand dollars an ounce. Some DNA samples are just as precious. By handling such tiny quantities, microfluidics makes possible a proliferation of experiments that would have been unaffordable a few years ago.The eclectic teams of scientists and engineers drawn into the new field believe that the low cost and high speed of tiny-scale reactions will do for chemistry what the microelectronics revolution did for computing. They even talk of a "Moore's law" for chemistry, a reference to the doubling of computer power every 18 months or so.

The new technology is already beginning to revolutionize the way drugs are developed. Pharmaceuticals companies have been forming partnerships with pioneering microfluidics startups to bring the speed and cost benefits of miniaturization to the tedious process of research. The advances come not a bit too soon. According to the Pharmaceutical Research and Manufacturers of America, a Washington, D.C., trade group, drug companies spend $500 million and 12 to 15 years to bring a new medicine to market, an investment that only three out of ten new drugs earn back in sales. Another party putting its weight behind microfluidics is the Pentagon, which can't wait to equip the Armed Forces with those portable labs for quickly detecting biological warfare threats.

Microfluidics is headed for other big jobs, such as environmental monitoring and medical diagnostics. Visionaries foresee a day when workers taking soil samples at suspected pollution sites will be able to identify contaminants on the spot. Or a doctor will be able to place a drop of your blood or urine into a disposable device that will quickly spot the bug that ails you by recognizing the pathogen's DNA. Another blockbuster medical application could take the hit-or-miss element out of the way doctors prescribe drugs: Future instruments with microfluidic chips could identify the genetic variations in patients that make a drug helpful to Smith, ineffective for Jones, and possibly lethal for poor Throckmorton.

One of the most ardent believers in microfluidics is Rick Kniss, who runs the chemical-analysis group at Agilent Technologies, a subsidiary of Hewlett-Packard that's slated to become a separate company. Says Kniss: "The worldwide market for various analytical instruments used in chemistry and biotechnology is about $16 billion a year, and I think a large portion of it can be miniaturized. We expect a tenfold increase in the quality and amount of information generated as a result of this technology."

Agilent, which makes the machine that sorts DNA fragments, is betting big on microfluidics. The device, called the HP-2100 Bioanalyzer, uses LabChips, a trademarked name, from Caliper Technologies, a startup in Mountain View, Calif. The companies say that together they will spend $100 million over the next several years to develop and commercialize lab instruments using microfluidics.

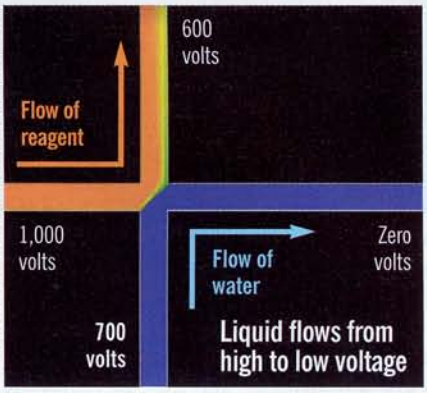
About 80 companies worldwide are working in the field, estimates John West, marketing director at Microcosm Technologies, a company in Research Triangle Park, N.C., that develops software for designing lab-on-a-chip systems. About 50 of the companies are in the U.S., West says, and range from large established firms such as Motorola and PE Corp. to a flock of startups founded in the past few years.

Microfluidics is a logical outgrowth of the automation that has been playing a growing role in biochemical experiments. Small robots adapted from the electronics industry have proved a boon to researchers by taking over repetitive tasks such as pipetting so-

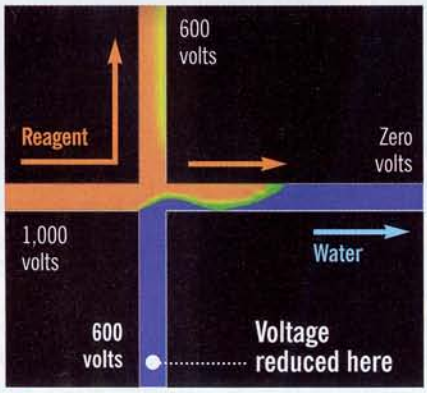
Coming Right Up, One Nanodollop of a Chemical

One building block of a lab-on-a-chip is a "virtual valve." Using a principle called electro-osmosis, it dispenses chemicals in amounts as small as a fifty-millionth of a drop of water. Microcosm Technologies of Research Triangle Park, N.C., which produced the simulation below, has worked with Caliper Technologies of Mountain View, Calif., on developing virtual valves.

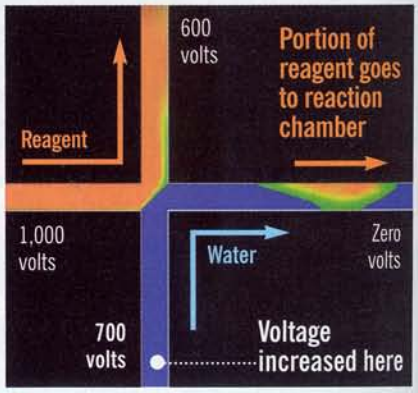
FORTUNE DIAGRAM / SOURCE: MICROCOSM TECHNOLOGIES



1. In this computer simulation, fluids flow from high-voltage to low-voltage areas. The electrical differential regulates the pressure and keeps the fluids from mixing.



2. A reagent is dispensed when the voltage propelling the water is briefly cut to 600 volts. This lowers water pressure at the junction, allowing a bit of reagent to enter the water channel.



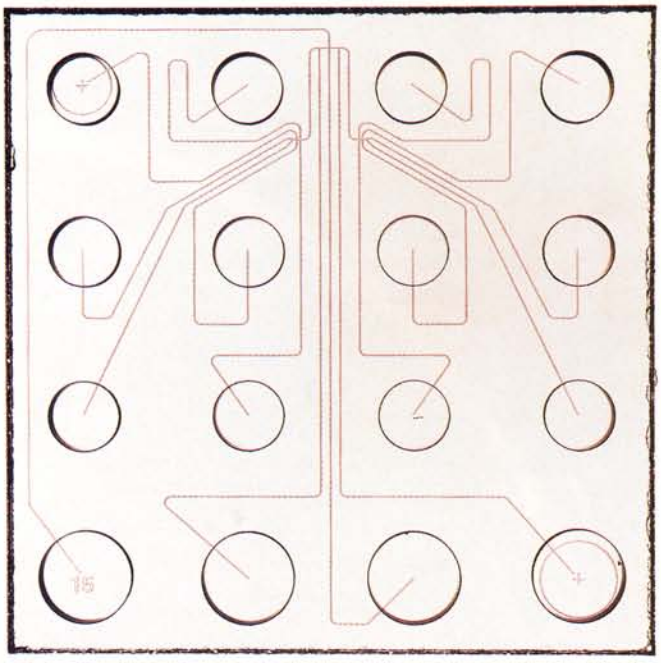
3. Voltage is returned to 700 volts, restoring the separation of the fluids at the junction while a precise amount of reagent is conveyed to a reaction chamber.

lutions into thousands of test tubes. But why stop at substituting mechanical arms for human ones, said the people who dreamed up the technology. Let's shrink the whole process to a minute fraction of the scale of benchtop chemistry. One advantage, they reasoned, is that in microscale chemistry, the force of electricity can be used to move streams of chemical solutions in a precise way that the science of fluid dynamics can predict. Thus was born the idea of labs-on-a-chip.

Key parts of the plumbing needed for microfluidics emerged in the late 1980s, when scientists working in the U.S., Switzerland, and Canada began to experiment with moving chemical solutions through networks of microfabricated channels etched on a sheet of glass. One of the scientists, Michael Ramsey at Oak Ridge National Laboratory, patented a so-called virtual valve. Despite its name—the term "virtual" these days is often associated with computer simulations—this is a real X-shaped intersection of four tiny channels at which different fluids arriving at the crossroads can be electrically steered, dispensed, or mingled. The valve is virtual only in its lack of moving parts.

The technique for manipulating fluids in a lab-on-a-chip, called electrokinesis, works in several ways. One is electroosmosis, which uses electric fields to propel conductive, water-based solutions. Another is electrophoresis, which separates molecules in an electric field according to their charge. By connecting networks of channels, valves, and tiny chambers where reactions occur, microfluidics designers can accomplish all that's done by full-sized plumbing in many conventional chemistry experiments.

To track what's going on, microfluidics tags the samples and reagent chemicals pumped through the tiny devices with dyes that fluoresce, or glow, when a reaction has taken place. A laser-diode sensor at the end of the pipeline monitors the fluoresence, which sometimes varies in intensity or color, depending on the outcome. Then the sensor converts the optical signal into an electrical one that's displayed on a computer screen, where researchers can interpret the results.


TOM SCHIERLITZ

The top 12 "wells" in the LabChip used by Agilent Technologies (shown actual size in its housing at left) hold DNA fragments that get sorted by size as they flow through its channels. Other wells hold materials used in the sorting process.

Caliper Technologies, a leader in the microfluidics business, is working in two important areas. To speed the search for new medicines, it has produced LabChips for the high-throughput screening of drug candidates. Using a method called combinatorial synthesis, pharmaceuticals companies are now able to generate "libraries" containing hundreds of thousands of compounds that they want to test against "targets"—biological molecules inside the human body, including gene segments, that they hope to block or activate to effect cures.

Before long, each leading drugmaker's library is expected to contain a million or more compounds, all of which it may want to aim at multiple targets in a deluge of individual reactions. Working with pharmaceutical partners like Hoffmann–La Roche, Amgen, and Eli Lilly, Caliper has built high-throughput screening machines. Recently it shipped two to drugmakers, and it keeps two more running in-house for pharmaceutical clients that want to purchase screening services rather than hardware.

In a typical experiment, an enzyme and a biological molecule are first brought together inside a high-throughput screening chip, where they react in a way that produces fluorescence. Then, adding samples of drug candidates from the library one at a time and watching for changes in fluorescence, the machine can spot compounds that alter the reaction in a potentially beneficial way. To add the drug samples, the machines use miniature capillary "sippers" that draw dollops measured in nanoliters—a billionth of a liter—in succession from tiny wells on 96-well "sample plates" and load them in succession in the chips.

A robotic feeder keeps sample plates moving into the apparatus, which can conduct 100,000 experiments a day. Like a microprocessor doggedly crunching through huge numerical calculations, the chip's economics bring the brute-force search—unthinkable until now—within the budgetary reach of drug researchers. Assays that cost between $1 and $10 apiece using benchtop methods cost as little as a dime when performed on a chip.

Caliper's other work led to its partnership with Agilent Technologies. DNA "sizing" was chosen as the first task for Agilent's HP-2100 Bioanalyzer because it is so widely performed. But the machine, which with peripherals costs $19,500, can run any experiment for which Caliper's designers have created a specialized LabChip. Like kids popping game cartridges into their Nintendo players, scientists will be able to load the appropriate square plastic modules, which encase a disposable glass chip etched with microplumbing tailored to each experiment. LabChips for other experiments are on the way, Caliper executives say.

DNA sizing is an essential verification step for researchers working with biology's master molecule. Using substances called restriction enzymes, they "cleave" gene segments of potential medical interest from the very long DNA strands of which they form a part. Before investing any more effort in an experiment, they need

to know whether the DNA sample includes the correct segments, each of which can be identified by its unique length, and if so, in what proportions they are present.

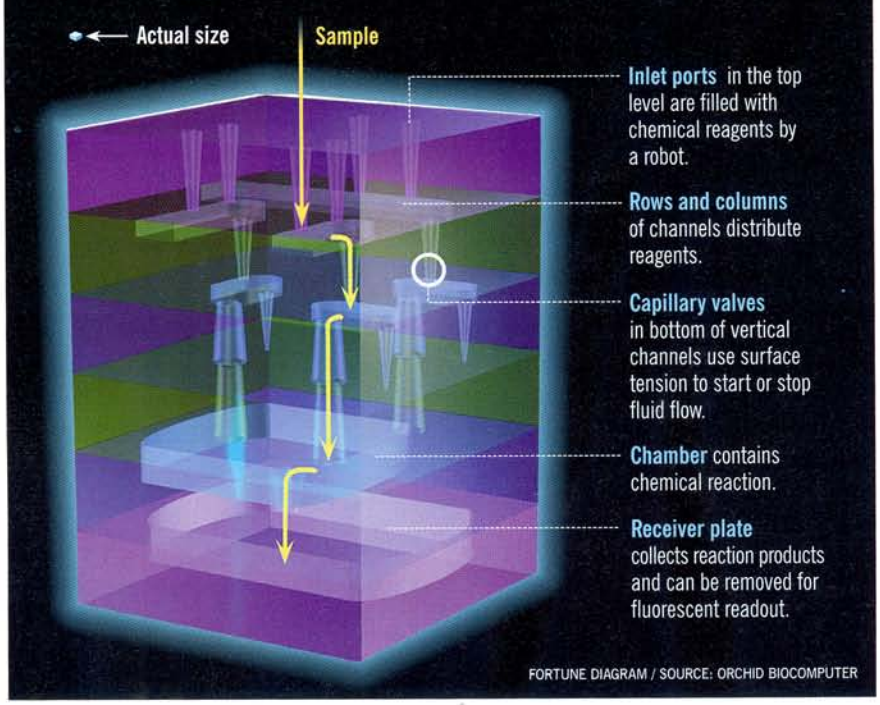
The conventional way to find out is through gel electrophoresis, a process that typically uses a thin slab of Jell-O-like agarose goo sandwiched between two vertical plates of glass about eight inches square. A DNA sample is loaded into a reservoir at the top of the device, and an electric current is applied across the gel, drawing the DNA slowly down through it. The smaller the segments are, the more quickly they reach the bottom, causing the sample to stratify by size into bands that become visible when the gel is stained in a subsequent step. Finally, the stained gel is scanned into a computer for analysis using special software. The whole process can take up to half a technician's day.

With Caliper's LabChip, the techie's job is far simpler. At the beginning of the test, the factory-fresh chip's internal plumbing is empty, so the first step is to fill all the passages with a linear-polyacrilamide gel, consisting of polymer balls floating in a solution, that is stored in one of the circular wells molded into the chip's plastic housing. The well also contains a fluorescent dye that will attach itself to DNA. Then all the researcher has to do is inject DNA samples into 12 other wells on the chip's housing, each of which can hold a different sample.

When the LabChip is loaded into the machine and the cover is closed, little wire electrodes descend into the wells. Hooked to power supplies, the electrodes drive the chip's electrokinetic choreography, pumping the DNA samples through the chip's tiny internal channels on cue from software running on a PC. As the DNA from the first sample well is pulled into the chip, it's directed into a separation channel 1.7 centimeters long, where a process known as capillary electrophoresis occurs. Big DNA segments get more bogged down by the stationary polymer balls in the gel than small ones do, so the segments arrive at the end of the channel in ascending order of size.

A MICROLAB WITH MANY LEVELS

Microfluidic chips made by Orchid Biocomputer are arrays of 96 to 1,536 tiny three-dimensional "microreactors" like the one shown here. They synthesize drugs and perform biochemical tests using as many as six interconnected levels.



FORTUNE DIAGRAM / SOURCE: ORCHID BIOCOMPUTER

By clocking the respective arrival times of the fluorescing DNA segments, the optical sensor scanning the channel's terminus can precisely determine their size. And by gauging the brightness of the fluorescence, it detects how many segments of each size are in the sample. Next the power supply pulls in the DNA from sample well No. 2, and so the test continues until all 12 samples have been sized. The process saves a lot of time because the DNA travels less than two centimeters through the polymer gel, instead of 20 centimeters through the agarose stuff used in the older method. Finally the used LabChip, which costs $12, gets chucked. So does any DNA that flunks the sizing test.

The stinginess with materials that labs-on-a-chip offer is just what many biochemists have been longing for. Eos Biotechnology in South San Francisco, Calif., a two-year-old startup pursuing the discovery of anticancer drug targets, has served as a "beta" test site, a sort of guinea pig volunteering to try out an early version of Agilent's machine in hopes of achieving better lab results. "A lot of our DNA samples are from biopsies of human tumors, and we don't get very much to work with," says scientist Keith Wilson. "When we were using gel electrophoresis with such small samples, we could only afford to do experiments without backup quality-control checks. But microfluidics takes almost nothing of our sample, and it's a very fast analytical tool that gives us exquisite separation of DNA fragments."

Caliper and other microfluidics companies are likely to come up with advanced versions of their creations in a lot less time than the computer-chip people did. That's because most of the needed manufacturing equipment has already been developed for the silicon world, and labs-on-a-chip are comparatively easy to fabricate. Compared with the 0.25-micron feature sizes on the latest computer chips, the microchannels on a microfluidic chip are big—about ten microns deep and 50 microns wide—though this is still only half the diameter of an average human hair.

Caliper's chips are made scores at a time on square plates of high-quality glass, much as batches of memory or logic chips are etched on a silicon wafer. LabChips require far fewer process steps. William Wright III, a silicon-fab veteran who recently became Caliper's vice president of operations, says the channels and other features are cut into the glass plate's surface with a pattern-and-etch process using hydrofluoric acid, aggressive stuff that eats glass. Then a plastic cover is bonded in place to form the roof of the labyrinth. This is done very carefully so as not to block any of its shallow features. Learning to do this consistently, and thus improving the yield of usable chips, is part of

the manufacturing wisdom Wright's group is accumulating.

Each completed chip is run through a videocamera inspection system that automatically checks its topography and looks for flaws in the glass or plastic that could cause unwanted fluorescence. To drive down costs for chips using mild chemical reagents that don't dissolve plastics, Caliper is working on all-plastic versions made by a "hot embossing" process; it uses a mold that's first etched into silicon and then plated with nickel.

Caliper has the capacity to produce about a million chips a year if the fab runs three shifts a day. Should the joint venture with Agilent generate higher demand, more fab equipment will be located somewhere cheaper than Silicon Valley. Becoming a high-volume chipmaker is the goal of this company, which uses Intel as its model. Says Calvin Chow, Caliper's chief operating officer and one of its three founders: "Intel looks for software developers to promote the need for their chips. We're looking for more partners like Agilent that will build systems around our microfluidics chips."

Caliper's chips are two-dimensional affairs, with all the plumbing etched on a single layer. Chips of a very different kind have been wrought by Orchid Biocomputer in Princeton, N.J. A privately owned spinoff from Sarnoff Corp., the former RCA research unit that's now part of SRI International, Orchid specializes in three-dimensional chips with as many as six interconnected layers. They have the gee-whiz quality of a 3-D chess game.

JOSEPH PLUCHINO

Orchid Biocomputer, headed by Dale Pfost, makes 3-D chips that help find key DNA segments. Pharmaceuticals companies also use the chips to churn out drug candidates.

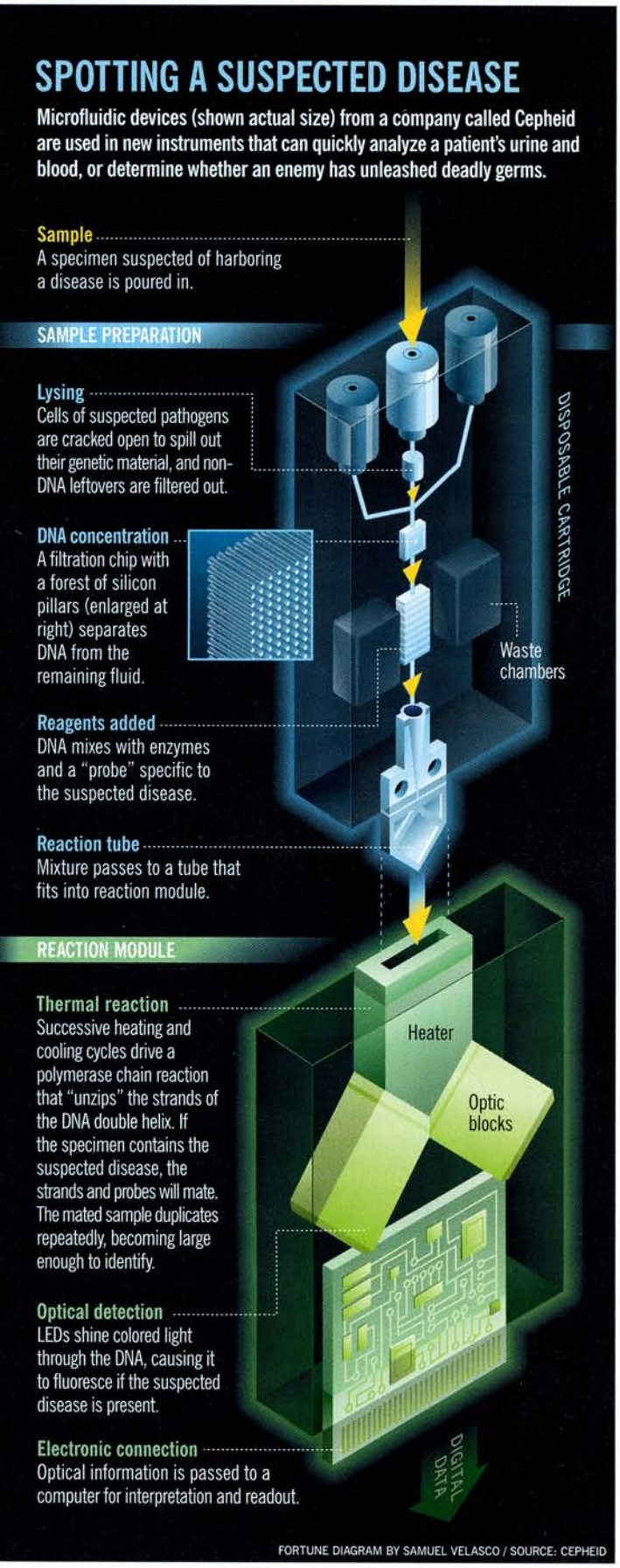
Orchid aims to assuage the pharmaceuticals industry's hunger for doing things more quickly and cheaply in two ways. The first is to supply combinatorial-synthesis chips. These enable drug companies to breed those millions of potentially therapeutic molecules that are screened for possible medical value in chips like Caliper's. Combinatorial chemistry involves shuffling the molecular building blocks of promising drug compounds, or "leads," to create dozens or even hundreds of variants that might have even better healing potential. Known as lead optimization, the strategy helps drugmakers pick the best candidates for further research and enables them to patent every closely related drug before a competitor does.

The Orchid chips that craft new molecules are arrays of "microreactors" less than a quarter of an inch on a side (see diagram). Each array is made in one piece from glass, silicon, quartz, or plastic, sandwiched in layers. The modular design permits multiples of 96 different reactions, up to 1,536, to be conducted simultaneously in pinhead-sized chambers located in each module's lower layers. In the works for collaborator SmithKline Beecham, which holds a stake in Orchid, is the mother of all high-volume synthesizers: a 12,228-well chip system called Chemstream 12K, which the drugmaker will use to expand its chemical libraries.

The upper layers of the microreactor modules form multistoried networks of valves and capillaries that deliver samples and reagents squirted in by pipetting robots. The modules can even conduct multistep reactions on the surface of special plastic beads located in the reaction chambers, which are rinsed between steps to remove leftover reagents. When the synthesis is complete, the final product is chemically stripped from the beads and piped away. While the upper layers of Orchid's chips are reusable, the bottom layer is thrown away when the experiment is done.

Orchid's other major effort is in the emerging field of pharmacogenetics, which could lead to the development of drugs targeted at particular subgroups of patients. Here the focus is on deviations from the usual DNA sequence called single-nucleotide polymorphisms, or SNPs. These naturally occurring anomalies, which biotechies call "snips," are believed to be the basis for many human diseases. They also make all of us different from one another and are responsible for the adverse effects some patients experience with drugs that don't bother other people. Side effects often vary because drugs are converted into somewhat different metabolites in different patients, some lethal. Last year the *Journal of the American Medical Association* reported that bad drug reactions accounted for more than 100,000 deaths a year among U.S. hospital patients.

SNPs occur in every 100 to 500 of the three billion nucleotides,



or chemical building blocks, that compose the human genome. Of these variations, perhaps 100,000 to 200,000 are located in protein-coding sequences that may make people vulnerable to various illnesses. Orchid builds 3-D chips for "scoring," or locating, SNPs along the DNA chain.

According to Orchid CEO Dale Pfost, SNP scoring is going to be the key to the second phase of the genomics revolution. The first phase is the sequencing of the human genome, in which the U.S. government and several private companies are mapping the procession of nucleotides, designated by the four letters A, C, G, and T, that make up human DNA. At several sites in the U.S. and abroad, rooms full of sequencing machines are grinding away right now to complete this monumental task.

To pinpoint SNPs, Orchid uses a method called genetic-bit analysis developed by Molecular Tool, a company it acquired last year. Genetic-bit analysis was originally developed for the lucrative business of racehorse paternity testing and was later used to identify deadbeat dads.

Marrying genetic-bit analysis and microfluidics, Pfost says, opens a much broader opportunity. It offers an "affordable way to zoom in" on SNP locations and "cull through to find the ones that will help choose the right drug for an individual patient." Already, he says, "our company has uncovered interesting correlations between certain SNPs and treatment practices."

But the search has only begun. A consortium of ten pharmaceuticals companies and a foundation has agreed to pool resources over the next several years to locate 300,000 SNPs. They hope to make the information available publicly before it's discovered independently by firms currently engaged in a kind of genetic land grab. To find which of these SNPs are associated with a given disease, drug researchers will have to get DNA material from perhaps 1,000 afflicted patients, including subgroups who have had a good reaction to a drug, a bad reaction, or no reaction at all. By scoring 1,000 potentially relevant SNPs in each of these patients—for a total of a million scores—researchers hope to identify the ones that really matter.

That's going to provide lots of work for Orchid's chips. But Pfost says this is cheap compared with the alternative. Running a million SNP scores with microfluidics can be done for about $200,000 today, he figures, vs. the $2 million to $3 million cost of sequencing all the patients' DNA and looking for differences. By the middle of next year, Orchid plans to have running a high-throughput "MegaSNPatron" facility designed to score more than a million SNPs daily, using samples provided by a network of university researchers. The pharmaceuticals industry hopes not only to devise drugs tailored to individuals but also to salvage potentially useful drugs that have ended up on the shelf because some patients had bad reactions during clinical trials. Someday a doctor may be able to "type" a patient's SNPs and safely prescribe a drug that would be ineffective, unpleasant, or fatal to another person.

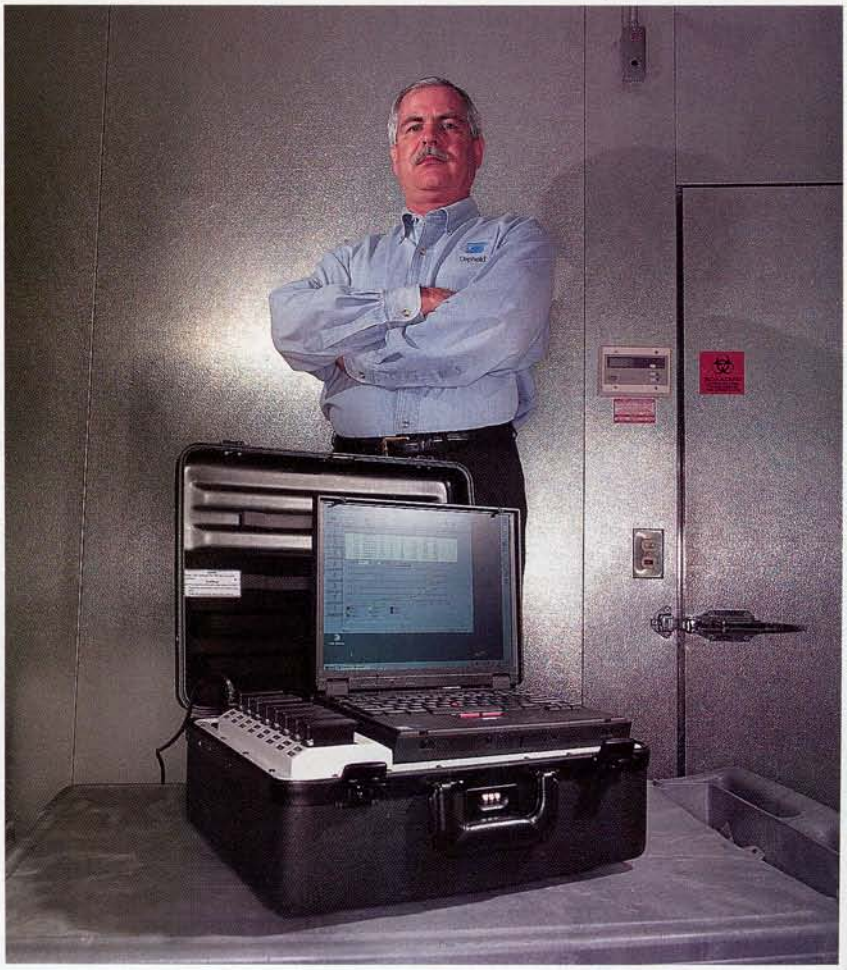
Another category of ingenious microfluidic devices, capable of performing DNA-based diagnosis in a clinic or detecting biological warfare attacks in a combat zone, is being developed by Cepheid in Sunnyvale, Calif. Companies working with picoliter and nanoliter volumes of fluids might regard Cepheid as ineligible to join the microfluidics club because its systems work with relatively giant samples measured in milliliters, or a thousandth of a liter. But the aim is the same: to conduct bio-

chemical tests on the smallest practical scale.

Cepheid's tests need large quantities because they work with raw specimens, such as urine or blood, in which pathogens are present in low concentrations. The instruments have microfluidic modules (see diagram) that prepare the sample, concentrate the DNA of a suspected pathogen, and then create the conditions under which it can be identified.

In preparatory steps, the specimen is run through a disposable, molded-plastic cartridge. Here the suspected pathogen's cells are "lysed," or cracked open, to spill out their genetic material. Then, after filtering to remove cellular leftovers, the DNA-bearing fluid passes through an extraction chip that reduces its volume from five milliliters to 100 microliters, or millionths of a liter. This little chip of micromachined silicon is populated with densely packed tiny columns that are 25 times as tall as they are wide. Produced by a recently developed process called deep reactive-ion etching, the chip's forest of columns has a surface texture with an affinity for DNA, which clings to the columns as the sample is pumped through. Think of the chip as a sieve.

In the next step, the concentrated DNA mixes with reagents needed to perform the same polymerase-chain reaction (PCR) that identified the famous stain on Monica Lewinsky's dress. These ingredients include the enzyme polymerase, which induces DNA to copy itself. The other key part of the recipe is what's called a DNA probe, a partial strand of a suspected pathogen's own DNA that can be synthesized in a laboratory. If the module is being used to test whether a patient has, say, gonorrhea, it will contain a gonorrhea DNA probe.



TIMOTHY ARCHIBALD

The Pentagon likes the Briefcase Smart Cycler from Cepheid, headed by Kurt Petersen. It uses miniaturized devices to test for eight different biological warfare germs in a hurry.

If the probe finds a matching stretch on the suspected pathogen's DNA and mates with it, the patient has the disease. But before this matchup can take place on a scale big enough for detection, the temperature must be repeatedly raised and lowered. The cartridge containing the lysed and concentrated DNA, plus reagents, is inserted into an "I-CORE" reaction module with miniature ceramic heating elements that perform the thermal cycling essential to the PCR process. When the heaters quickly raise the sample's temperature to 95°C for a few seconds, the spiraling ladderlike strands of the suspect's double-helix DNA molecules unzip into two.

Then the temperature is brought back down to 60° C. If the DNA half-strands are from gonorrhea, they will attract some of the probe segments. A kind of miracle occurs when the temperature is raised again to 95°C. The polymerase enzyme manufactures all the building blocks needed to complete the parts missing from the probes. Each half-strand grows into a complete, two-stranded DNA molecule, thus doubling the number of DNA molecules in the sample.

By cycling the temperature up and down 30 to 40 times—prompting further unzipping, matching up, and filling out of missing DNA sections—the device doubles and redoubles the sample size to the point where a disease can be identified. Now LEDs shine colored light through the sample, whose distinctive fluorescence is detected by an optical sensor. At a recent conference Cepheid demonstrated a prototype system for clinical use, containing a cartridge with two probes for simultaneous tests of two sexually transmitted diseases. By amplifying and detecting the two pathogens' DNA, the system in half an hour spotted both gonorrhea and chlamydia in a five-milliliter urine sample.

"Our system uses miniaturization to run five to ten times faster than more expensive PCR equipment that's on the market," claims Kurt Petersen, Cepheid's president. The amplification and detection method Cepheid uses was originally developed at Lawrence Livermore National Laboratory by Allen Northrup, now the startup's chief technical officer. Petersen was an early pioneer in commercializing tiny mechanical devices called MEMS, or microelectromechanical systems. He also founded Lucas Novasensor in Fremont, Calif., a maker of micromachined silicon pressure-sensing devices that will fabricate Cepheid's little DNA-extraction chips.

Cepheid's strategy is to have all its components made by outside suppliers, doing only assembly and testing in-house. Its customers will include makers of medical diagnostic and life-science research equipment, says Chairman Thomas Gutshall. The company has formed a partnership with Innogenetics in Ghent, Belgium, to develop gene-based diagnostic systems.

The military, which is in a bit of a panic about the threat of

biowarfare, has been Cepheid's biggest source of revenue to date. The U.S. Army's Medical Research Institute of Infectious Diseases at Fort Detrick, Md., has received two prototypes of Cepheid's Briefcase Smart Cycler, a portable lab that's right out of the movies. Up to eight amplification-detection cartridges, containing DNA probes for different pathogens that the enemy might be spreading around, can be inserted in the mobile lab for simultaneous tests. Late this year, Cepheid expects to be shipping production versions that can handle 16 cartridges.

Eclectic is the word for the teams of people working in the rapidly evolving microfluidics business. A visitor will find under one roof, striving to learn and speak one another's lingo, chemists, biologists, electrical engineers, mechanical engineers, chip-manufacturing experts, and uncommon folks like Andrea Chow. She's a fluid-dynamics specialist hired by Caliper from Lockheed-Martin, where she was working on rocket fuel manufacturing techniques. Says Chow: "I used to deal in millions of pounds of material for the space shuttle's solid rocket motors. Now I'm working with picoliters." Multidisciplinary types who got their degrees in fields that cross academic boundaries tend to thrive in such an environment.

The early generations of microfluidic devices were pretty simple, developed more or less through trial and error. But as designers strive to pack more experimental horsepower into the tiny labs, complexity starts to gang up on them. The microprocessor people went down that road years ago, and the design of today's multimillion-transistor chips would be unthinkable without so-called electronic design automation, or EDA. This consists of specialized packages of computer-aided design (CAD) software containing modular blocks of proven circuitry that can be combined to create much larger systems. Other tools help with layout, packing features efficiently on a chip and arranging them so that the interconnections are as short as possible to maximize operating speed. The microfluidics crowd is starting to develop a counterpart of EDA.

The need is growing. Wally Parce, Caliper's research chief, says some of his company's chip designs will soon be too complex to fit on one level and will require the move to 3-D architecture like Orchid's. Visualizing all the interrelated structures and chemical events won't be possible without computer modeling. Caliper is working with funding from the Pentagon's Defense Advanced Research Projects Agency (DARPA) to test a microfluidics-design software package called Flume-CAD, under development by Microcosm Technologies. An MIT spinoff, Microcosm got started with a micro-electromechanical systems design program called MemCAD, also funded by DARPA.

John Gilbert, chief technical officer at Microcosm's development center in Cambridge, Mass., says the task of modeling microfluidic chips is greatly helped by the fact that fluid flows in their small channels remain laminar, or unruffled. Thus, they are amenable to modeling by fluid dynamics equations. That's better than the everyday world of sinks and lawn sprinklers, where fluid flow becomes chaotic and unpredictable. Laminar flow is a liability, however, when it comes to mixing things. The orderly streams can be reluctant to merge, so designers sometimes split flows into interleaved liquid fingers that diffuse into one another quickly.

Timing is everything in a lab-on-a-chip. The size and routing of channels determine when fluids reach reaction or separation chambers and how much time the chemicals have to complete their reactions. "These are time-of-flight and time-of-arrival problems," says Gilbert. "We use a technique called time-domain modeling to help predict how all the components in a system will work, and whether the diameters, radii, and other features are correct to synchronize events while keeping everything as small as possible and creating the best conditions for the chemicals."

Whatever the methods used to design them, new chips are on the way that will perform impressive tasks. Parce at Caliper likes to display a prototype of a Lab-Chip designed to discover the dose-response curve of a drug that blocks a receptor on a cell. In steps, it will add increasing concentrations of the blocker and measure the outcomes. Says Parce: "This is the equivalent of setting up a whole test-tube rack of experiments, and they've got to be done one at a time because there's a precise incubation time and you can't miss the response." Parce is pleased to report that the chip "was completely modeled with CAD, and it actually worked the first time we tried it, so our software is at this level." In their small-is-beautiful world, the microfluidics people are doing some really smart stuff. F