DNA Fingerprinting Takes the Witness Stand

DNA typing is providing forensic scientists with a formidable tool for identifying murderers and rapists

RANDALL JONES HAD A PROBLEM. His car was stuck in the mud and he needed a tow. Jones's solution to that problem would land him on Florida's death row—and his own DNA would provide part of the evidence that put him there. The Jones case is one of a growing number in which newly developed methods of DNA analysis are helping to identify the perpetrators of violent crimes, mainly rapes and murders.

The crimes for which Jones was convicted were especially vicious. After his car became stuck, he approached a pickup truck that was parked at a nearby fishing ramp. The occupants, a man and woman in their early twenties, were sleeping. Jones shot them both in the head with a high-powered rifle at close range. He then dragged their bodies into the woods and used the truck to pull his car out of the mud. About 40 minutes after murdering the victims, Jones went back to their bodies and raped the woman. By doing so he left behind a sample of his sperm, and therefore of his DNA, that would inextricably link him to the crime.

The older forensic tests that can be conducted on the biological evidence usually found at crime scenes, including semen, blood, and hair, can identify a suspect with only about 90 to 95% certainty, a circumstance that leaves room for argument that the blood or semen belonged to someone other than the accused. But the type of analysis that was run on Jones's DNA can rule out everyone else in the world as a possible perpetrator of a crime.

"Essentially what that means is that it is an absolute identification. Before, we never had that. It's the greatest boon to forensic medicine and law since fingerprinting," says an enthusiastic Mac McLeod of the State Attorney's office in Palatka, Florida, who prosecuted Jones.

In the limited number—a few more than a dozen—of cases in which the results of DNA typing have been used in court, the evidence has generally been allowed, although in some cases it could still be challenged if the verdicts are appealed. The legal system has not always been ready to accept new scientific technologies. But so far at least forensic DNA typing has not encountered major obstacles. "The legal community is moving about as fast as I've seen them move," says Daniel Garner of Cellmark Diagnostics of Germantown, Maryland, which is a subsidiary of the British conglomerate Imperial Chemical Industries.

Cellmark, where the analysis of Jones's DNA was performed, is one of three commercial companies in the United States currently doing the forensic DNA work. The other two are Lifecodes Corporation of Elmsford, New York, and Forensic Science Associates of Richmond, California. The forensic application of the technology is expected to spread rapidly, however. Public law enforcement agencies, notably including the Federal Bureau of Investigation, are also gearing up to do DNA typing.

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According to the FBI's John Hicks, the agency expects to begin doing forensic DNA typing by October. Moreover, the FBI is instituting a program for training forensic scientists from state crime laboratories around the country in the technology. "In about 3 years, you will see it as a routine sort of thing," Hicks predicts. One likely outcome is the eventual establishment of DNA data banks that could be used much as are the FBI's fingerprint files for identifying the perpetrators of violent crimes.

The method that Cellmark used to type Jones's DNA was devised by Alec Jeffreys of the University of Leicester, England, and goes by the name of DNA fingerprinting (*Science*, 1 August 1986, p. 521). Jeffreys found that the human genome contains many "minisatellite" regions that consist of tandemly repeated nucleotide sequences, all of which have a common core sequence. The number of repeated units in the minisatellites, and thus their length, varies from one individual to the next.

These variations in the DNA can be detected by restriction fragment length polymorphism (RFLP) analysis, a now standard research technique in molecular biology. (Polymorphism is molecular biology jargon for variation.) To do the analysis, a DNA sample is first digested with a restriction enzyme that cuts it at specific sites into fragments, which are then separated according to size by gel electrophoresis.

For detecting the fragments from the minisatellite regions, Jeffreys produced probes, short pieces of radioactively labeled DNA that specifically bind to the core sequences. The locations of the fragments that bind the radioactive probes are detected with x-ray film. This produces a picture of a strip of 30 to 40 dark bands, which looks something like the bar codes used to identify items at the grocery store. The degree of length variation in the minisatellite is so high that every individual, with the exception of identical twins, produces a unique band pattern, much as every individual has a unique set of ordinary fingerprints.

More recently, Jeffreys has produced probes that recognize single sites in the minisatellite regions. These probes, which Cellmark uses for its forensic work, produce simpler banding patterns than the multisite probes and have also improved the sensitivity of the DNA typing. A high degree of specificity can still be obtained, however, by using several probes.

"You can differentiate one person from the rest of the world's population, which is about as definitive as you can get," Garner says. Garner could estimate, for example, that Jones's DNA pattern, which matched that of the sperm recovered from the woman's body, would be found in only one person in every 9.34 billion, which is about double the current world population.

Although this all sounds very complicated, the scientific testimony at the Jones trial apparently did not faze the jury. It brought in a guilty verdict—with a recommendation for the death penalty—in 12 minutes flat. McLeod attributes this partly to the clarity of Garner's testimony about DNA fingerprinting. In addition, McLeod says, "When you put the x-ray films on a light box, anybody can see the match."

He also says that he could have won a conviction without the DNA analysis, but points out, "In any murder case, you want to present as much evidence as you can. It can be scrutinized on appeal. We had overwhelming evidence of guilt, especially with the DNA fingerprinting."

In other cases convictions may not have been obtainable without DNA typing. In two of the rape cases in which Lifecodes has participated, the victims, one a blind woman and the other a woman with Alzheimer's disease, could not identify their assailants. Yet with the aid of DNA typing the prosecutors were able to win convictions.

According to Michael Baird of Lifecodes, that company's DNA-typing technique is comparable in specificity to Cellmark's, essentially allowing one person to be picked out of the world's population. Lifecodes' test is another RFLP analysis, but uses a different set of probes for detecting the DNA polymorphisms. Most of these probes detect specific variable number tandem repeat (VNTR) sequences.

The VNTRs resemble minisatellites in that they consist of a repeated sequence, with the number of copies varying from one person to the next. However, where there are usually many minisatellites of a given type in the genome, there is only one VNTR of each type. The VNTR probes also produce simple banding patterns.

The high specificity of VNTR typing is achieved by using several probes, each of which recognizes one VNTR site, to characterize a DNA sample. After the frequencies of the various patterns produced by each probe have been established for different ethnic groups, they can be used to calculate the probability of any particular combination of patterns occurring in an individual. With the probes Lifecodes uses, Baird says, that probability is about 1 in 10 billion.

Lifecodes has already provided DNA evidence in 11 rape or murder trials, one of which is still under way. Of the others, one resulted in a hung jury, one in an acquittal, and the rest brought in convictions. The hung jury, which occurred in a rape case in Orlando, Florida, represents one of the new technology's few stumbles.

The defense challenged the admissibility of the statistical evidence needed to support the identification of the suspect by DNA typing. The judge's refusal to allow the statistical arguments may have contributed to the jury's failure to reach a verdict. When the case was retried, however, the statistical evidence was allowed, and the jury returned a guilty verdict.

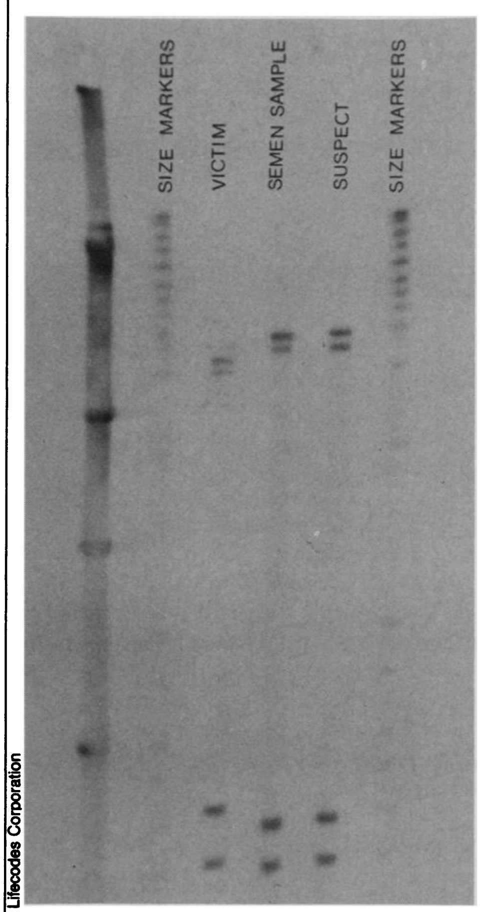
The acquittal came in a murder trial conducted last fall in Oklahoma. In this case, a man had disappeared and murder was suspected. Although no body had been found, investigators had uncovered a blood stain on a vacuum-cleaner hose in the apartment of a man that the presumed victim had visited. Researchers from Lifecodes were called in to perform what amounts to a paternity test on the blood sample.

They compared the DNA from the bloodstain with the DNAs of the missing man's mother and father. Because each parent contributes half of an individual's DNA, half of the bands in a child's DNA print should come from the mother and half from the father. That is what the Lifecodes group found in their analysis of the bloodstain DNA. "The DNA evidence showed that the blood could have come from the offspring of the parents tested," Baird says.

The suspect was subsequently tried for murder, but was acquitted. Obtaining a murder conviction without a body is generally difficult. The defense can always argue that the victim is still alive, or at least was alive when the suspect last saw him, even though there was some bloodshed. The missing man in this case is not still alive, however. His skeleton was found about a month ago—and once a suspect has been acquitted in a jury trial, he cannot be retried for the same crime.

As this case illustrates, the various methods of DNA typing can be used for paternity testing and for identifying the victims, as well as the perpetrators, of crimes.

Although Cellmark and Lifecodes' methods for obtaining DNA prints have the capability of identifying an individual with absolute certainty, they have a disadvantage in that a DNA sample has to be in relatively good condition to be analyzed. The DNA fragments identified by the probes range in size from 2 to 20 kilobases, but DNA breaks down very readily if it is not stored properly.



Lifecodes Corporation

Identifying a rape suspect. *The suspect's DNA produced a banding pattern identical to that of the DNA of semen obtained from a rape victim. One probe was used here, but three or four would be used to nail down an identification.*

Here the method used by Forensic Science Associates, which was developed by the Cetus Corporation of Emeryville, California, has the edge. Not only is it applicable to broken-down DNA, say 100 to 200 base pairs in length, but it also works with vanishingly small samples. This procedure uses a gene amplification method, known as the polymerase chain reaction (PCR), to pick out a specific DNA sequence in the genome and then multiply the number of copies of that sequence to produce sufficient material for analysis. (*Science*, 10 June, p. 1408).

Recently, Russell Higuchi and Henry Erlich of Cetus, in collaboration with Cecilia Beroldingen and George Sensabaugh of the University of California, Berkeley, have shown that a single cut hair can provide sufficient DNA for PCR amplification and subsequent analysis. "There is so little DNA inside a hair that we can't measure it, but we can amplify it," Erlich says.

The DNA inside hair shafts is not of nuclear origin but comes from the mitochondria, the cell's energy powerhouses, which have small genomes of their own. Analysis of mitochondrial DNA cannot yet provide sufficient information for a positive identification of a crime suspect.

More useful right now for forensic analysis is the genomic DNA in the hair follicle cells that cling to the roots of hairs that have fallen, or been pulled, out. The nuclear DNA contains the genes for the major histocompatibility proteins, which have sufficient individual variability to be useful for identification purposes, especially if more than one is typed. Erlich and his colleagues have shown that the typing of histocompatibility genes can be achieved with the DNA in the cells on a single hair root, after the DNA is amplified by the PCR.

In the past, when forensic scientists wanted to match hair found at a crime scene with that of a suspect, they could only look at such superficial characteristics as color and structure. But DNA typing goes directly to an individual's most characteristic feature, his genetic material.

The Cetus group's method does not provide as a high a degree of certainty in an identification as RFLP analysis does, however. According to Edward Blake of Forensic Science Associates, use of several probes will eventually produce gene patterns that might be restricted to 1 of 10,000 to 100,000 people, rather than to 1 of 10 billion. The PCR-based method can be used on samples that could not be analyzed by the other procedures, however.

A somewhat bizarre murder case in which Blake testified provides a case in point. A very old man had apparently died of starvation in a rest home in Pennsylvania, and the operators of the home, Helen and Walter Pestinikas, had been charged with negligent homicide. When the defense attorney questioned the credentials of the autopsy physician who had concluded that the man had starved to death the State ordered a second autopsy. The man's body was exhumed, but the original autopsy physician indicated that the remains might have been tampered with.

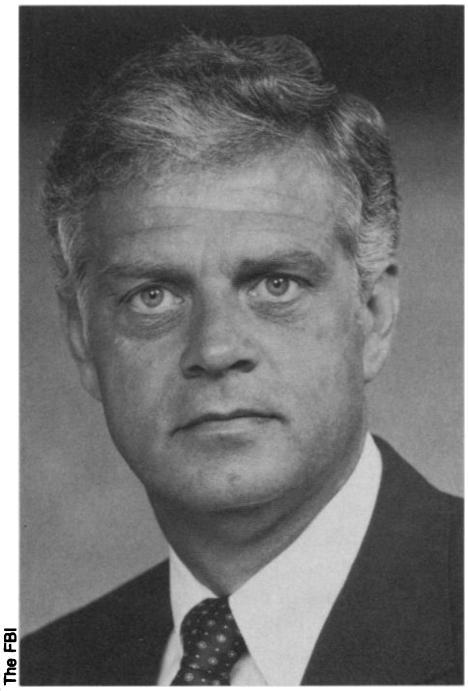
The body had been in the physical custody of the defendants after the autopsy, and there were suspicions that they had switched the internal organs with those of another body in an attempt to hide the man's cause of death. Blake's group was asked to find out if the organs found with the exhumed body actually belonged to it. At this point the tissues had been in contact with embalming fluid and in the ground for about a year. They were not in good shape. "All the DNA was very degraded," Blake says. "The average size was about 100 base pairs."

Nevertheless, the Cetus workers were able to amplify and compare an 82-base-pair fragment of a histocompatibility protein gene from both the internal organs and from other body tissues. "The bottom line was that the [histocompatibility] type of all the tissues was the same. The finding tended to undermine the idea that the tissues had been switched," Blake explains. Ultimately the defendants were convicted of negligent homicide, but acquitted of the charge of tampering with the body.

The FBI is currently setting up its own program of forensic DNA analysis, which it plans to have in place by October. The agency also wants, Hicks says, to bring some degree of uniformity to DNA testing nationwide. This would facilitate the establishment of a DNA data bank and the comparison of DNA samples from persons and crime scenes in different jurisdictions. "To do that," Hicks notes, "you would have to use the same DNA probes."

A uniform computerized method of storing and retrieving the information on the DNAs would also be needed. States may vary in the computer systems that they use for storing information on ordinary fingerprints, and this can complicate comparisons.

Researchers at the FBI's Forensic Science and Research Training Center in Quantico are evaluating the various DNA-typing methods to see which might be most suitable for a nationwide program. The agency appears to be leaning toward typing with VNTR probes, partly because the banding patterns obtained with them are easy to interpret.



The FBI

The FBI's John Hicks *estimates that forensic DNA typing will be widespread in 3 years.*

Even if forensic DNA typing is largely taken over by the FBI and other publicly supported crime laboratories, the commercial companies could still have a lucrative business in supplying the probes and other reagents needed for the tests. There are some 300 state and local crime laboratories in the United States.

Another goal of the FBI is to establish more fully the validity of the DNA-typing technology. The concern is not about DNA analysis per se, since the various methods are already well established in the research laboratory, but about their application to the types of biological samples likely to be encountered in forensic laboratories. "We don't want to have a system that may possibly be flawed," says Bruce Budowle of the Quantico laboratory. "There are going to be evidentiary hearings. The burden of proof is on the people who use [a new technology]."

In most states, scientific evidence must meet the "Frye rule," which holds that "the thing from which the deduction is made must be sufficiently established to have gained general acceptance in the particular field in which it belongs." Defining what is "generally accepted" can be difficult. Electrophoresis is certainly a generally accepted method of analyzing mixtures of proteins. Nevertheless, the forensic application of the technique to the analysis of blood and semen samples encountered problems before becoming acceptable in court. At one time, both the Michigan and California state supreme courts ruled inadmissible the results of the electrophoretic analysis of blood or semen stains on the grounds that the prosecutions failed to establish that the analytical methods enjoy general scientific acceptance.

The courts also indicated that the validity of a technology needs to be established, not just for the relatively ideal samples encountered in the research laboratory, but also for the less than ideal samples often encountered in forensic medicine. These may be hours or days old, contaminated with bacteria or fungi, dried out, or exposed to hostile environmental conditions such as heat, humidity, and sunlight. The FBI's validation of the DNA-typing methods will assess how these environmental influences might affect the outcome of the analyses.

Once DNA typing becomes widespread, data banks of DNA patterns will begin to accumulate. A good many questions remain about the operation of such data banks. One question concerns where to deposit the information. The FBI's National Crime Information Center is a possibility.

Another concerns whose DNA patterns should be included, although individuals convicted of sex crimes would be the logical first choice. Even before DNA typing became possible, the state of California required the collection of blood and saliva samples from sex criminals before their release from prison.

Members of the U.S. Armed Services and employees of the federal government are routinely fingerprinted, but DNA typing raises issues not raised by ordinary fingerprinting, Sensabaugh says. Fingerprints are used only for identification, but DNA contains additional information, about parentage, for example, or genetic predisposition to disease. "I may not want to give up my DNA," Sensabaugh says, "unless I know there are constraints on its use."

Routine forensic use of DNA typing, is also likely, Sensabaugh notes, to change the nature of the defense in cases in which identity is an issue. A test that can identify a rape suspect with absolute certainty makes it hard for the suspect to argue that someone else committed the crime. In addition, plea bargaining may become more common. "Faced with evidence that appears to be overwhelming, the defense counsel may decide that it will be better to cop a plea," Sensabaugh explains.

Of course, DNA typing can exonerate a suspect as well as incriminate him (or her). All three companies report cases in which DNA analyses showed that a suspect's DNA did not match that from a blood or semen sample. "It's important to recognize," Blake asserts, "that this kind of evidence is a defendant's best friend if he is falsely accused." ■ JEAN L. MARX