BIOPHYSICS

Double Helix Does Chemistry At a Distance—But How?

It's hard to be surprised anymore by DNA's repertoire of talents. It is a genetic archive with a remarkable combination of security and accessibility, a powerful probe that can seek out and bind to matching DNA molecules, even a potential computer. Now add yet another startling ability to the DNA résumé. In a paper in this issue of *Science* (p. 1465), chemists at the California Institute of Technology (Caltech) led by Jackie Barton present evidence that the DNA double helix can perform what they call chemistry at a distance. A DNA molecule with a chemical group artificially tethered to one end appears to mediate a chemical change far down the helix, causing a patch of damaged DNA to be mended.

The DNA damage repaired in the experiment—a small kink in the helix known as a thymine dimer—is the kind of damage caused by the sun's ultraviolet rays, and it can be a first step toward the deadly skin cancer melanoma. While the chemical groups Barton and her colleagues used for their demonstration aren't found in the body, long-range DNA repair of some kind might play a role in normal cells, and Barton thinks the finding might point the way to therapies that could patch up damaged DNA after severe sun exposure.

The result may point to an even more impressive attribute of DNA—if it means what Barton thinks it does. The ability of DNA to carry out long-range repair launches this paper into the heart of an already heated controversy over the possibility that DNA's unique structure allows it to behave like a conductive wire, utterly unlike the insulating behavior of proteins. The paper is the latest of four from Barton and her colleagues supporting the proposition that electrons can flow freely through the channel that runs down the center of the joined bases of the helix—in this case, traveling from the thymine dimers to the added chemical groups and repairing the dimers in the process. "There is no question that these results are saying DNA is a different system than proteins," says Barton.

If Barton is right and DNA readily transports electrons, the implications could go well beyond DNA repair. In living things, the transfer of electrons in DNA plays a crucial role in DNA regulation and other biological processes. And the technological possibilities are alluring as well: Knowing the precise electrical properties of DNA, says Georgia State University chemist Tom Netzel, could allow chemists to tailor artificial DNA molecules to serve as sensitive biological probes and minute photochemical machines.

But Barton faces some determined skeptics. By taking a variety of different experimental tacks, her group has finally proved its case to the satisfaction of some colleagues. Columbia University's Nick Turro, for instance, who collaborated on the first of Barton's papers, says the four experiments taken together "show unambiguously that there's long-range chemistry that can be performed on DNA, and that electron transfer can be accomplished." Stanford University biologist Philip Hanawalt, a leader in the study of DNA damage and repair, calls Barton's latest work "convincing." Others, however, see loopholes in each of the earlier papers—interpretations equally consistent with the data that do not require a paradigm shift. As University of Pittsburgh theoretical chemist David Beratan puts it, "It's a mystery story. You have to decide what data are convincing and try to piece together a coherent story."

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> —Tom Netzel

Easy as π. At issue is exactly how electrons move through large organic molecules. Twenty-five years of study have convinced chemists that in proteins, electrons move only by the laborious process of quantum-mechanical tunneling through pathways that connect one atom to the next along the protein's backbone. Researchers have suspected that DNA might be different. They have pointed out that the arrangement of bases on the complementary strands allows the electrons shared by multiple atoms to inhabit donut-shaped electron clouds above and below each ring of bases. The interior of the helix can be thought of as a stack of these π orbitals. If electrons could be injected into this stack, so the theory goes, they might easily tunnel from one end of the DNA to the other. While this would still be a quantum-mechanical effect, the electron transfer would be as effortless as moving current through a wire.

But the π-stack conductivity theory has always been a minority opinion. DNA simply does not fit the expected criteria for conductors, says Beratan: "What we know about it from basic physical chemistry doesn't make it look like a wire." Even biology has argued against the theory. If electrons could scoot around DNA with such facility, says Beratan simply, "we'd all be in a lot more trouble when we walk out in sunlight."

In 1993, however, Barton, Turro, and their colleagues in effect hooked a DNA strand up to a circuit, tested its conductivity, and came up with evidence that seemed inconsistent with the theory of DNA as a resistor (*Science*, 12 November 1993, p. 1025). They had created metal complexes that slipped between adjacent base pairs in the DNA. One arm of the complex would stick into the core of the DNA helix, or intercalate, "like one blade of a propeller," says Barton, injecting an electron into the core or retrieving one, depending on the complex.

The two chemists attached an electron-donating ruthenium complex near one end of a 15–base pair synthetic DNA helix and an electron-accepting rhodium complex near the other end. When hit by photons, the ruthenium would be excited and begin to glow until it could transfer an electron. If no rhodium acceptor was attached to the other end of the DNA, the ruthenium continued to glow. But if a rhodium-acceptor complex was in place, says Barton, "the glow was quenched because of the presence of electron transfer."

Indeed, Barton and Turro saw no detectable glow at all, which they interpreted as evidence that the DNA shuttled electrons between the metal compounds so fast that the quenching happened before it could be measured. The implication, they said, was that electrons could move huge distances through the DNA at speeds a million times faster than would be possible if the electrons had to tunnel laboriously from atom to atom, as they do in proteins.

The result shook up the field. As Beratan says, "I don't even have to do much theory to tell you the Barton '93 result is extremely provocative." Chemists were skeptical, and they were especially troubled by the lack of any glow from the ruthenium, says Tony Harriman, a spectroscopist at the University Louis Pasteur in Strasbourg, France. "[Barton and Turro] took a very, very negative result and converted it into an extremely positive conclusion. Many people would interpret not seeing the luminescence as failure of the experiment. To interpret it in a very spectacular way and a very positive way is going to raise a few eyebrows."

Indeed, Harriman promptly set off to do an experiment using organic molecules as donors and acceptors, spaced at random dis-

tances on the DNA. He was able to see his complexes luminesce and could measure the rate at which the glow was quenched, which indicated electron transfer rates "a little faster" than would be expected from a protein, but consistent with the mundane DNA-as-resistor theory. And Caltech chemist Tom Meade tethered donor and acceptor metal complexes to opposite ends of an 8–base pair DNA helix and found a similar, modest electron-transfer rate.

Barton agrees that "there was no question that [the 1993 finding] was a surprising result and required lots of controls." To show that the lack of luminescence in her experiments really was due to electron transfer and to measure its rate, she enlisted University of Minnesota chemist Paul Barbara, an expert in ultrafast spectroscopy. These experiments required concentrations of complexes too high for them to be tethered a fixed distance apart on the DNA helix. Instead, the two chemists simply mixed them with the DNA and allowed them to intercalate randomly along the helix, presumably some distance apart. The disappearance of the glow, as indicated by the spectroscopy, turned out to be as fast as Barton and Turro had reported in 1993, providing further evidence that the reaction was driven by rapid electron transfer down the helix (*Science*, 26 July 1996, p. 475).

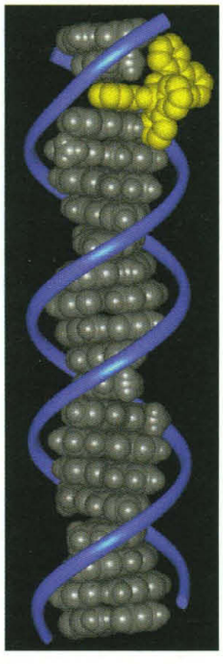
This result came with a caveat of its own, however. Because the metal groups weren't tethered to the DNA, as Barton explains, the distance the electrons had to travel could not be established. It was conceivable that the metal complexes were clustering together, in which case adjacent molecules would be swapping electrons over a very short distance. "Without tethering the complexes to the DNA," says Barton, "we couldn't rule clustering out." They did consider it unlikely, she adds.

But Barbara reports in the 16 January issue of the *Journal of Physical Chemistry B* that he has now reanalyzed the data from his experiment with Barton and found that the "data are completely consistent with clustering combined with short-range electron transfer." In the 16 February issue of the *Journal of the American Chemical Society*, Bengt Nordén and his colleagues at the Chalmers Institute of Technology in Sweden come to the same conclusion from a similar experiment. Clustering, he says, is "a much more plausible explanation" than conductive DNA.

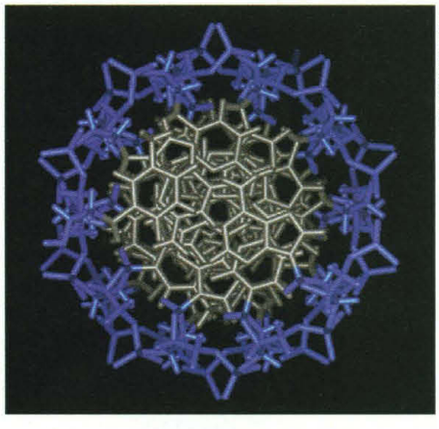
A charged issue. For Barton, the experience just underscored the need to lock the metal complexes onto the DNA. Her latest experiments rely on a chemical change in DNA, driven by charge transfer to a distant chemical group, to make a case for the electrical conductivity of the π stack. In the 22 August 1996 issue of *Nature*, she and her colleagues describe an experiment in which a metal complex, excited by a photon, stole an electron from a pair of guanine bases at a distant site on the DNA. The result was what is known as oxidative damage to the DNA, apparently triggered by the electron transfer down the double helix.

The ratio of reactions to photons going into the system—the so-called quantum yield—was extremely low, however: just one in 10 million. That seemed to leave room for alternative explanations. "Maybe [the reaction happens] the one time the duplex opens up and something strange happens," says Netzel. "If we were dealing with a quantum yield of 30%, we could be pretty sure we're dealing with a phenomenon with an intact helix. If we're dealing with 10^{-7} quantum yield, the room for nature to be fooling us is much greater."

In this week's *Science* paper, Barton presents a long-range chemistry experiment that she says isn't open to this objection, and now she's winning some converts. Barton and her colleagues fabricated DNA helices with a built-in thymine dimer, then intercalated an electron-accepting metal complex at the end of the DNA. Exposing the sample to light excites the rhodium compound, triggering it to absorb an electron from the thymine dimer and repair the DNA damage. "Even California sunlight works just fine," says Barton. And because the rhodium complex can catalyze the repair reaction over and over, she says that the experiment "may represent a strategy to rationally design molecules that can accomplish this kind of repair therapeutically."



Wired? In DNA's core, base pairs form a so-called π stack (gray, shown in top and side views) along which electrons may tunnel from a distant site to an artificial electron acceptor (yellow).



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It also constitutes the first systematic measurement of how electron transfer in DNA changes with distance, says Barton. In a resistor, such as a protein, the rate or efficiency of electron transfer falls off very quickly with distance. In contrast, Barton and her colleagues found that the repair efficiency didn't change with the distance between the metal-acceptor complex and the thymine dimer. But specially fabricated disruptions in the base-pair stack did cut into the efficiency. "The bottom line," she says, "is we were carrying out a long-range electron-transfer reaction that depended on the π stack."

Barton says that because the reaction was able to repair every one of the damaged strands, there is no room to argue that what is happening is a fluke that depends on some rare change in the double helix. And Claude Helene, a biophysical chemist at CNRS in Paris, says the data are convincing, and they "open up the possibility that people will be searching for [evidence of chemistry at a distance in DNA] in living organisms."

But even after this latest experiment, just how each electron makes its way down the double helix is still an open question. "We don't yet understand the mechanism," says Barton. Instead of tunneling from the thymine dimer to the rhodium in one step, she says, "maybe it's hopping" down the helix. If the energies of the electron orbitals and that of the electron accepted by the intercalator are close enough, then the electron might easily tunnel from base to base down the π stack, virtually unaffected by distance.

This mechanism would not work in proteins, where the gap between the energies of the orbitals and that of artificial electron donors or acceptors would be too large. And Barton's critics say it can't explain the results reported in her 1993 and 1996 *Science* papers. But it is consistent with prevailing theory, says University of North Carolina chemist Holden Thorp: "This may really be chemistry at a distance, with a believable mechanism. And there's a lot of cool stuff she could do with that."

What's clear to everyone is that the field needs more data to shake out the reality. Perhaps half a dozen labs, Barton's among them, have experiments or papers in the works that might pin down DNA's electrical properties and what their mechanism might be. The double helix is not accepting its new accolades easily. "The burden of proof for such a startling result," says Netzel, "is simply higher than for a boring result."

–Gary Taubes