A New Role For Gases: Neurotransmission

The remarkable finding that nitric oxide carries nerve impulses initiates a novel concept of neurotransmission

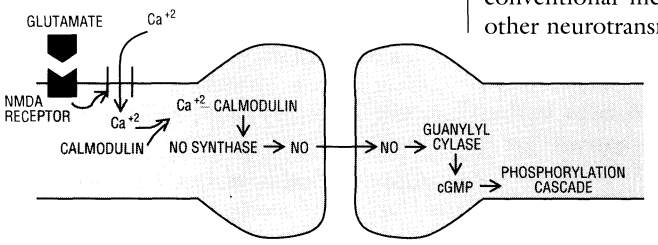
HUMAN BIOLOGY IS DOMINATED BY COMplex molecules such as proteins and nucleic acids, and no one would have predicted a role for something as disarmingly simple as nitric oxide. So back in the early 1980s when pharmacologists first began suggesting that nitric oxide, which is a gas, might be an important regulator of the body's activities, their proposals were greeted with skepticism. But expectations aside, nitric oxide is turning out to be one of the most pervasive regulators of human physiology yet described.

It helps maintain normal blood pressure by dilating blood vessel walls, and has lately been implicated in several additional—and critical—activities: Immune-cell killing of foreign organisms, the normal contractions of the gastrointestinal tract, and penile erection. Now, neurobiologists Solomon Snyder, David Bredt, and their colleagues at Johns Hopkins University School of Medicine have evidence for yet another unexpected role for nitric oxide—this time in the brain.

According to recent work by the Snyder group, some of which is not yet published, nitric oxide in low concentrations acts as a neurotransmitter, carrying nerve impulses from one cell to another. It may be, Snyder says, the first member of a new neurotransmitter family. Neuroendocrinologist Karl Knigge of the University of Rochester Medical School calls the work one of the "hottest, most exciting things in neurobiology. Now a new kind of transmitter turns out to be a gas. Good God!"

But while nitric oxide is a normal neurotransmitter, the work of Snyder's group also shows that when nitric oxide is produced in abnormally high concentrations, it can be toxic to neurons. Indeed, it may act as an agent of cell death in stroke and neurodegenerative disorders such as Huntington's and Alzheimer's disease (also see *Science*, 7 June, p. 1380).

Because of its possible importance in brain pathology, as well as in normal brain activities, neurobiologists would like to know a great deal about what nitric oxide does, and the Snyder group has just taken a step that could help answer that question. One of the problems that has impeded nitric oxide research in the past is its short lifespan; it lasts for only seconds after its manufacture. But



It's a gas. *The path by which nitric oxide is produced and has its effects in nerve cells.*

in the 27 June issue of *Nature*, the Johns Hopkins workers describe the cloning of the gene encoding nitric oxide synthase, the enzyme that makes nitric oxide in nerve cells and also in the endothelial cells that line blood vessels.

Having the gene sequence may also help to erode any remaining pockets of resistance to the idea that nitric oxide has biological functions. "People used to say, 'You need thunder and lightning to make nitric oxide in the atmosphere, so how does the body manage to do it?'" recalls John Vane of the William Harvey Research Institute in London, who studies nitric oxide action in endothelial cells. Now, he adds, it will be possible to explore exactly how the body does do it.

What's more, the cloning of the gene may make it easier to develop improved diagnostic tests and therapies for diseases that may result from abnormalities in nitric oxide synthase, which include high blood pressure as well as the neurodegenerative conditions mentioned previously.

Snyder says he got interested in nitric oxide a few years ago after Salvador Moncada of Wellcome Research Laboratories in Berkenham, England, had identified it as the mysterious "endothelial relaxing factor," which is released by endothelial cells and causes blood vessels to dilate. Snyder decided to take a look at nitric oxide in the brain, but assaying for it directly would have been hopeless, he says, because of its instability. So instead he and his colleagues looked for the synthase enzyme.

The strategy paid off when they showed that, within the brain, the synthase occurs exclusively in neurons. Then the researchers went on to show that nitric oxide fulfills all the classical criteria for a neurotransmitter. It is made in nerve cells, for example, and inhibiting its synthesis can block nerve stimulation. "The evidence," he says, "is better than the evidence for the well-established transmitters acetylcholine and norepinephrine. But it [nitric oxide] is a bizarre neurotransmitter."

Unlike other neurotransmitters, which are made in advance and stored in small vesicles until needed, nitric oxide is made and used immediately. Nor does it act through a conventional membrane receptor the way other neurotransmitters do. Instead, Snyder says, nitric oxide's "receptor" is the iron bound to enzymes, including guanylyl cyclase, which synthesizes cyclic GMP, an important regulator of cellular activities.

What Snyder is proposing, based on results from both his lab and others, including those of Moncada, James Ferrendelli of Washington University in St. Louis, and Ferid Murad of Stanford University, is that stimulation of nerve cells by the excitatory neurotransmitter glutamate causes calcium ions to move into cells. Then the calcium ions, working in conjunction with the regulatory protein calmodulin, turn on the synthesis of the nitric oxide, which in turn diffuses to adjacent cells where it activates guanylyl cyclase, resulting ultimately in the appropriate physiological response.

But trouble develops if glutamate stimulation is excessive. A great deal of evidence now indicates that this neurotransmitter can have neurotoxic effects if present in higher than normal amounts, and the Snyder group's results show that glutamate-related cell death is mediated by nitric oxide. That might cause the loss of brain neurons in Huntington's and Alzheimer's, Snyder proposes.

He now plans to determine whether the nitric oxide synthase from patients with the neurodegenerative diseases behaves differently from the enzyme of normal controls. That might provide clues to why some people get the diseases and others don't, as well as providing information that might be useful for diagnosis or therapy.

Finally, the Johns Hopkins group has found that the sequence of nitric oxide synthase resembles that of another enzyme, called cytochrome P450 reductase, which is best known as a liver enzyme involved in drug metabolism. But Snyder and his colleagues have found that the enzyme also occurs in brain neurons, and specifically in cells that do not make nitric oxide synthase. What could that mean? "Maybe there are more bizarre gas-like transmitter molecules in the brain," Snyder speculates. "It's exciting to us. It's a completely new concept of neurotransmission." ■ MICHELLE HOFFMAN