BRAIN ATTACK

Progress is slow in finding better ischemic-stroke therapies

BY ANDREAS VON BUBNOFF

About a year ago, E. Gail Anderson Holness was in church when she suddenly felt lightheaded. At first, the then-49-year-old minister, motivational speaker, and Washington, D.C.–based writer didn't think that the episode was serious, even though she'd had headaches for several days. She was healthy, after all, and she ran 4 miles almost every day.

"Because I am an athlete, I could not understand what was happening to me," she says. "I said, 'Just let me go home.'" But when her speech became slurred and the left side of her face started to droop, a friend convinced her to go to the hospital.

Anderson Holness had had an ischemic stroke, the type caused by a blocked blood vessel in the brain. The stroke killed some of her brain tissue by cutting it off from its oxygen and nutrient supplies.

About 600,000 people suffer ischemic strokes each year in the United States. The other kind of stroke, which occurs when blood spills into the brain, affects 100,000 people a year. With more than a million survivors facing varying degrees of permanent damage, strokes are the leading cause of serious, long-term disability in the country.

Still, there is currently only one drug treatment approved by the Food and Drug Administration for use at the time an ischemic stroke occurs. That drug is tissue plasminogen activator (tPA), given intravenously to dissolve blood clots. FDA approved the famous "clot buster" in 1996.

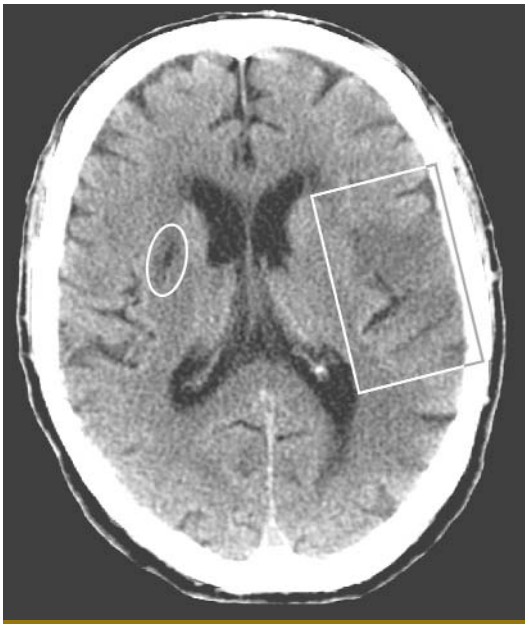
Treatment with tPA saves many patients—as it did Anderson Holness—by opening obstructed blood vessels in their brains. But the drug has severe limitations. Doctors have to first do a computerized tomography scan to make sure that the person isn't experiencing a bleeding stroke, which tPA would make worse. And the clot-busting drug must be administered within 3 hours of the onset of a stroke—which isn't always possible because many delays occur on the way to emergency treatment. After 3 hours, the risk of bleeding outweighs the potential benefit of tPA, at least according to current medical judgment. The potential complications have made emergency room physicians reluctant to give tPA, says Costantino Iadecola, chief of the division of neurobiology at Weill Cornell Medical College in New York City.

As a result, less than 5 percent of ischemic-stroke patients in the United States get treated with tPA. "Very few patients can benefit from the only treatment we have available for stroke," Iadecola says. "There is a tremendous interest to develop new treatments."

Indeed, trials of many treatments are under way. Some test alternative clot busters that may be less likely than tPA to provoke bleeding. Other studies focus on drugs that keep brain cells from dying once a stroke has occurred. Some researchers are using genetics to look for yet more such neuroprotective compounds to test.

Other scientists are trying combinations of drugs because each drug typically targets just one or a few of the many processes that lead to brain-cell death after a stroke.

All in all, researchers have come up with a number of approaches that have shown promise in animal studies. Even so, making such treatments work in people has proved to be a stubborn problem.



SHADES OF DANGER — Darker tissue in circled area at right in this computerized tomography brain scan was damaged by an ischemic stroke that occurred about 6 hours earlier. Smaller dark area, circled at left, represents damage from an older stroke.

UNIV. TEXAS-HOUSTON STROKE TEAM

BUSTING OUT Given that clot busting is the only approved and effective treatment for ischemic stroke, some researchers are trying to expand on it. For example, Anand Vaishnav of the University of Kentucky Medical Center in Lexington and his stroke team advise doctors at rural hospitals on whether tPA will help or hurt a particular stroke patient.

Many doctors are insufficiently trained or too conservative to use the drug, Vaishnav explains. "We'll give them the green signal to give tPA" when it's appropriate, he says. After a preliminary study, Vaishnav concludes that a national telemedicine system, which would provide instant expert advice to any location, could increase the proportion of ischemic-stroke patients getting tPA to 10 or even 20 percent.

Other doctors are starting to extend the time following a stroke during which tPA can be used, using brain imaging to show that a person still has salvageable tissue in the damaged area. Last year, a team led by researchers at Stanford University published a study suggesting that such patients can benefit from tPA treatment up to 6 hours after a stroke.

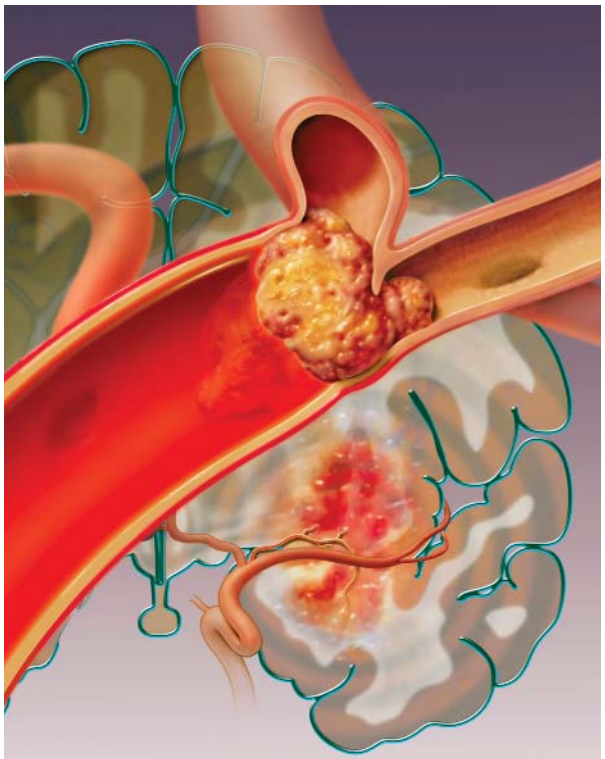
Still others are using ultrasound to make tPA more effective. In a small clinical study published in 2004, Andrei Alexandrov, now at the University of Alabama at Birmingham, found that applying ultrasound through the skull triples the effectiveness of tPA in dissolving ischemic-stroke clots. The sound waves agitate

the stagnant blood in the blood vessels affected by the stroke, Alexandrov says, allowing tPA to better reach and penetrate the blood clot. "It's like a spoon that stirs sugar in a cup of tea," he says.

More recently, Alexandrov has been working with ImaRx Therapeutics of Tucson, Ariz., which makes tiny, gas-filled bubbles by enclosing minute quantities of perfluoropropane in fatty shells that are similar to cell membranes in humans. Administered intravenously, the bubbles aren't dangerous, he says, because perfluoropropane is chemically inert and present in only small amounts. But when the bubbles reach the site of stroke damage, ultrasound vibration makes them oscillate or even explode. This further agitates the stagnant blood around a clot, Alexandrov says. In February, he told the International Stroke Conference in San Francisco of early success combining tPA, ultrasound, and gas bubbles to dissolve clots in stroke patients.

Until recently, many doctors had high hopes that a drug called desmoteplase, derived from the anticlotting saliva of vampire bats, would prove to be an effective clot-busting alternative to tPA. Last year, a study funded by the biopharmaceutical company PAION, based in Aachen, Germany, reported in *Stroke* that desmoteplase appeared to clear brain blood clots in patients up to 9 hours after a stroke, without causing excessive bleeding.

In late May of this year, however, PAION announced that the drug didn't show significant benefits in a larger trial. Success might have positioned desmoteplase for FDA approval within a few years, says Marc Fisher of the University of Massachusetts Medical School in Worcester. "It's very disappointing that it didn't show anything," Fisher says. "That was the only promising thing. Now there is nothing."



VICIOUS BLOCK — Foreground illustration shows a blood clot lodged in a cerebral artery, the location of which is diagrammed in the brain at rear. Such an event, an ischemic stroke, starves tissue of oxygen and kills brain cells unless treatment can quickly clear the blockage.

MEDISCAN/CORBIS

FOR PROTECTION Since therapies that restore blood flow in the brain have limitations, scientists have for decades also searched for means to protect neural cells while they're starved of blood and oxygen. Ideal neuroprotective drugs would interfere with the stroke-related events that cause those cells to die.

By now, researchers have a pretty complete understanding of these events, Iadecola says. As soon as blood flow is interrupted, nerve cells in the immediate area of the blockage start dying for lack of oxygen and energy-providing glucose. Most of the cells at the core of a stroke can't be rescued unless blood flow is reestablished within a few minutes.

The area around the core still has some blood flow and remains salvageable for hours, Iadecola says. But dying cells in the core make matters worse by releasing glutamate, a chemical that normally transmits nerve signals.

In this case, glutamate is dangerous because it triggers brain cells outside the stroke core to open critical pores to an influx of calcium ions. That starts a wave of inflammation, free radical production, and DNA and protein destruction that leads, about a day after the stroke, to the activation of a cell-suicide program called apoptosis.

Researchers have been looking for compounds that target these processes by soaking up free radicals, for example, inhibiting the effects of calcium and glutamate, or fighting inflammation. In March of last year, David Howells of the University of Melbourne in Australia and his colleagues reviewed in the *Annals of Neurology* about 1,000 animal studies and 100 human trials that have tested such compounds, as well as clot busters, since 1957.

But so far, none of the neuroprotective compounds that worked in animals or lab-cultured cells has benefited stroke patients, says Ashfaq Shuaib of the University of Alberta in Edmonton. "Everything was negative," he says. "The record is pretty bad."

Shuaib says that researchers tried to improve the situation by establishing quality criteria for testing stroke drugs in 1999. The Stroke Therapy Academic Industry Roundtable (STAIR) standards declared, for example, that a drug should be tested in animals in several labs, including ones other than that of the company developing the drug; that it should be tested in several animal species, including primates; and that the drug's physical and behavioral effects should be monitored for weeks after an animal undergoes an induced stroke.

One current study that follows most of the STAIR criteria is testing infusions of magnesium sulfate. Magnesium can keep calcium from rushing into cells. The drug can be given even before a patient reaches the hospital because it carries no known risk of increased bleeding. A team at the University of California, Los Angeles School of Medicine is studying the effects of magnesium sulfate given within 2 hours after a stroke. So far, 300 patients have received the infusions, says Anna Yanes, the chief nurse coordinator of the trial. Another 1,000 patients will be recruited, and indications of the long-term effects of the treatment should emerge in 2 to 4 years.

But the STAIR criteria don't seem to be enough. That became clear last October, when the drugmaker AstraZeneca announced that the first trial in people of a drug that satisfied all STAIR criteria had failed. The trial found no significant benefit to stroke patients from an antioxidant called NXY-059, which was intended to inhibit damage by free radicals. As a result, AstraZeneca abandoned its efforts to seek FDA approval for the drug.

"We were devastated, really disappointed," says Shuaib, who was the lead investigator of the trial.

Meanwhile, researchers are trying cutting-edge approaches to finding new neuroprotective-drug candidates. The drug company Wyeth, for example, is analyzing dying nerve cells to determine what genes and proteins are active in them. The company's scientists plan to screen libraries of chemicals in search of compounds that modulate these genes or proteins and so might keep cells from dying, says Giora Feuerstein, who heads Wyeth's department of discovery and translational medicine in Collegeville, Pa.

Seong-Seng Tan of the Howard Florey Institute for brain research in Melbourne, Australia, and his colleagues are also looking for proteins that help cells survive. Tan's group has identified about a dozen proteins that become more active in brain cells that survive experimental strokes in animals.

One of them is a mouse protein called BP5. It's 10 times as abun-

dant in brain cells that survive a stroke as in other cells in the same area. Further animal experiments showed that BP5 can keep nerve cells alive in strokelike conditions. Tan says that he hopes to find a drug that can activate or mimic BP5.

COMBO LESSONS The NXY-059 failure illustrates another reason why it has been so difficult to find neuroprotective drugs: Many candidate compounds affect only one or a few of the mechanisms that kill brain cells after a stroke, Shuaib says. "A single drug is not really the answer," he says.

So Shuaib and others have turned to treatment combinations. The Canadian researcher recently gave each of nine stroke patients six treatments normally given in isolation or in limited combinations. Within 3 hours of their strokes, the volunteers received tPA, and for 12 hours they wore helmets that cooled their brains by about 3°C. The cooling slows many processes that cause nerve cells to die.

The other treatments were an infusion of albumin to inhibit free radicals and improve blood flow; a statin drug, also for improving blood flow; an anti-inflammatory agent; and a day of magnesium infusions. Shuaib says the test established the safety of the overall therapy but that it wasn't designed to determine its effectiveness at protecting brain cells. The combination treatment has protected brain cells in animals, however.

James Grotta of the University of Texas in Houston combines cooling with an infusion of the equivalent of Irish coffee: caffeine and ethanol in a combination researchers call caffeinol. Caffeine, Grotta says, blocks dying cells' glutamate release, and ethanol blocks glutamate's action on surviving cells. Both compounds are cheap, their side effects are well-known, and they can easily enter the brain, he says.

In rats, a caffeinol infusion 2 to 3 hours after an induced stroke can reduce the brain area in which cells die by about 60 percent, compared with the area in animals receiving no treatment. If the regimen includes cooling the animals by 2°C, the area of dead cells is reduced by 80 percent. A person would have to get the equivalent of two cups of strong coffee and one cocktail to get a dose equivalent to what the animals received, Grotta says. He's currently testing the treatment's safety in people.

DON'T DELAY Stroke researchers seem to be a long way from any breakthrough in stroke treatment, with current progress measured in small increments. For instance, doctors physically removed blood clots from the brains of about 2,000 stroke patients last year in the United States using a device called the Merci (Mechanical Embolus Removal in Cerebral Ischemia) Retrieval System made by Concentric Medical in Mountain View, Calif. During this procedure, a catheter threaded from the groin into a brain artery is used to deploy a retriever that snags and pulls out the clot. Catheters inserted in this way can also deliver tPA directly to where it's needed.

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>
> — ASHFAQ SHUAIB, UNIVERSITY OF ALBERTA IN EDMONTON

The FDA has approved such devices for safety, but there's no solid evidence that any of them benefit stroke victims, says S. Claiborne Johnston, the director of the stroke service at the University of California, San Francisco. What's more, a Merci device can cause bleeding, he says.

With tPA still the only sure ischemic-stroke treatment available, doctors and researchers emphasize the importance of getting victims to the hospital quickly. That gives a person a chance to receive tPA and prevent the death of more and more brain cells over several hours. "Time is brain," Wyeth's Feuerstein says.

Anderson Holness, for her part, is glad to be out of a wheelchair—the stroke had paralyzed her left leg for a few weeks, but she's now back to running every day. She says the specialist told her, "It's a good thing they didn't take you home. You would not have survived." ■