**Story 1**

November 27, 1997

Tests on Mice Block a Defense by Cancer

By NICHOLAS WADE

Many drugs work well at first against cancer, but the tumor cells quickly develop resistance. A concept for getting around the resistance problem has now been proved, at least in laboratory mice, and may well prove relevant to the treatment of human cancer.

The idea is to attack the cells that build capillaries, a network of fine blood vessels that nurture fast-growing tumors. It exploits the fact that the capillary-making cells are normal, genetically stable cells and should therefore, unlike tumor cells, be incapable of developing resistance.

Researchers at Children's Hospital in Boston report that the capillary-making cells in mouse tumors did not develop resistance to repeated doses of a recently discovered agent that inhibits their activity. A second finding, both unexpected and unexplained, was that although the tumors at first grew back when the drug was withheld, they did not recur after two or more repetitions. The results are being published today in the journal Nature.

''For years, we thought you could hold tumors in check with angiogenesis inhibitors, but to actually cure them is amazing,'' said Dr. Noel Bouck, an expert on blood vessel formation, or angiogenesis, at Northwestern University Medical School in Chicago.

Dr. Douglas Hanahan, a cancer biologist at the University of California at San Francisco, said that ''the reality is not going to be as exciting as the response in these mice'' but that he expected angiogenesis inhibitors in time to become a useful component of cancer therapy.

The idea of retarding tumors by attacking their blood supply system is not new. The first generation of angiogenesis-inhibiting drugs is now undergoing clinical trials. But at least some of these drugs attack the tumor cells themselves, by aiming at the chemical signals with which they trick the vessel-making cells into building a blood supply for them. The tumor cells can switch to other chemical signals and thus become resistant.

In January, researchers from Dr. Judah Folkman's laboratory at Children's Hospital reported their discovery of an agent that singles out active capillary-making cells and shuts them down. The researchers, led by Dr. Michael O'Reilly, named the substance endostatin. Unlike the first generation of angiogenesis inhibitors, which merely retard the growth of tumors in experimental mice, endostatin makes tumors shrink to microscopic size, as does another agent named angiostatin. A speck of angiostatin was first distilled by Dr. O'Reilly in 1994 from about 21 quarts of mouse urine.

With endostatin in hand, the Boston team was able to aim specifically for the capillary-making cells and address the question of whether they would develop resistance. Using mice with a particularly aggressive kind of tumor, they injected endostatin in stop-and-start cycles, a pattern that favors the emergence of resistance to drugs.

At first, the tumors shrank when their blood supply was cut off and then they grew back when endostatin was stopped. As predicted, in cycle after cycle the capillary-making cells did not develop resistance to the agent. Around last Thanksgiving, Dr. O'Reilly noticed that the tumors were no longer growing back after endostatin was withdrawn. Tumor cells were still there but had reverted to a dormant cluster, too small to need its own blood supply, and had not grown back throughout the lifetimes of the mice.

Dr. Folkman reported these results at a recent meeting of cancer biologists in Frederick, Md. When he presented a graph showing the mouse tumors growing and regressing several times and then lapsing into dormancy, there was an audible gasp, Dr. Bouck said.

''It was really one of those magical moments in science,'' she said.

The ability of endostatin to make mouse tumors regress without developing resistance is ''unprecedented and could herald a new era of cancer treatment,'' Dr. Robert S. Kerbel wrote in a commentary in Nature, adding, however, that the era ''could be years away.''

It was Dr. Kerbel, a cancer biologist at the Sunnybrook Health Science Center in Toronto, who suggested six years ago that targeting the capillary-making cells should be the way around the problem of drug resistance. Dr. Kerbel said that neither he nor others could test the idea at the time because suitable agents to be directed at the capillary-making cells were not available.

The patent on both the mouse and human forms of angiostatin belongs to Children's Hospital. It has licensed them to Entremed, a small biotechnology company in Rockville, Md., which has been a major supporter of Dr. Folkman's research since 1994. Entremed has in turn licensed Bristol Myers-Squibb, a large pharmaceuticals company, to produce and market angiostatin. The company also has an option, not yet exercised, to develop endostatin.

''We are several years away from clinical trials,'' Dr. Folkman said, adding that at present his laboratory could barely produce enough endostatin for its own research needs.

Both the angiostatin and endostatin, isolated by Dr. O'Reilly and his colleagues in Dr. Folkman's laboratory, are from mice, but there is every reason to assume that the human versions have the same or similar properties. By one of nature's economies, both agents are part of larger protein molecules. Angiostatin is a fragment of a substance called plasminogen, involved in blood clotting, and endostatin is the tail end of a kind of collagen, a protein of connective tissue.

Both agents become active when cut loose from their parent protein. These fragments may be produced all the time to make sure the blood vessel system is firmly restrained, Dr. Hanahan said, or they may be generated just after processes that require new blood vessels, like wound healing.

Dr. Folkman said he did not yet know how or why the endostatin forced the mouse tumor cells into dormancy, nor is it known how endostatin acts on the capillary-making cells.

**Story 2**

May 3, 1998

HOPE IN THE LAB: A special report:

A Cautious Awe Greets Drugs That Eradicate Tumors in Mice

By GINA KOLATA

Within a year, if all goes well, the first cancer patient will be injected with two new drugs that can eradicate any type of cancer, with no obvious side effects and no drug resistance -- in mice.

Some cancer researchers say the drugs are the most exciting treatment that they have ever seen. But then they temper their enthusiasm with caution, noting that the history of cancer treatments is full of high expectations followed by dashed hopes when drugs with remarkable effects in animals are tested in people.

Still, the National Cancer Institute has made the drugs its top priority, said Dr. Richard D. Klausner, the director. Dr. Klausner called them ''the single most exciting thing on the horizon'' for the treatment of cancer.

''I am putting nothing on higher priority than getting this into clinical trials,'' Dr. Klausner said. The mouse studies are ''remarkable and wonderful,'' he said, and ''very compelling.'' But he pointed out that the studies were in mice and so, when it comes to humans, he said he wanted to emphasize ''the if's.''

The new drugs, angiostatin and endostatin, work by interfering with the blood supply tumors need. Given together, they make tumors disappear and not return.

Dr. James M. Pluda, who is directing the cancer institute's planned tests of the drugs in patients, said he and others at the institute were ''electrified'' when they heard the drug's discoverer deliver a lecture about the newest results. ''People were almost overwhelmed,'' Dr. Pluda said. ''The data were remarkable.''

Although the discovery of the drugs, and some of their effects, have been reported over the past few years, Dr. Pluda said that ''if people understood how many steps ahead'' the research was compared to what had been published, ''they'd be even more in awe.''

But Dr. Jerome Groopman, a cancer researcher at the Harvard Medical School, was wary. ''We are all driven by hope,'' Dr. Groopman said. ''But a sober scientist waits for the data.'' And until the drugs are given to humans, he said, the crucial data simply do not exist.

So far, the drugs are the only ones ever tested that can seemingly eradicate all tumors in mice, even gigantic ones, equivalent to a two-pound growth in a person. The best that other cancer drugs have done is slow the growth of these large tumors. Mice are the traditional test animals in cancer research.

But even the drugs' discoverer, Dr. Judah Folkman, a cancer researcher at Children's Hospital in Boston, is cautious about the drugs' promise. Until patients take them, he said, it is dangerous to make predictions. All he knows, Dr. Folkman said, is that ''if you have cancer and you are a mouse, we can take good care of you.''

Other scientists are not so restrained. ''Judah is going to cure cancer in two years,'' said Dr. James D. Watson, a Nobel laureate who directs the Cold Spring Harbor Laboratory, a cancer research center on Long Island. Dr. Watson said Dr. Folkman would be remembered along with scientists like Charles Darwin as someone who permanently altered civilization.

The long trail to the discovery of the new drugs began more than 30 years ago when Dr. Folkman became obsessed by what many saw as a quixotic notion: that cancers cannot grow beyond the size of a pinhead unless they have their own blood supply. If he could block a tumor's blood supply, he reasoned, the tumor should shrink to a minuscule size.

The first major break in the efforts came a decade ago when Dr. Folkman and his collaborators found drugs that did what he envisioned. He called them anti-angiogenesis drugs because they stopped the process of developing new blood vessels, or angiogenesis. They slow tumor growth in animals but do not eradicate the tumors. Early results in patients indicate that the drugs may slow human cancers. Dozens of companies are developing such drugs.

The results with these weaker drugs were ''a proof of principle,'' said Dr. Bart Chernow, a professor of medicine and dean for research and technology at the Johns Hopkins University School of Medicine. Dr. Chernow is a founder of Entremed, a company in Rockville, Md., that was formed to make and market angiostatin, endostatin and other weaker drugs that can slow cancer growth.

But the real breakthrough -- and the two new drugs -- came from Dr. Folkman's efforts to understand a peculiar phenomenon that has been known to cancer surgeons for 100 years: sometimes a patient will have a single tumor, with no evidence whatsoever of metastases, the satellite cancers that can pepper a patient's body. A doctor will remove the tumor and all will seem fine. But then, a few months later, a whole series of metastases will appear, grow, and kill the patient.

In 1989, Dr. Folkman proposed a reason for the effect, which he wrote on a large white board in a room where his laboratory group had its weekly seminars. Is it possible, he asked, that a tumor could be making both stimulators and inhibitors of blood vessel growth? If so, the inhibitors might travel through the bloodstream, squelching metastases. When the large tumor was removed, it would no longer be a source of inhibitors, allowing the tiny metastases to proliferate.

Dr. Folkman tried to get one of his doctoral or post-doctoral students to work on that idea. ''Each Friday, at our meeting,'' he said ''I would say, 'Here's a great experiment.' But no one wanted to work on it.'' It seemed too wild, Dr. Folkman said, too unlikely to result in findings that would end up in a scientific journal, a major goal of young scientists.

**Undertaking The Big Challenge**

Then, in 1991, a post-doctoral student, Dr. Michael O'Reilly, decided to take on the challenge. Dr. O'Reilly focused on a particularly deadly mouse cancer that grows to the equivalent of a two-pound tumor in a person.

As long as mice had the large tumor, they had no signs of metastases. But five days after the tumors were surgically removed, metastases invariably sprang up in the animals' lungs. Within 15 days, the animals would be dead, their lungs packed with large red tumors, like grapes.

Eventually, after arduous work in collaboration with chemists, Dr. O'Reilly discovered that the large tumors made a substance that stymied the growth of other tumors. This substance showed up in the animals' urine, but was present in such minute quantities that Dr. O'Reilly had to collect 10 quarts of mouse urine to obtain 30-thousandth of an ounce of the mysterious substance. It turned out to be a piece of a larger and very common protein, plasminogen, that the body uses in blood clotting. Dr. Folkman named the new substance angiostatin.

Apparently, cells can use the plasminogen gene for two purposes: they can use it at its full length to make plasminogen, or they can use just a piece of it and make angiostatin. Plasminogen does nothing to stop tumor growth. The question was, would angiostatin?

Dr. Folkman and Dr. O'Reilly discovered that angiostatin also appears, in minute quantities, in human blood. Using outdated human blood discarded by the Red Cross, they extracted enough angiostatin to treat mice. Then they began their experiment.

They had 20 mice with large tumors on their backs. The investigators removed the tumors and then injected half of the mice with angiostatin each day and the others with salt water, as a comparison.

After 15 days, the researchers killed the mice and cut them open. As more than a dozen scientists gathered around a table in the laboratory, Dr. O'Reilly opened the first mouse. It had huge tumors filling its lungs. Then Dr. Folkman checked a notebook to see what the animal had received: salt water. They looked at the next mouse. No tumors. Dr. Folkman checked to see the treatment: angiostatin. And so it went. All 10 of the mice that had been injected with angiostatin were free of cancer. All 10 of those that had been received salt water had huge new tumors.

**A Jubilant Celebration And a Second Discovery**

The room was buzzing, the scientists were grinning. Dr. Folkman said. Everyone in the room knew what the results meant, and they were elated. They responded, he said, like men at a football game. ''Everyone clapped O'Reilly on the back,'' Dr. Folkman said.

Then the researchers found a second protein fragment, secreted by tumors, that also squelches metastases, Dr. Folkman said. It was a piece of a different protein, collagen 18, that is in all blood vessels but by itself has no effect on cancer. They named the collagen fragment endostatin.

''It was even more potent than angiostatin,'' Dr. Folkman said. If he gave it to a mouse with a huge tumor, he said, the equivalent of one weighing a pound and a half in a human, endostatin would shrink the cancer down to a microscopic size.

Moreover, tumors never became resistant to endostatin, said Dr. Folkman, who added that he had given the drug to mice with large tumors and they had shrunk to almost nothing. He stopped the drug, he said, and the tumors grew back. Then he gave the drug continuously for the rest of the animals' lives. The tumors remained small and harmless and the animals remained healthy.

Dr. Robert S. Kerbel, a cancer researcher at Sunnybrook Health Science Center in Toronto, said he was not surprised that the cancers never became resistant to endostatin. Tumors become resistant to chemotherapy drugs, Dr. Kerbel said, because cancer cells constantly reshuffle their genetic information. The result, he said, is that the tumors spin off mutant cells that resist the drugs and, ultimately, the tumors grow back, invulnerable.

But, Dr. Kerbel said, angiostatin and endostatin do not act on tumors. Instead, they act on normal blood vessels that feed tumors. And normal cells, he said, do not reshuffle their genes and so do not develop drug resistance. That is why chemotherapy drugs continue to devastate normal cells -- causing bone marrow suppression, loss of hair, nausea and vomiting -- even when the cancer cells have grown impervious to their effects, Dr. Kerbel said.

**Drug Combination Knocks Out Tumors**

Then Dr. Folkman discovered that he could actually obliterate tumors in mice with his new drugs. He gave endostatin and angiostatin together, treating mice for 25 days. To his surprise, Dr. Folkman said, ''there was no tumor left -- we couldn't even find it with a microscope.'' The tumors, he said, ''were eradicated.''

And the drugs seem to have no side effects at any stage of life, at least in mice, something that Dr. Folkman said is hard for researchers to believe. But, he said, he had given mice up to four times the doses needed to eliminate cancer and could not find any adverse effects. These two human proteins may be, he said, ''exquisitely aimed -- we do not know why -- at cancer.''

In contrast, Dr. Folkman said, mice become very ill when they receive commonly used chemotherapy -- their hair falls out, they bleed, they refuse to eat.

For the past four years, Dr. Folkman said, he and his colleagues have found that all tumors responded to the drugs in the same way. Even leukemia, a blood tumor, responds, he said, because it turns out that leukemia needs to form new blood vessels in the bone marrow to grow. Leukemia tumors grow on these blood vessels, ''like berries on a bush,'' Dr. Folkman said, shedding cancer cells into the blood.

But Dr. Folkman is the first to urge caution in leaping to conclusions about what might happen when patients try the drugs. ''Going from mice to people is a big jump, with lots of failures,'' he said.

Hopes were high for chemotherapy drugs that worked well in mice but turned out to be less successful in people. Therapies that used the immune system to rid the body of cancer also worked in mice but were disappointing when they were tried in people. Gene therapy treats mouse cancer, but has had limited success in people.

From bitter experience, most cancer researchers have learned to be leery of what one called ''that four letter word'' -- cure.

Meanwhile, Entremed is working as fast as it can to produce angiostatin and endostatin for studies in humans. Dr. John Holaday, Entremed's president and chief executive, said his company was working with the Bristol-Myers Squibb Company to develop angiostatin and had not yet decided on a corporate partner to develop endostatin. The drugs are being made in genetically engineered yeast growing in 20-gallon vats.

Dr. Pluda, of the cancer institute, said the first patients to get the drugs would have cancers that were growing quickly and were essentially untreatable. The institute will start by giving the drugs separately by the end of the year, he said, then hopes to combine them.

Already, Dr. Folkman said, he gets hundreds of calls a day from cancer patients, pleading for the drugs.

Dr. Folkman, in an interview on Friday, said one call had come from an old friend from medical school with prostate cancer that had spread to his bones.

''He's terrified,'' Dr. Folkman said. But there were no strings Dr. Folkman could pull. He said he had to tell his friend what he told all the other callers: ''You can't get it because it isn't being made.''