NEUROSCIENCE

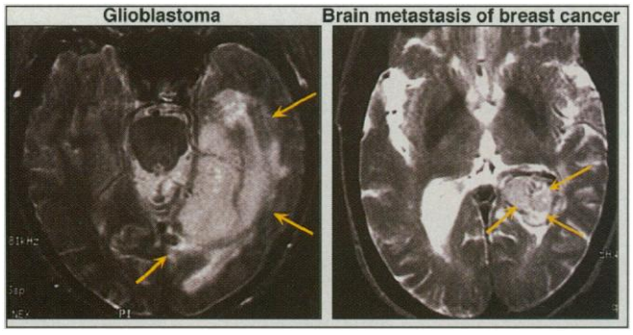
Molecules Give New Insights Into Deadliest Brain Cancers

NEW ORLEANS—Glioma is among the deadliest of tumors: Most of the 20,000 people diagnosed each year in the United States with this form of brain cancer die within 2 years. The tumors—which derive from support cells of the brain known as glia—are eerily invasive: Glioma cells spread freely into healthy surrounding brain tissue, making it impossible to remove all of the tumor surgically and virtually assuring that the cancer will soon return elsewhere in the brain. "We have so little to offer these patients," says Bruce Ransom, chair of neurology at the University of Washington Medical Center in Seattle. "This is an area of neuroscience that is long overdue for a significant breakthrough."

Ransom and other researchers fighting glioma would settle for some real progress on two fronts: clues to what makes glioma insidiously invasive, and ways to direct drugs specifically to the malignant cells. Findings presented last month at the meeting of the Society for Neuroscience here could offer both. Two research teams have identified different proteins—one an ion channel and the other a secreted protein that sticks to the matrix of molecules surrounding brain cells—that are produced by all the human gliomas tested, and apparently by no other cells in normal adult human brains.

Because they are so specific to glioma, the proteins could make good targets for directing antiglioma therapy right to the culprit cells. What's more, both teams, one led by Susan Hockfield of the Yale University School of Medicine and the other by Harald Sontheimer of The University of Alabama, have evidence that the proteins may help glioma cells invade surrounding brain tissue. The studies "both may be very important," says neuro-oncologist Henry Brem, co-chair of a multicenter consortium for testing brain-tumor therapies headquartered at The Johns Hopkins University School of Medicine. "Anything we can do to better understand and control [glioma's] invasiveness is a major advance."

Hockfield's team was initially looking for proteins important in brain development, not glioma. Postdoc Diane Jaworski was searching for a brain-specific member of a class of proteins that bind hyaluronan—a sugar component of the extracellular matrix, the interlocking web of molecules surrounding cells. Previous work had shown that hyaluronan-binding proteins help cells to move around in tissues during development. Jaworski hit pay dirt with a protein she called BEHAB, for brain-enriched hyaluronan binding. Hyaluronan-binding proteins have also been associated with invasive cancers, so Jaworski checked and found that BEHAB is made and secreted by human gliomas, but by no other cells in the adult human brain. What's more, it appears to contribute to the invasiveness of glioma cells: Rat glioma cell lines that form invasive tumors make high levels of BEHAB, as do surgical samples of highly invasive human gliomas that she checked. Rat glioma cell lines that form tumors without invasive properties don't make the protein.



SUSAN HOCKFIELD/YALE UNIVERSITY

Invasion. Cells fan out into healthy tissue from the border of a human glioma *(left)*. A breast cancer tumor that has metastasized to the brain shows no such invasiveness *(right)*.

BEHAB is normally cut in two as soon as it is secreted, and to see whether the protein's hyaluronan-binding portion could boost invasiveness, Hockfield's team put a gene encoding that fragment into a noninvasive rat glioma cell line. When the cell line was put back into rats' brains, expression of the BEHAB fragment "increased the invasive potential of these cells," says Hockfield. She notes, however, that the cells making the BEHAB fragment aren't quite as invasive as the most invasive rat cell lines or human gliomas. "My sense is we don't have all the elements there that an invasive cell needs."

Indeed, tissue invasion probably depends on multiple proteins, and Sontheimer's team has discovered another one that seems to play a role: an ion channel that allows chloride and other negatively charged ions to pass through the glioma cell membrane. They found the ion channel by chance while they were doing electrophysiological studies on glioma cells. "Then the story got really exciting," Sontheimer says. His group found no evidence of the channel in normal glial cells or normal brain tissue, but it was present in "every single [human glioma] we studied." What's more, in the higher grade, more invasive, gliomas, nearly 100% of the cells had the channel, while in the lower grade gliomas, the channel turned up in just under 50% of the cells.

Using a molecule from scorpion venom called chlorotoxin, which specifically blocks this new channel, Sontheimer's team probed the channel's role in the glioma cells' ability to slip through tiny spaces. They found that the chlorotoxin reduced the glioma cells' ability to penetrate a mesh of extracellular matrix proteins in a culture dish. Sontheimer speculates that for glioma cells to migrate straight through dense and healthy brain tissue, they must lose fluid so they can become "thin and spindly" and slip through small molecular openings. The chloride channels, he suggests, may help them lose salts, promoting water loss.

Ransom says proteins like these "must contribute something to [the cells'] behavior, since [they are] so absolutely uniform across this whole class of cells that have in common unrestrained growth." But glioma researcher David Louis of Harvard Medical School in Boston warns against jumping to conclusions because, he says, experimental tests of invasiveness don't truly replicate the kind of pernicious tissue invasion seen in human gliomas. Hockfield and other researchers agree that it will take a lot more study to learn whether blocking either protein will lead to therapies that can counter glioma invasiveness.

On the other hand, Ransom notes, "it doesn't matter" whether you even know what a protein does if your aim is to use it as a target for anticancer drugs. Sontheimer is already pursuing that strategy. He and others have founded a Birmingham start-up company, Transmolecular Inc., which is planning to use chlorotoxin to direct cell-killing agents to glioma cells. In preclinical trials in animals, researchers at Transmolecular have shown that chlorotoxin can ferry radioactive iodine specifically to the tumors, and Sontheimer says they are currently juicing up the chlorotoxin molecule with enough radiation to actually kill the tumor cells when the toxin binds to them. If the animal tests go well, the team plans to apply for permission to conduct clinical trials in human glioma patients by next year.

"We are very anxious in the [brain cancer] consortium to test this hypothesis," says Brem. "If it shows promise clinically, it will be a major breakthrough"—and one that glioma researchers and patients would welcome as long overdue.

–Marcia Barinaga