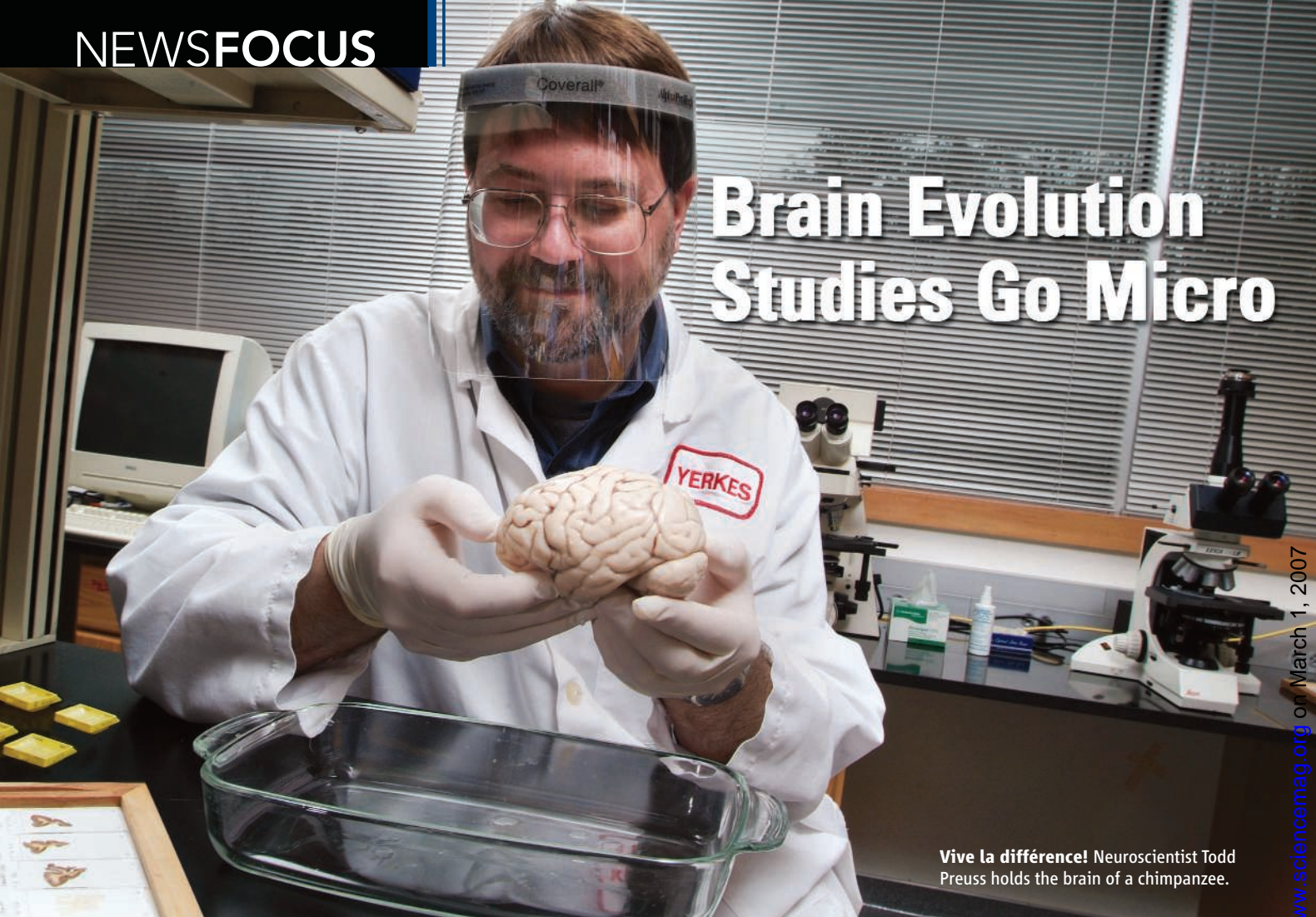


Brain Evolution Studies Go Micro



Vive la différence! Neuroscientist Todd Preuss holds the brain of a chimpanzee.

What makes the human brain unique? Researchers are coming up with new answers to that question as they shift their focus from large-scale brain structures to individual neurons and their complex wiring

NEW YORK CITY—When it comes to brains, Patrick Hof has plenty. Plastic containers filled with the brains of macaques, gorillas, chimpanzees, bonobos, and humans cram the shelves of the walk-in refrigerator in his lab at Mount Sinai School of Medicine here. During the 1990s, Hof and his team were studying human brains when they spotted a type of nerve cell they had never seen before, in a small area associated with higher cognition. At first they thought the long, narrow cell was an artifact. But then they realized that they had rediscovered a cell type first described during the 1920s. So Hof turned to his collection and got an even bigger surprise: These cells were found only in apes and humans, not other primates.

His discovery was the first demonstration that the ape lineage had evolved an entirely new type of brain cell. Since then, he and other neuroscientists have been putting primate brains under the microscope,

looking for clues to how the extraordinary information-processing capabilities of the human brain evolved.

On the macro level, many of the differences between human and other primate brains have long been obvious. Researchers have known since the early 19th century that the average human brain is nearly four times as large as that of a chimpanzee. And for decades, anthropologists have analyzed the relative sizes and visible structures of brain regions such as the frontal and temporal lobes in humans and in other living and fossil primates.

Yet in recent years, a growing number of researchers have become convinced that the size isn't the whole story. Work over the past decade by Katerina Semendeferi, an anthropologist at the University of California, San Diego (UCSD), suggests that the human frontal lobes, the seat of many advanced cognitive functions, are not pro-

portionately larger than those of other apes (*Science*, 5 May 2000, p. 798). Her work remains controversial, but it has spurred many scientists to look elsewhere for explanations. "Having a big brain is necessary but not sufficient" to explain human cognition, says UCSD glycobiologist Ajit Varki. "Neandertals had brains bigger than ours, but they did not paint on cave walls."

Now, armed with new histological and imaging techniques to identify and trace individual nerve cells, a growing number of researchers have begun looking for signs of human uniqueness that can only be spotted under the microscope. They are discovering microanatomical structures and enhancements in the wiring and connectivity of nerve cells that our ape cousins lack. "Brain size is one thing, and brain organization is something else," says neuroscientist Todd Preuss of Emory University in Atlanta, Georgia, a leading member of this avant-garde movement in evolutionary neuroanatomy. "There is a whole microuniverse of human nature for us to explore."

Like Hof's slender neuron, some small-scale innovations are shared by humans and

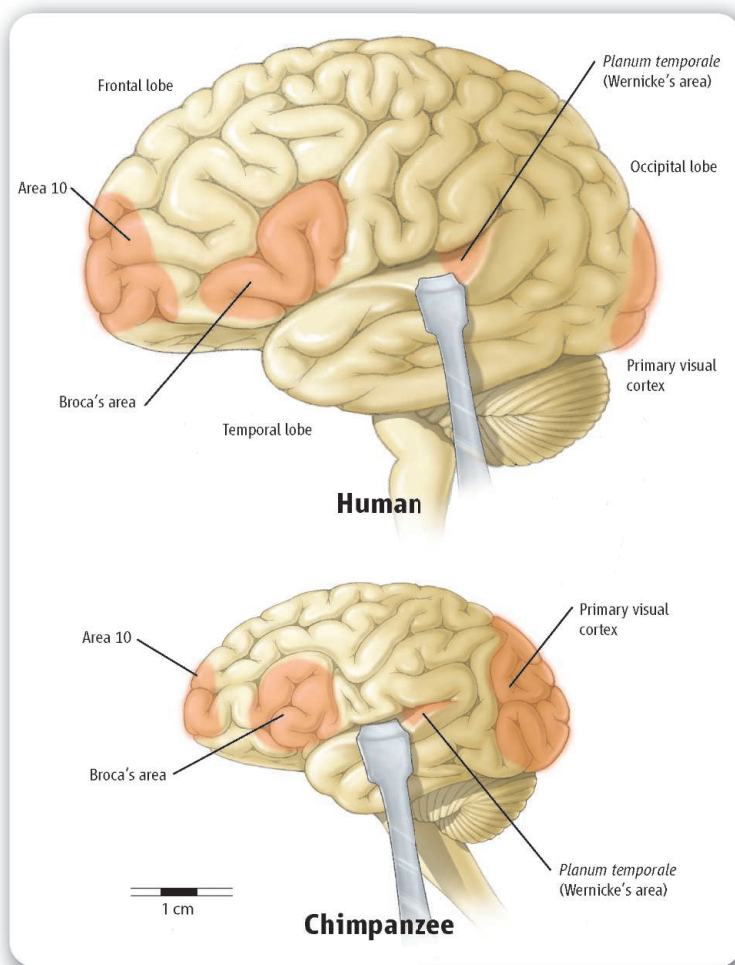
great apes but not other primates, implying that they arose after the great apes evolved about 15 million years ago but before humans came on the scene, about 5 million to 7 million years ago. Yet in nearly all cases—including Hof's discovery—these novelties show additional differences between apes and humans. Indeed, most of the ape-human distinctions are seen in parts of the brain implicated in advanced functions such as social cognition and language. "This is the first set of [microscopic] differences that define the human brain as more than just another great ape brain," says Chet Sherwood, an evolutionary neuroanatomist at George Washington University in Washington, D.C.

Despite considerable progress, the field is still in the basic discovery stage, identifying new features and trying to decipher their functions. Researchers can't point to a recently evolved nerve cell type and say with confidence that it helps humans to plan ahead or negotiate delicate social situations, for example.

On the other hand, the emerging micro differences are encouraging new hypotheses about brain evolution. These studies "have a beautiful potential and open a whole new window on the evolutionary history of [primates] that we never had before," says anthropologist Ralph Holloway of Columbia University.

Of apes and whales

Although researchers have long studied the anatomy of the brain, until recently many had assumed that all mammalian brains are basically the same at the microscopic level. "Many neuroscientists haven't wanted to imagine that the human brain is anything more than a rat or mouse brain done a little differently," Varki says. As a result, researchers have overlooked important differences between humans and their close primate kin, Preuss says. He adds that the roots of the problem go all the way back to Charles Darwin, who argued that humans were



essentially big-brained apes. Well into the 1980s, he says, neuroscientists continued to argue for what they called the "basic uniformity" of the mammalian brain.

This simple picture began to change during the 1990s, when researchers began to find subtle differences in the shapes and biochemical properties of neurons across mammalian species. They were greatly aided by new histological techniques that allowed them to label specific nerve cells and neurotransmitters. In 1999, Preuss and his co-workers were the first to show more significant microscopic differences in brain organization between apes and humans. They reported in the *Proceedings of the National Academy of Sciences* that one layer of the human primary visual cortex, which is located in the back of the brain (see diagram), differs markedly from that of



Built for speed. Slender Von Economo neurons may relay nerve impulses swiftly.

that these neurons are located in only two parts of the brain: the anterior cingulate cortex, deep in the center of the brain, and the fronto-insular cortex, located inside the frontal lobes. In humans, both of these structures appear to be involved in aspects of social cognition such as trust, empathy, and feelings of guilt and embarrassment. Not only were VENs unique to great apes, but humans had many more VENs than other apes. And the human VENs were markedly larger.

What do humans use those big VENs for? No one knows for sure, but a few hints are emerging. Last year, Allman's team reported in *Neuroscience* that human VENs seem to make fewer connections with adjacent nerve cells than do other types of neurons. And because the speed of nerve impulse conduction generally increases with the diameter of a nerve fiber, Allman hypothesizes that the large VENs might relay information rapidly from the anterior cingulate and fronto-insular cortices to other parts of the brain. "We think of them as a Ferrari relative to a Chevrolet," Allman says. "They are really stripped-down, high-performance kinds of cells."

monkeys as well as apes such as chimps and orangutans. In this layer, which helps relay visual information from the retina to the parietal lobe, nerve cells are organized in a complex meshlike pattern very different from the simpler vertical arrays of cells found in other primates. Preuss's team concluded that the meshlike arrangement was an evolutionary innovation on the human line and might help explain humans' superior ability to detect objects against a background. "This was very nice work," says Holloway.

That was the same year Hof reported the elongated neurons he had rediscovered, called spindle neurons because of their tapered shape or Von Economo neurons (VENs) after the Austrian neurologist who originally spotted them. Work by Hof, neuroscientist John Allman of the California Institute of Technology in Pasadena, and Semendeferi has shown



Brains aplenty. Patrick Hof's extensive collection helped him rediscover a specialized nerve cell.

He and others think that one target for nerve impulses from the VENs is a part of the frontal lobes called area 10 (see diagram, p. 1209), which is involved in taking initiative and advance planning; Semendeferi has argued that this region, unlike the frontal lobe as a whole, is expanded in the human line relative to its counterpart in other apes. Allman hypothesizes that the big VENs might help humans adjust behavior swiftly in response to rapidly changing social situations.

New data on dementia seem to fit that notion. Last December, a team led by William Seeley at UC San Francisco reported in *Annals of Neurology* that subjects afflicted with a type of dementia that causes inappropriate and impulsive social behavior had 74% fewer VENs in their anterior cingulate cortex compared to normal controls.

But other researchers note that it's too early to draw functional conclusions about the role of VENs in the normal brain. "They do have a [shape] that suggests they are designed for conduction of more rapid output than surrounding cells," Sherwood says. "But what they are connected to we don't know yet."

Whatever the VENs do, primates may not be the only creatures doing it. In a surprise finding last year, Hof and his Mount Sinai co-worker Estel Van der Gucht found that some large whales—including humpbacks and fins—have VENs too, as they reported in the *Anatomical Record*. This apparent case of parallel or convergent evolution could help explain the cognitive talents of some whale species, including singing and other forms of complex communication, says Hof.

Marching in column

Whereas VENs seem to be restricted to certain mammal species and specific brain regions, other researchers are exploring the uniquely human specializations of a feature shared by all mammals: the minicolumn. Discovered in the 1950s, each minicolumn is comprised of 80 to 100 nerve cells bundled together vertically in the cerebral cortex. Most neuroscientists now consider the minicolumn to be the basic modular unit of neural information processing, one that can respond to many simultaneous stimuli at once. "The minicolumn serves as a parallel processor in the brain," explains neurologist Manuel Casanova of the University of Louisville in Kentucky.

And certain human minicolumns apparently have unusually great processing capacities. In 2001, Casanova and biological anthropologist Daniel Buxhoeveden, now at the University of South Carolina in Columbia, examined minicolumns in the left planum temporale, a part of the temporal lobe involved in uniquely human activities such as language and perhaps music. As they reported in the *American Journal of Physical Anthropology*, they found that human minicolumns in this region were organized much differently than those of chimps and rhesus monkeys. Human minicolumns were much wider, an average of 51 micrometers compared to about 36 micrometers in both chimps and monkeys. This increased size was apparently due to an increase in the so-called neuropil space at the minicolumn's periphery, which contains the axons, dendrites, and synapses that make neural connections. The neuropil space was expanded even more by a tighter packing of nerve cells in the center of the minicolumn in humans compared to other primates.

This suggests that the organization of nerve cells in the planum temporale has evolved since the human-chimp split, says Casanova. In a follow-up study, the team also showed that in humans, the minicolumns of the left planum temporale are wider and have more neuropil space than those of the right planum temporale, whereas in chimps and rhesus monkeys the left and right sides are similar. And recent unpublished work by Semendeferi's graduate student Natalie Schenker shows a significant enlargement of minicolumns in area 10 as well as Broca's area, an area on the left side of the brain involved in language processing.

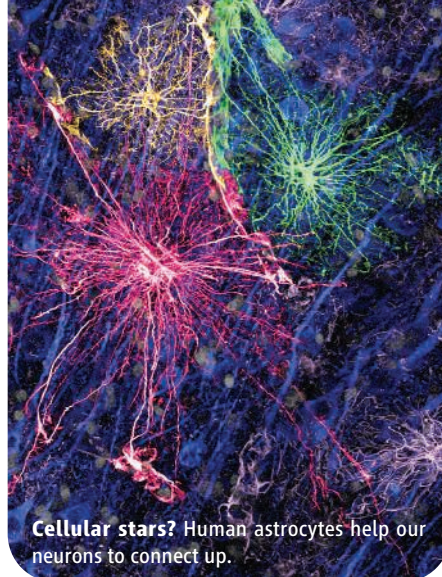
These microlevel asymmetries fit with macrolevel results: In most humans, certain areas are bigger on the left side of the brain than on the right, and some of the left-side regions, such as Broca's area, are apparently involved in language. Sherwood suggests that

the macrolevel asymmetries may reflect an underlying left bias at the micro level.

All this work suggests that the human minicolumn has reorganized during evolution to allow greater connectivity, says Casanova. That reorganization may have helped make the expansion of the human brain possible, he says: “To have a big brain, you need more connections.”

Making connections

In the nervous system, making connections is everything—and usually, the more the better. Until recently, however, little was known about what triggered the formation of synapses between neurons. Then in 2001, a team led by neurobiologist Ben Barres of Stanford University in Palo Alto, California, reported that specialized neural cells called astrocytes—which make up nearly half the cells in the human brain, but whose functions had remained a mystery—must be present for synapses to form. Astrocytes do not form synapses themselves, but Barres’s work showed that they play some sort of supporting role in creating synapses between the axons and dendrites of impulse-carrying nerve fibers. Later, Barres and his colleagues reported that astrocytes trigger synapse formation by secreting large proteins called thrombospondins (*Science*, 21 November 2003, p. 1323).



Cellular stars? Human astrocytes help our neurons to connect up.

“Thrombospondin secretion is an astrocyte function with a high impact on the capacity for neural processing,” agrees Maiken Nedergaard, a neurologist at the University of Rochester Medical Center in New York. In general, the more synapses, the greater the brain’s ability to transmit messages and process information.

Intrigued by Barres’s results, Preuss wondered whether there were any differences in thrombospondin secretion among primates. He and co-workers looked at the gene expression of thrombospondins in the brains of humans, chimps, and macaques. The team hit the jackpot: As reported online last December in *Cerebral Cortex*, human brains produce up

to six times as much thrombospondin messenger RNA and protein than do either chimps or macaques. Moreover, the differences were seen in the cerebral cortex but not in the cerebellum and nonbrain tissues.

“Todd’s findings are extremely interesting,” Barres says. “They raise the question of whether the human brain can form more synapses,” at least in adulthood. Varki agrees: “This work is excellent. It is exactly the kind of approach needed for the future.” Semendeferi adds that these results are completely consistent with her lab’s finding that minicolumns in area 10—one region where Preuss found enhanced thrombospondin expression—have larger neuropil space and thus more room for synaptic connections.

Just how much the relatively new field of comparative microneuroanatomy will contribute to our understanding of human brain evolution remains to be seen. “Some of it may work out, and some might not,” Holloway says. “What we need now is to establish a solid relationship between these structural elements and actual behavioral variations” between humans and other primates. Nevertheless, says Holloway, a pioneer in macrostudies of brain evolution, “If I were 42 years old instead of 72, I would throw all my brain endocasts away and get right into this new field.”

—MICHAEL BALTER

ONCOLOGY

Recruiting the Cell’s Own Guardian for Cancer Therapy

Reactivating the *p53* tumor suppressor gene has given promising results in mice, reversing and even temporarily eradicating some tumors

Within the past few years, biologists have begun to see their study of cancer cell genetics pay off in the best way possible: through the development of new drugs that can improve patient survival. Some specifically block the oncogenic proteins that drive tumor growth; Herceptin is a recent example. But oncogenes are only one part of the equation. Many if not all human cancers also have defects in so-called tumor suppressor genes that would normally restrain cancer development. And now, researchers are increasingly turning their attention to the tumor suppressor genes to see whether it’s possible to develop therapies that work by restoring their activity.

The lion’s share of attention has focused so far on the tumor suppressor gene known as

p53. This work, still in its very early preclinical stages, looks promising. One line of evidence comes from three recent studies showing that restoring *p53* activity can halt the growth of cancerous tumors in mice, and in some cases, even cause tumors to disappear. The papers “eloquently show that restoration of *p53* function in every cell is effective in suppressing tumors,” says Wafik El-Deiry of the University of Pennsylvania School of Medicine in Philadelphia.

In addition, researchers are hot on the trail of the field’s Holy Grail: the development of small molecule drugs that reactivate the *p53* protein. Some of this work was sparked by the discovery 3 years ago of a drug called nutlin that has shown promise in

preclinical testing; now several additional drugs are also in the pipeline. “The whole field is in a stage of very serious optimism,” says *p53* pioneer David Lane of the Institute of Cell and Molecular Biology in Singapore.

The reason drug developers are so interested in *p53* is that mutations in the gene contribute to the development of about 50% of all human cancers. In addition, tumors lacking mutations in *p53* itself often carry mutations in other genes that produce proteins that interact with and regulate *p53*. Indeed, Lane says, one way or another, the *p53* pathway may be inactivated in all human cancers.

The *p53* pathway may have evolved as a protection against cancer, helping cells cope with stresses such as DNA damage triggered by exposure to environmental toxins or radiation. When activated, the *p53* protein turns on genes that can halt cell division until the DNA damage is repaired, or it can set off a form of cell suicide called apoptosis. Thus, *p53* can help prevent the accumulation of potentially cancer-causing mutations and also put the brakes on abnormal cell growth. That’s why Lane once christened *p53* “the guardian of the genome.”