

# NIDA Notes

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## INNOVATIONS Special Issue

### Basic Science Discoveries Yield Novel Approaches to Analgesia

Research on glial cells and receptors leads to compounds that outperform morphine in preclinical trials.

BY LORI WHITTEN,  
NIDA Notes Staff Writer

For millennia, opioid medications have provided the most potent tools for pain relief. Yet, although they are the best we have, opioids fall short of being ideal analgesics: They have unpleasant side effects, their efficacy diminishes with ongoing use, they cause physical dependence, and they pose significant risks for abuse and addiction. Moreover, opioids do not quell all types of pain. For example, they provide only partial, often temporary relief to people with neuropathic pain, which commonly develops when disease or trauma injures nerves and typically produces burning, stinging, or tingling that doesn't cease.

NIDA-supported research teams are

advancing along separate paths to develop new compounds that match or exceed the pain relief provided by opioids while avoiding their shortcomings. Dr. Linda R. Watkins and colleagues at the University of Colorado at Boulder are pursuing the implications of their discovery that glia—nonneuronal nervous system cells—play a previously unrecognized key role in the generation of neuropathic pain and in opioid tolerance and withdrawal. Dr. Philip S. Portoghesi and collaborators at the University of Minnesota and Louisiana State University are tinkering with opioid molecules to capitalize on recent findings showing that opioids produce pain relief by mechanisms separate from those that produce their less desirable effects. During the past year, both groups have reported highly promising results.

#### THE GLIAL STRATEGY

Dr. Watkins' research team, in collaboration with Dr. Kirk W. Johnson and colleagues at Avigen Inc. of Alameda, California, recently relieved neuropathic pain in rats by giving them AV411 (also called ibudilast; see box, page 13), a compound that inhibits glial cell activity. In other animal studies by Dr. Watkins and other researchers, various compounds that inhibit glial cells have reduced or abolished pain that simulated many types of human chronic neuropathic pain. They also have boosted eight-fold the acute pain relief afforded by morphine.

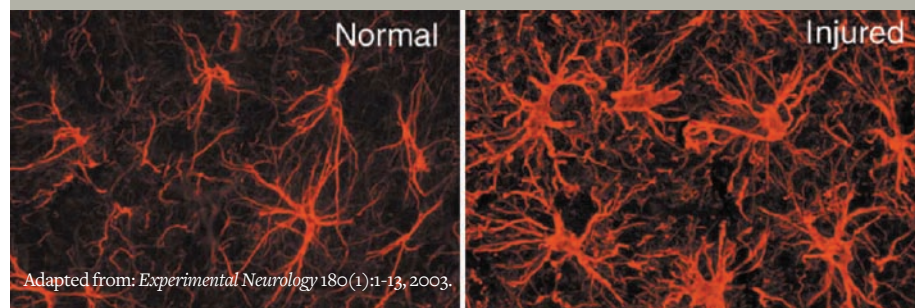
Dr. Watkins and colleagues discovered the analgesic potential of glial cell inhibition through pioneering research that implicates glial cells in the amplification and long-term maintenance of pain. Their investigations showed that tissue or nerve inflammation or damage that is associated with neuropathic pain heightens activity of two types of glial cells—microglia and astrocytes—in the spinal cord. The activated glia release biochemicals—including proinflammatory cytokines and chemokines, nitrous oxide, and prostaglandins—that excite spinal cord neurons to transmit strong and persistent pain signals to the brain. The finding that glial cells modulate pain in the spinal cord

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AFTER SPINAL CORD DAMAGE, GLIAL CELLS CALLED ASTROCYTES PROLIFERATE AND EXPAND.



Adapted from: *Experimental Neurology* 180(1):1-13, 2003.



# Research Breakthroughs in Drug Abuse Have Wide Applications in Other Fields

Each of the innovative and exciting research achievements described in this special issue of *NIDA Notes* represents a benchmark advance in NIDA's work to reduce the health and social effects of drug abuse and addiction. Each also provides a conceptual research or clinical tool that promises to transform knowledge in other health areas.

We describe, for example, a new analgesic that appears to not only lower the risk of addiction but also successfully reduce neuropathic pain that is often unresponsive to current medications in a wide range of conditions, including cancer and diabetes (page 1). Optical technologies that have the power to turn neural circuits on and off may yield information about the underpinnings of depression and serious neurological conditions (page 9). Analysis of genome-wide datasets sheds light not only on vulnerability to addiction but also on Alzheimer's disease and bipolar disorder (page 6). Research on brain development in newborns offers insights into adult neurodegenerative diseases such as multiple sclerosis and glaucoma (page 3).

Just as NIDA research contributes to other fields of science, discoveries in other fields hold promise for new drug abuse strategies and therapies. To encourage cross-pollination between drug abuse researchers and other scientists and to foster breakthrough research, NIDA has developed two innovative programs: Cutting-Edge Basic Research Awards (CEBRA) and the Translationally Oriented Approaches, Devices and Strategies (TOADS) Workgroup.

CEBRA supports new high-risk, potentially high-impact lines of research. Through CEBRA, NIDA both encourages the researchers that it funds to adapt technologies from other fields and invites scientists from other disciplines to apply their technologies to addiction research. In one CEBRA success story, a team led by physicist Dr. Mark J. Schnitzer developed a portable tool for visualizing blood vessels deep inside the brains of living mice, a technique expected to permit scientists to image neural circuits as they work (page 9).

Through its TOADS Workgroup, NIDA encourages creative interaction among NIDA-based researchers in basic science and those in clinical research. Staff scientists who participate in the TOADS Workgroup scout other fields for state-of-the-art technologies and treatment tools that they can adapt for use in substance abuse therapy. For example, in 2002, the Workgroup began investigating whether virtual environments might function as a behavior-modification tool to help substance abusers fight cocaine and cigarette cravings. Subsequently, CEBRA and other grants were awarded to investigate this promising area of research.

TOADS Workgroup members are now exploring online communities—on educational sites, virtual clinics, and support groups—as possible avenues for NIDA to provide millions of people with science-based information on drug abuse prevention and treatment. These developments and the research triumphs described in this special issue of *NIDA Notes* contribute to a growing web of connections among drug abuse research, health applications, and the wider world of scientific discovery. ■

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# Immune System Plays Unexpected Role in Brain Development

In adulthood, however, the immune complement cascade may contribute to neurodegenerative disease.

BY NIDA NOTES STAFF

**N**IDA-supported scientists have demonstrated that the immune system participates in the shaping of brain circuits during a child's development. The finding represents a breakthrough in understanding how our early experience conditions our responses to events later in life, and it may also shed light on the origins of neurodegenerative diseases, such as Alzheimer's.

Dr. Ben A. Barres and postdoctoral fellow Dr. Beth Stevens at Stanford University studied the phenomenon called neural pruning, which eliminates unused and little-used synaptic connections during development. To their surprise, the investigators found that the mechanism responsible for neural pruning is the same one that marks and destroys damaged and infected cells in the body throughout life. Their findings also indicate that the mechanism, called the immune complement cascade, may destroy healthy brain circuits in adulthood, with drastic consequences.

## SUPRISING SCULPTOR

We are all born with brain circuits ready to respond to a vast variety of experiences. As we call upon some circuits and not others, the former strengthen and the latter languish. The presence of unused or little-used circuits can interfere with brain efficiency, so the brain trims them away.

One example of neural pruning is retinogeniculate segregation, a process

that remodels visual circuits during development. Visual information from the retina travels along nerve fibers to brain areas called the right and left lateral geniculate nuclei (LGN). In the period immediately after birth, neurons in some areas of the LGN possess structures—synapses—to receive information from both eyes. As the infant develops, however, the brain eliminates many of these synapses. Most neurons lose the synapses that receive information from one eye, and from then on receive information from the other eye only.

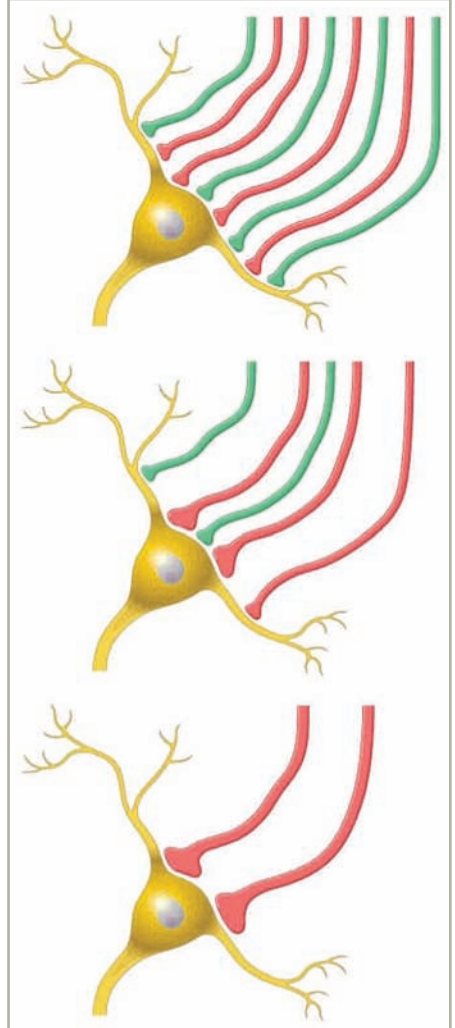
Drs. Stevens and Barres focused on retinogeniculate segregation and asked the question: What mechanism underlies this process? The electrical activity associated with normal vision certainly plays a role, but molecules must also be involved. Which ones?

To begin their inquiry, the researchers hypothesized that cells called astrocytes start the segregation process by activating genes for molecules that eliminate underused synapses. Assuming this hypothesis, they exposed purified mouse retinal ganglion cells to astrocytes. To their surprise, they found that astrocytes activated the genes for all three subunits of the protein, called C1q, that initiates the immune complement cascade.

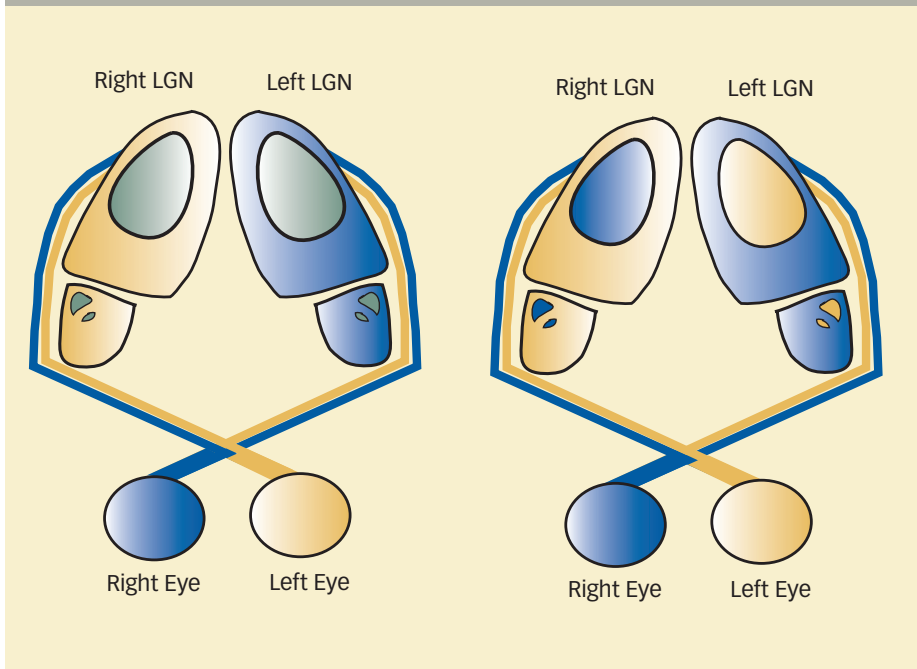
Additional experiments produced further evidence that C1q is instrumental in retinogeniculate segregation. They showed high C1q levels in the retinogeniculate system, and the rest of the nervous system, precisely during the age span when the mouse brain accomplishes neural pruning.

## WINNOWING THE CONNECTIONS

In a newborn mouse, neurons in certain areas of the lateral geniculate nucleus receive synapses from nerve cells originating in either eye (red and green). The mouse's eyes open when it's about 12 days old (top), and by day 17 (middle), some synapses from one eye have become stronger and begin to predominate. After neural pruning (bottom), only the strongest synapses remain, and most LGN neurons in these areas receive input from only one eye.



**RETINOGENICULATE SEGREGATION** In a newborn mouse (left), most of the neurons in the lateral geniculate nucleus (LGN) receive input from only one eye (tan or blue), but cells in some locations (green) receive input from both eyes. After retinal pruning (right), neurons in those areas receive input from one eye only.



The levels—along with those of other complement cascade proteins—then declined, virtually disappearing by adulthood.

These results suggested a mechanism for neural pruning: Astrocytes trigger the complement cascade, which eliminates underused synapses in the same way it disposes of dying or mutated cells and viral or bacterial invaders. To confirm this, the researchers bred mice that lacked the gene that produces C1q. As predicted by the theory, these mice did not accomplish neural pruning. Similar experiments demonstrated that C3, another component of the complement cascade, is also required for neural pruning. The team concluded that the classic complement cascade mediates synapse elimination in the developing brain.

“Neural pruning is a good thing when the brain is developing. Synaptic connections that we use frequently are strengthened, and those we don’t use enough to

justify maintaining are simply eliminated,” says Dr. Stevens. However, further research by the Stanford team and others indicates that the same synaptic pruning mechanism that is beneficial in youth may recur at older ages and, when it does, may give rise to serious health problems.

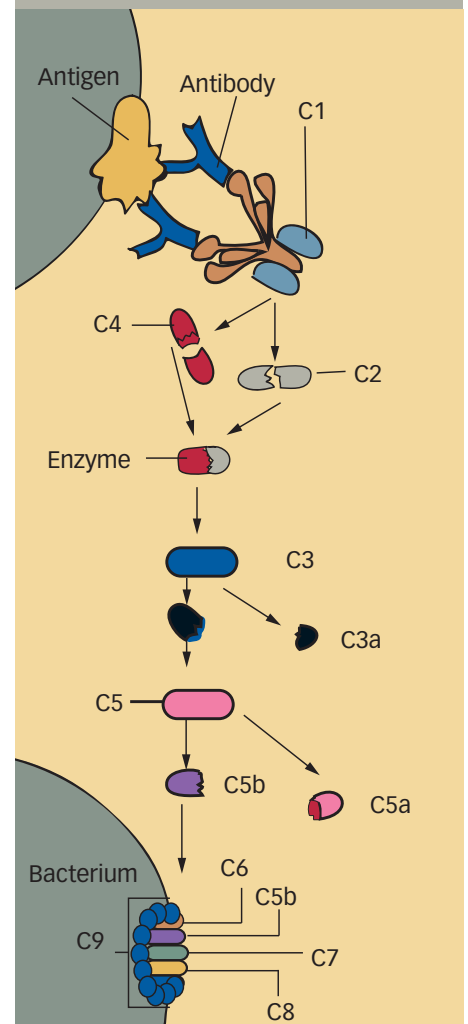
#### HELPER TURNS DESTROYER

Dr. Barres proposes that many adult neurodegenerative diseases are initiated when a neural injury activates an astrocyte reaction that, in turn, triggers the complement cascade. The nature of the injury is different for each disease. For example, multiple sclerosis begins with a severe autoimmune reaction, Alzheimer’s disease with a buildup of misshapen proteins, and glaucoma with plumbing problems that damage the retina. In each condition, however, the result is the elimination of useful synapses and destruction of essential neural circuits.

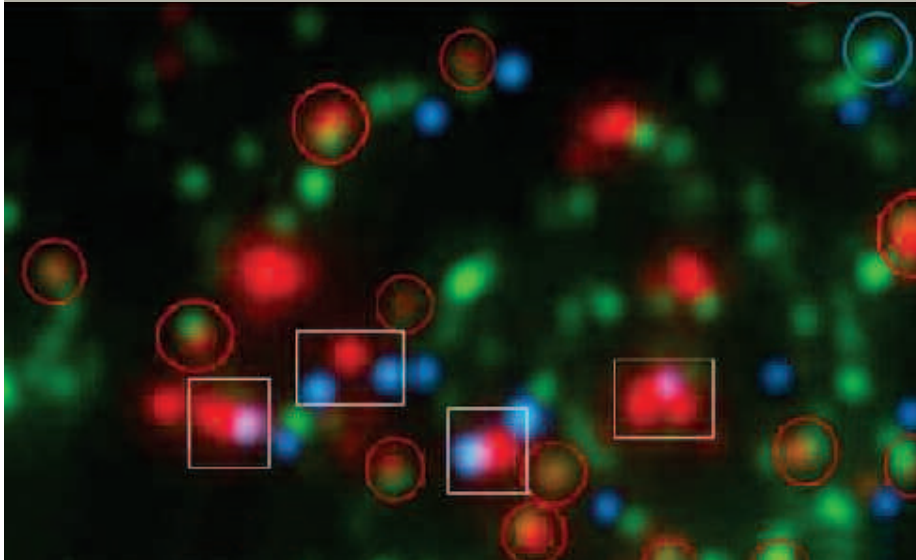
Previous observations are consistent with this idea. For example, although the adult brain does not normally express significant levels of complement proteins,

#### THE COMPLEMENT SYSTEM AT A GLANCE

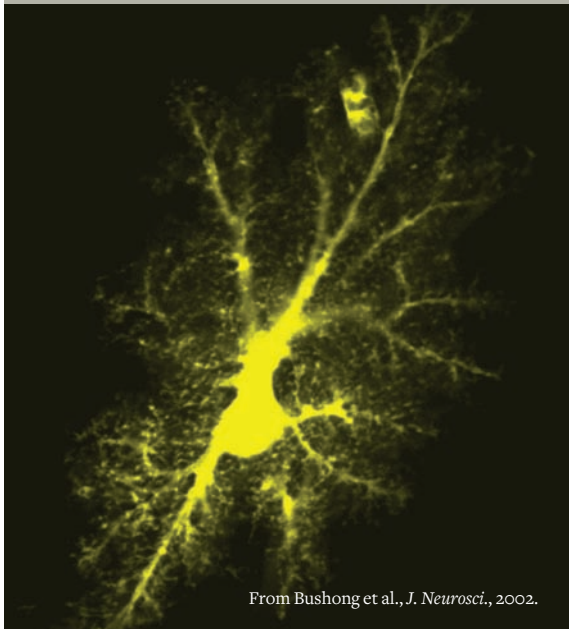
The proteins newly identified as instigators of neural pruning in the brain are integral to the cascade of activity that the immune system uses to defend the body. The first complement protein, C1, is a complex of C1q and two other proteins. C1 initiates enzymatic activity that acts on C2 and C4 to create a new enzyme that then acts on C3. The products of that reaction, in turn, mark a pathogen’s surface for immune attack, stimulate inflammation, or join with other complement components to break down C5. To kill an invading bacterium, the system finally produces a cylinder that punctures the pathogen’s cell membrane.



**IMMUNE PRESENCE** The initial protein (C1q) of the immune system's complement cascade appears green in this image of the lateral geniculate nucleus (LGN). That protein tends to associate with immature synapses (circles) but not mature synapses (boxes). Mature synapses are distinguished by the close proximity of retinal nerve inputs (red) to LGN nerve processes (blue).



**ASTROCYTE AGLOW** This dye-filled astrocyte was imaged in the brain of a living rat. A single astrocyte interacts with five or six neurons at about 100,000 synapses.



From Bushong et al., *J. Neurosci.*, 2002.

they are present in large amounts in the brains of people with neurodegenerative diseases. Individuals with Alzheimer's disease, for example, may have complement

protein levels a hundred times the normal levels.

To test the theory in one neurodegenerative disease, glaucoma, the researchers collaborated with Dr. Simon John of the Howard Hughes Medical Institute's Jackson Laboratory in Bar Harbor, Maine. Using mutant mice that were specially bred to develop glaucoma, the team discovered that complement proteins begin to build up in the adult mouse retinas long before synaptic connections are destroyed and the animals begin losing vision. Moreover, by simply measuring the C1q level in each retina, the researchers determined whether mouse retinas pro-

vided to them without labels came from animals with mild, moderate, or severe glaucoma.

In work now in progress, the

researchers are studying the severity of glaucoma that results when the mice bred to have the disease are crossbred with complement-deficient mice. If complement deficiency proves protective, then new drugs that block complement activity may provide a new therapy for this disease and perhaps others.

Drs. Stevens and Barres hold that it might some day be possible to use measures of complement proteins to diagnose neurodegenerative diseases in their earliest stages, even before they produce symptoms. To that end, the Stanford team is looking for ways to measure C1q quantities in blood. The team is also looking for ways to block the action of C1q in order to slow or prevent the development of these diseases.

Furthermore, Drs. Stevens and Barres are now investigating whether C1q and other complement proteins tag synapses early in a wide spectrum of neurodegenerative disorders. They are collaborating with neuropathologists who have access to human neural tissue in brain banks around the world.

Dr. Jonathan Pollock, chief of NIDA's Genetics and Molecular Neurobiology Research Branch, is enthusiastic about the research. "It's fabulous," Dr. Pollock says. "It's a significant breakthrough and helps us understand how synapses are eliminated during development. These researchers are expanding on Drs. David Hubel and Torsten Wiesel's Nobel Prize-winning work that proved that experience strengthens neural pathways that support function and eliminates those that don't. If this elimination does not take place, the brain receives too much input and can't process information properly." ■

#### SOURCE

Stevens, B., et al. The classical complement cascade mediates CNS synapse elimination. *Cell* 131(6):1164-1178, 2007.

# New Technique Links 89 Genes to Drug Dependence

Results point to importance of memory in addiction.

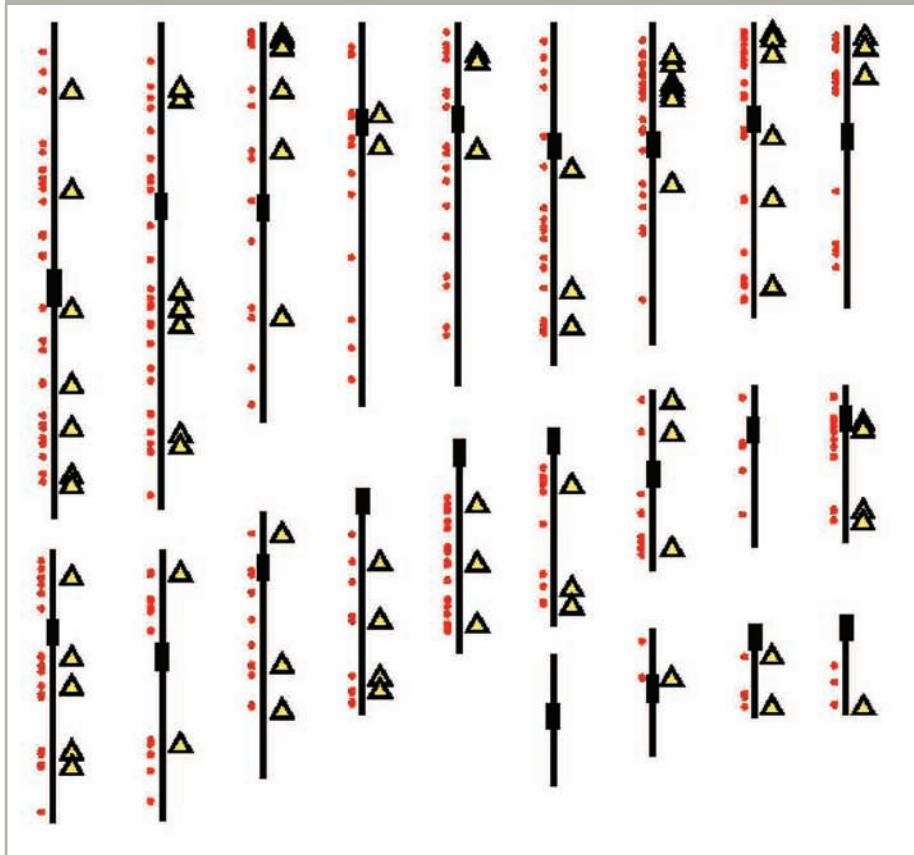
BY NIDA NOTES STAFF

**A**fter a person's first exposures to a drug, genes exert a major influence on whether he or she will go on to become dependent. Over the past decade, researchers painstakingly identified a handful of genes that appear to contribute to this influence. Recently, however, Dr. George Uhl and colleagues at NIDA's Intramural Research Program (IRP) in Baltimore, Maryland, announced that, using a powerful new technique for identifying genes that are associated with diseases, they have linked at least 89 genes to drug abuse and dependence.

The technique, called genome-wide association studies (GWAS), rapidly examines individuals' entire genomes. Researchers learn which individuals have variant forms of each of our 30,000 or so genes and then correlate these findings with other data—for example, in the present study, the subjects' drug histories. Whereas other techniques for genetic analysis can only handle small study populations of genetically similar people, researchers using GWAS can compare hundreds or thousands of individuals' genomes and establish relationships between gene variants and traits in unrelated and ethnically diverse populations. This extra power has revealed new relationships.

The findings do not indicate that someone who has one or more of the predisposing variants is bound to abuse drugs or develop addiction. Some variants will promote the disorders more and some less strongly; some may make a significant difference only if an individual also has certain others; and some

**LOCATION OF GENES ASSOCIATED WITH DRUG DEPENDENCE** On this schematic representation of human chromosomes, triangles show the location of the genes linked to abuse of multiple drugs by both European-Americans and African-Americans. Red dots indicate clusters of variations of a single molecular unit in a DNA sequence (SNP) found to differentiate drug abusers from nonabusers.



may turn out to be chance associations of genes that actually have no role in drug dependence.

Dr. Uhl, chief of NIDA's Molecular Neurobiology Research Branch, explains, "Unlike cystic fibrosis, which is caused by a single gene, in addiction and a number of complex disorders, many different genes must act together with environmental factors to create the illness. No single gene is likely to have a large effect by itself; it's the combination of effects that produce the

vulnerability to the problem."

The study's immediate significance is that it greatly expands the breadth of the genetic input that researchers recognize as potentially influencing drug abuse. Scientists can now turn to investigating how each of the 89 genes might influence the response to drugs. Among the immediate leads, Dr. Uhl's team points out that many of them appear to play roles in memory formation and processing.

## THE GENES

Dr. Uhl and his colleagues analyzed DNA samples that they collected between 1990 and 2005 from 420 European-American and 560 African-American drug abusers and from 680 ethnically matched nonabusers. Each drug abuser had used a variety of substances and was addicted to at least one illegal substance. The nonabusers had either never used drugs or had only modest exposure; none had ever been addicted. No two study participants belonged to the same family.

The researchers pooled the DNA samples into groups of 20 and applied the pooled DNA samples to microarrays containing millions of DNA probes that bind to specific short sequences of DNA. A powerful computer analysis of the bound DNA revealed single nucleotide polymorphisms (SNPs)—substitutions of a single molecular unit (nucleotide) in a DNA sequence (see box, page 8). Thousands of SNPs occurred more often in the DNA of addicted participants than in that of controls. Some of the SNPs occurred in clusters within 89 genes, where the variations in DNA would be expected to alter the way the gene performs its protein-building function.

Most of the 89 genes were associated with substance dependence among both European-Americans and African-Americans, although some appeared to affect risk in only one ethnic group. The researchers propose that this finding supports the idea that many of the variants that predispose to addiction first occurred relatively early in human evolution, before humankind's diaspora out of Africa led to the formation of separate Asian, African, and European ethnicities.

Many of the genes identified in the study were associated with addiction to several different drugs. This finding accords with those of other GWAS studies conducted by Dr. Uhl and colleagues, which revealed “a remarkable degree of overall convergence” among the SNPs related to European-Americans who were addicted to alcohol,

Asians who were addicted to methamphetamine, and smokers with European heritage. The IRP team suggests that genes that predispose to addiction to multiple drugs do so because they affect basic brain features and processes, such as nerve connections and how the brain handles information. Overall, the genes implicated by the current study meet that criterion. Most are active in the brain, where they affect such fundamental characteristics as the ways that nerve cells recognize their neighbors (via cell adhesion molecules); cell structure; enzyme activity; and actions of receptors and other cell membrane components.

## MEMORY CONNECTIONS

Among the 89 genes that Dr. Uhl and colleagues identified, the team is most intrigued by 21 that regulate cell adhesion processes. Those processes establish the correct wiring of the brain during development, maintain communication between brain cells, and keep the brain adaptable so it can modify old connections and make new ones in adulthood. Almost all of the cell adhesion genes

**In the new study, many of the genes were associated with addiction to multiple classes of drugs rather than a single drug.**

that the study linked to drug abuse are active in brain regions that play roles in memory.

“Cell adhesion molecules control the formation, stabilization, enhancement, and elimination of contacts between brain cells, which are at the core of memory formation,” Dr. Uhl says. “Finding this group of gene variants in people who are dependent on addictive substances underscores the important role memory plays in addiction and will help us understand why addicts can relapse decades after their last use of an addictive drug.”

Dr. Uhl suggests that addiction involves the implicit, as well as the explicit, memory systems. Explicit memory is immediate and conscious. Implicit memory, in contrast,

influences our behavior unconsciously. “Remembering what you need to pick up from the store tonight is a good example of explicit memory,” Dr. Uhl says. “But if you are considering those items while you are driving to work and suddenly realize you’ve gone 10 miles along your route without thinking about it, your implicit memory system is what got you there.”

Dr. Uhl speculates that cell adhesion genes may influence the risk of drug abuse and addiction by affecting both memory systems, with special impact on implicit memory systems. When vulnerable people experience the effects of an addictive drug, he suggests, their brains may store that experience deep in the implicit-memory system, where it exerts a stronger-than-usual influence on their behavior.

“Implicit memory may play a more important role than euphoria in the long-term story of a drug addiction,” says Dr. Uhl. “Although drug highs produced by the brain’s reward system motivate repeated use in the early stages of addiction, many addicts in later stages say they rarely experience dramatic euphoria and complain instead that

their drug use is driven by compulsion. They also say they can crave their drug of choice long after they stop using it when they are reminded of it by environmental or social situations. That testimony is consistent with a process in which the brain’s pleasure centers help addiction get started, but memory-like features maintain it over time.”

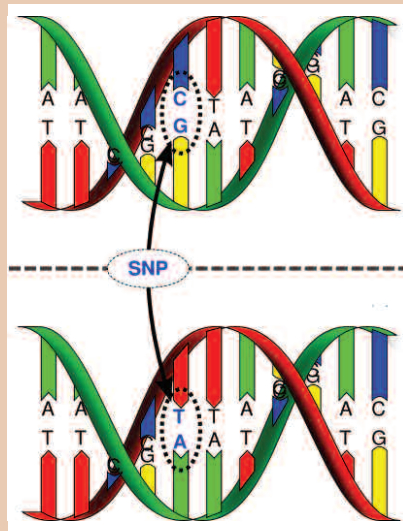
Dr. Joni Rutter of NIDA’s Division of Basic Neuroscience and Behavioral Research says, “The link between vulnerability to addiction and genes that influence the formation of neural connections and how the brain adapts to experience raises intriguing questions about how drug abuse affects the function of these genes.”

## Learning From Single Nucleotide Polymorphisms

Simple variations in genetic material are providing profound insights into the roles of specific genes in complex functions, such as behavior. Genes consist of strings of molecular units called nucleotides. Nucleotides come in four varieties, which differ from each other only in the subunits known as bases. Each nucleotide is named after its base: adenine (A), thymine (T), cytosine (C), or guanine (G).

At many locations in the human genome, the nucleotide string that makes up a particular gene is identical in everyone. That is, if you start at one end of the gene and count off the nucleotides in order along one of the two DNA strands, the result is the same—for example, AAGGGATCCAC.... At certain places along the string, however, some people have one nucleotide and others have a different one—for example, AAGGAATCCAC... instead of the more common sequence. Such a variation is called a single nucleotide polymorphism (SNP).

The variations may or may not make a difference in the way a gene functions. However, in either case, scientists can take advantage of the variation to discover associations between genes and critical traits, such as vulnerability to drug addiction.



only of addiction but of a variety of other brain-related disorders as well,” Dr. Uhl says. “In our ongoing research, we have found significant overlaps between our addiction genetics datasets and datasets for other brain disorders including Alzheimer’s disease and bipolar disorder, leading us to believe that many of these gene variants play roles in a number of brain-related problems.”

Dr. Uhl notes that the majority of a person’s 30,000 genes are expressed in the brain but that many fewer are likely to affect brain function. “It’s logical that there would be overlaps in the genetic underpinnings for various neurologic problems,” he says.

“We hope our work will eventually allow clinicians to match at-risk people with the right prevention programs and develop more effective treatment strategies for people who are already using drugs,” Dr. Uhl adds. “Just doing that will be reward enough. The wider reach of our research for other brain disorders is icing on the cake.” ■

### SOURCES

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### BEYOND ADDICTION

The IRP team is now applying GWAS in investigations of a variety of brain disorders.

“We think the 89 genes we found are the beginning of an alphabet that eventually will allow us to spell out the genetic basis not

## ADDICTION SCIENCE & CLINICAL PRACTICE

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# Optical Technologies Expand Vistas Into the Brain

Researchers in optics, genetics, and bioengineering have developed a powerful tool for visualizing cells deep in the brain and a remote control for activating brain cells.

BY LORI WHITTEN,  
NIDA Notes Staff Writer

**T**he brain, like an ocean, guards mysteries of the deep. Many of the structures and activities that make us what we are, mentally and emotionally, reside or occur far from the surface. Until now, researchers have observed deep-brain processes in living subjects only indirectly, through nuclear imaging or magnetic resonance techniques. Soon, however, if new technologies being developed by NIDA-funded scientists bear out their promise, investigators will have the means to view structures anywhere inside the organ and even activate or deactivate specific brain circuits at will. The technologies powerfully enhance neuroscientists' ability to understand how the brain functions—not only in drug abuse but across the spectrum of neurological disease. One of the new tools also opens the possibility of controlling neural circuits to counter neurological disease.

## WEE BUT MIGHTY SCOPE

In an impressive feat of engineering, researchers have created a device about the size of a dime that performs the job of a powerful tabletop microscope. The new tool sends an optic fiber bundle measuring less than 1 millimeter in diameter into a test animal's brain. The fibers deliver light to the target site within the brain and then relay images to a packet of miniaturized external components so small that a mouse can carry it on its back.

This microendoscope, designed by Dr. Mark J. Schnitzer and colleagues at Stan-

*The optical technologies described in this article have earned the National Institutes of Health Director's Pioneer Award. Dr. Karl Deisseroth was recognized in 2005, and Dr. Mark J. Schnitzer, in 2007.*

ford University's James H. Clark Center for Biomedical Engineering and Sciences, can provide clear images of structures up to about three-tenths of a millimeter away from the probe tip, which can reach a depth of 1 centimeter or so into the brain. The tool is more powerful, but has a thinner probe and smaller overall size, than earlier microendoscopes

Dr. Schnitzer's team plans to use the technology to reveal critical neurological processes as they happen. For example, researchers might observe structural changes in the circuitry of the hippocampus during memory formation or monitor a tumor growing on the brain stem.

"The technology will provide a great deal of information about how the living brain works at a very deep level—a scientific opportunity unavailable before," says Dr. Thomas Aigner of NIDA's Division of Basic Neuroscience and Behavioral Research.

## ACHIEVING DEPTH

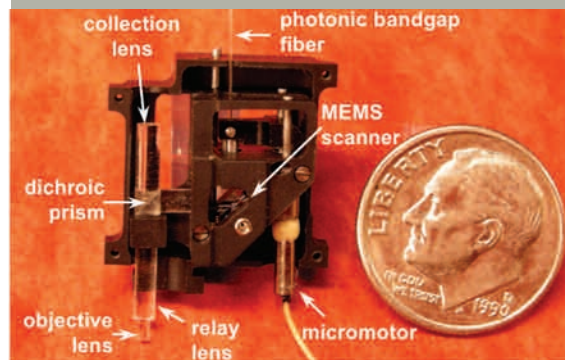
A physicist who recently turned his attention to neuroscience, Dr. Schnitzer and his colleagues built the new tech-

nology on the foundation of fluorescence microscopy. In a well-established technique called one-photon fluorescence microscopy, researchers infuse tissue with a light-sensitive dye. Shining a bright light on the tissue causes the dye to emit photons that form an image. However, because brain tissue can scatter the photons before they reach the detector, the images lose resolution when the technique is applied more than about 100 micrometers deep. Consequently, researchers developed two-photon fluorescence imaging. Although this technique produced clear images just below the brain surface, it could only penetrate about 500 micrometers into the brain.

To achieve deeper visualization, Dr. Schnitzer and colleagues inserted an ultrathin probe—0.35 to 1.0 millimeter in diameter—through the skull and into the brain. The device's external components include microlenses and micromotors that

## RESEARCHERS DEVELOP MINIATURE MICROENDOSCOPE

The external components of a two-photon fluorescence microendoscope consist of a combination of tiny lenses, fiber-optic technology, and a microelectromechanical systems (MEMS) scanning mirror. In this image, the structure is shown next to a dime. The microscope is sufficiently small that an adult mouse can carry the 2.9-gram device on its head.



power scanning, alignment, and focusing mechanisms.

The first portable two-photon fluorescence microendoscope weighed 4 grams and was the size of a matchbox. The team's latest version weighs only 2.9 grams and is about the size of a dime (see photo, page 9). In the external pack, a direct-current micromotor drives the focusing mechanism and controls silicon mirrors smaller than one square millimeter.

In one test with anesthetized mice, the 4-gram microendoscope produced detailed images of blood vessels located in the hippocampus, a structure located near the core of the animal's peanut-sized brain. In an initial test of the optical capabilities of the smaller microendoscope, the team imaged single grains of pollen and more recently microvasculature. That resolution is sufficient for portraying cells within the brain.

"Other teams are starting to use our probe technology for deep brain imaging in anesthetized animals," says Dr. Schnitzer. "My colleagues and I are developing microendoscopes that can image the brains of awake animals. We believe that scientists will find many uses for these devices in their research."

Even more valuable would be imaging technology that visualizes an animal's brain over weeks, months, or longer. Such a tool would have wide applicability in neuroscience, especially in the study of neural circuit development and cellular changes in response to experience. "Our goal is to build a general imaging tool that scientists can use to study the brain for a protracted period of time," Dr. Schnitzer says.

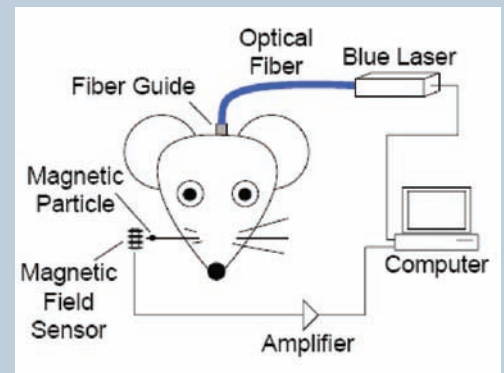
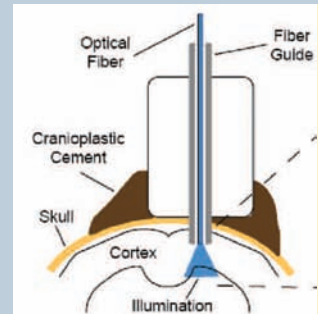
Ultimately, scientists may also combine two-photon fluorescence microendoscopy with genetic techniques to view particular neural circuits in active animals.

### REMOTE CONTROL OF BRAIN CELLS

Dr. Karl Deisseroth of Stanford University led the team that has developed opti-

## Blue Light in Brain Twitches Rodent Whiskers

Stanford University researchers used their optical remote to activate neurons in the brains of rats and mice. Dr. Karl Deisseroth and colleagues targeted neurons in a layer of the motor cortex that controls movement of a whisker. First, the scientists introduced a gene into the neurons that made them produce the light-sensitive protein channelrhodopsin-2 (ChR2). Next, the investigators directed an optical fiber 200  $\mu\text{m}$  in diameter to the cells and illuminated the protein with a blue laser diode. In response, positive ions rushed into the neurons, the cells fired, and the whisker moved. The diagrams show the optical-neural interface for cell stimulation and the magnetic field sensor used to measure the movements of the whisker, to which a magnetic particle had been attached.



Source: *Journal of Neural Engineering* 4:S143-S156, 2007.

cal remote control of discrete neural circuits. With the new technology, researchers activate highly selective sets of neurons in an animal's brain by shining a blue light on them. To deactivate the neurons, the researchers hit them with an amber light.

In the most dramatic demonstration of the optical remote to date, the researchers twitched rodents' whiskers by directing blue light at the controlling neurons within the animals' motor cortex (see box). In previous trials, they made worms (*Caenorhabditis elegans*) wriggle and become still by alternately exposing them to blue and amber light. (To see a video of this experiment, visit [www.nature.com/nature/videoarchive/braincellonoffswitch/index.html](http://www.nature.com/nature/videoarchive/braincellonoffswitch/index.html)).

Two proteins are keys to Dr. Deisseroth's system, which he developed in collaboration with his graduate student Feng Zhang, other colleagues in their laboratory at Stanford University, and collaborators at the Max Planck Institute of Biophysics, the Johann Wolfgang Goethe University, and the University of Wuerzburg in Germany. To make cells responsive to the "on" and "off" light signals, the researchers introduce DNA for the blue light-reactive protein channelrhodopsin 2 (ChR2) and the amber light-reactive protein *Natronomonas pharaonis* halorhodopsin (NpHR) into the cells they will target. For example, the rodents whose whiskers twitched carried DNA for these proteins in their cortical motor neurons, and the worm that moved and halted in response

to colored light carried it in its muscle walls and motor neurons. Cells with this DNA manufacture the proteins and store them in their membrane surfaces.

The Stanford team's system brings a long quest to a successful conclusion. ChR2 and NpHR belong to the class of proteins called opsins, which biophysicists had for years sought to utilize as neural switches. When an opsin in a cellular membrane is exposed to light, it changes shape, allowing ions to flow into the organism. Shining a blue light on a cell that features ChR2 opens a molecular gate, and positive sodium ions rush in, generating action potentials. Shining an amber light on NpHR precipitates an influx of negative chloride ions that silences the cell. The natural sources of opsins are some single-celled organisms, which rely on the proteins to regulate their activities or internal processes. The researchers culled the DNA for ChR2 from a green algae, *Chlamydomonas reinhardtii*, and the source of DNA for NpHR is a bacteria-like organism called *N. pharaonis*.

"As a research tool, optical remote control of neurons represents a huge leap forward for scientists who explore neural circuitry and neural circuit dynamics," says Dr. Geraline Lin of NIDA's Division of Basic Neuroscience and Behavioral Research.

#### FINER CONTROL

The optical remote represents a marked advance over brain stimulation by electrodes, which has been the mainstay of neurological research for more than a century. The optical filament that the researchers insert into the brain to deliver laser light to cells is much finer than a standard electrophysiology electrode (roughly the difference between the diameter of a hair versus that of a sewing needle) and so disrupts less brain tissue. The optical system can turn cells on and off at millisecond intervals, compared with second-long intervals with electrodes. Scientists can choose which cells to make light-reactive with opsin DNA and use the

optical remote to zero in on specific types of neurons. Electrodes, in contrast, are much less selective; they stimulate all the cells in a region.

With these advantages, the optical remote has wide potential application for addiction research as well as basic neuroscience. "This tool can help scientists see how neural circuits go awry in drug abuse and other brain disorders, information that may ultimately translate into therapeutic strategies," says Dr. Deisseroth, a psychiatrist with expertise in bioengineering.

NIDA's Dr. Lin says that combining the optical technology with other existing investigative techniques offers unprecedented opportunities to advance understanding of addiction. For example, researchers might document the neural activity in a particular circuit of an animal's brain during drug withdrawal, then test whether producing the same activity through use of the optical remote would also cause withdrawal symptoms. If it did, the scientists would know that the circuit was instrumental in withdrawal, and the next step might be to test whether suppressing the activity—again using the optical remote—could alleviate withdrawal.

"Before using the optical remote in people, we must proceed carefully with all the appropriate testing," notes Dr. Deisseroth. "Such an implant is somewhat invasive, but no more than current deep-brain-stimulation devices, such as the electrical pacemakers that therapeutically stimulate areas deep in the brain of Parkinson's patients."

Because optical remote technology requires that neurons produce opsins, the technique would necessitate the introduction of genetic material into human cells. Daunting as that may sound, medical scientists are already employing such techniques. For example, the U.S. Food and Drug Administration has approved studies in which researchers use a virus to introduce genes into the human brain as a potential therapy for Parkinson's disease.

The team led by Dr. Deisseroth is disseminating the optical remote technology and has provided the genetic tools and technical support upon request to scientists exploring a variety of neuroscience questions, including how fish move, how neuromodulators control neural circuit function in the mammalian brain, and how seizures begin and terminate.

For their part, Dr. Deisseroth and colleagues are currently using their invention to elucidate the causes of narcolepsy. In one experiment, they stimulated cells in sleeping mice that release the neuropeptide orexin (also called hypocretin) in the lateral area of the hypothalamus of sleeping mice. The animals awoke, verifying that these neurons, which are deficient in people with narcolepsy, regulate wakefulness.

"I believe this breakthrough technology will spur an enormous amount of research that examines and elucidates neural-circuit-activity dynamics under normal and disease conditions. It will also enable researchers to develop strategies to normalize the perturbed neural circuit activities," Dr. Lin says. "The research will ultimately translate into help for patients with various psychiatric diseases, including drug addiction, and neurological disorders." ■

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## ■ ANALGESIA

[ Continued from page 1 ]

prompted the researchers to explore glial cell inhibition as a broad strategy for controlling many types of pain.

Dr. Watkins' findings extend the roster of important functions attributed to glial cells. It was previously known, for example, that glia maintain hospitable microenvironments for neurons and provide them with molecular support for carrying out their cellular functions. Another glial cell activity, clearing cellular debris away from neurons, can last for years—a fact, the researchers suggest, that may have important implications for the long duration of chronic pain.

## ENHANCING OPIOIDS AND MAKING THEM SAFER

Working with researchers at the University of Adelaide in Australia, Avigen has completed an exploratory clinical trial to test AV411 in patients with neuropathic pain. Patients in the study, who were predominantly diagnosed with painful diabetic neuropathy, received fixed regimens of AV411 as well as concomitant analgesics, including opioids. Preliminary results indicated that patients receiving AV411 reduced their opioid use, suggesting that AV411 relieved pain or reduced opioid tolerance.

The idea that glial modulatory compounds might counteract opioid tolerance

emerged from studies to understand the biochemical basis for that phenomenon. In experiments with animals, Dr. Watkins' team demonstrated that opioids, like neuropathy, stimulate glial cells to release substances that in turn incite spinal cord neurons to amplify pain signaling. This response combines with the neuroexcitatory glial response triggered by neuropathy itself to offset the opioid's analgesic efficacy.

AV411's ability to enhance opioid efficacy could be especially valuable for chronic pain patients with histories of drug abuse. At a NIDA-sponsored panel at the annual Society for Neuroscience meeting on November 2, 2007, Dr. Watkins reported preliminary

## AV411 for Pain Relief Without Opioid Side Effects

AV411 (also called ibudilast) is prescribed in Asia to treat asthma and post-stroke dizziness. It is also being tested in Eastern Europe for treatment of multiple sclerosis and in the United States for other neurological conditions. The medication inhibits glial cells from triggering inflammatory responses that target neurons.

In Dr. Watkins' studies with laboratory animals, AV411 alleviated some types of chronic pain more effectively than morphine. Given in combination with morphine, AV411 enhanced analgesia in acute pain, retarded the development of morphine tolerance, reduced the severity of morphine withdrawal, and made morphine less rewarding.

### Alleviation of chronic neuropathic pain

#### *Two types of chronic pain from traumatized nerves*

To simulate traumatic nerve damage that causes two types of chronic pain in people, the researchers operated on two groups of rats. They surgically tied off either the sciatic nerve or two nerves that emerge from the spinal column. They then measured how hard they might press fibers against the paws served by these nerves before the animals felt uncomfortable enough to pull them away.

Among the findings were:

- In both groups of rats, twice-daily AV411 injections enhanced the animals' pressure tolerance up to three-fold for up to 16 hours, depending on the dosage;
- In rats with sciatic nerve damage, oral AV411 increased the pressure tolerance for 2 to 4 hours and was well-tolerated with only a few, passing adverse effects.

#### *Chronic pain from cancer chemotherapy*

The researchers gave rats an anticancer agent, paclitaxel, which irritates peripheral nerves. The animals' pressure tolerance declined steadily with continuing chemotherapy but stabilized and reverted toward pre-chemotherapy levels following initiation of AV411 12 or 19 days into the treatment.

### Improvement of the efficacy and safety of morphine

#### *Enhancement of morphine analgesia for acute pain*

The researchers measured how much heat a rat would tolerate on its tail before moving the tail away. Animals given AV411 plus morphine tolerated heat for a longer period of time than rats given morphine alone. AV411 boosted morphine's pain-killing potency about eight-fold.

#### *Suppression of morphine tolerance*

The researchers induced damage to rats' sciatic nerve. They then treated some of the animals with repeated injections of AV411 plus morphine and others with morphine alone. For the first 12 days, animals receiving both regimens withstood as much pressure on the paws served by the disrupted nerves as they had prior to surgery, indicating ample pain suppression. By day 16, rats that received morphine alone flinched away from lower pressures, demonstrating that the relief given by the medication was waning with repeated exposures; animals on the combination treatment, however, continued to withstand as much pressure as they had on day 12.

#### *Alleviation of morphine withdrawal*

The researchers gave healthy rats either morphine alone or morphine with AV411 for 5 days. Next, they stopped the injections and observed the animals for signs of withdrawal—including excessive chewing, jumping, grooming, and exploring. The animals that received only morphine exhibited such signs much more markedly than did the animals that received morphine with AV411.

#### *Treatment of addiction?*

Using a standard behavioral assay, the researchers found that morphine was less rewarding to rats who received it together with AV411. Similarly, AV411 dampens the neurochemical surge that underlies morphine reward in rats. Taken together, the preliminary findings suggest that AV411 has promise as a treatment for morphine addiction.

## Beyond the Central Nervous System: Analgesic Opportunities on the Periphery

The discovery of cannabinoid and vanilloid nerve cell receptors during the 1990s opened up still another avenue for the development of non-addictive pain medication. Animal research suggests that these receptors affect nerve cells' responses to pain and that modifying the activity of these receptors can produce analgesia.

### Focus on one type of cannabinoid receptor

Efforts to modulate the cannabinoid (CB) system for analgesia are currently centered on the CB2 receptor, one of the system's two known receptors. Both CB1 and CB2 receptors respond to naturally occurring neurochemicals and to drugs (for example,  $\Delta^9$ -tetrahydrocannabinol, the active ingredient in marijuana). CB2 receptors are generally not found in the brain and spinal cord but are located on nerve cells throughout the body and seem to modulate pain through actions close to the site of injury or inflammation. Because of that localization, stimulation of the CB2 receptor is not expected to cause the sedation and impaired motor coordination effects associated with CB1 receptor stimulation (see "Novel Cannabinoid Appears Promising for Treatment of Chronic Pain," *NIDA Notes* Vol. 19, No. 2).

"A focus on CB2 receptors in pain therapeutics seems a promising strategy," says Dr. Rao Rapaka of NIDA's Division of Basic Neuroscience and Behavioral Research. "Because CB2 receptors are present on immune system cells, scientists were concerned that stimulating them for pain relief may induce immunological side effects. Recent animal studies, however, indicate that the compounds do not induce such side effects."

### Chili peppers, Moroccan plant provide pain killers

Attempts to reduce pain by modulating vanilloid receptor activation are focused on a molecule called transient receptor potential vanilloid 1 (TRPV1). TRPV1s on nerve cells throughout the body detect heat and irritating touch; when stimulated, they signal those sensations to the brain. Capsaicin, the active component of chili peppers and some topical analgesic ointments, is a member of the vanilloid family of compounds that activates TRPV1s. While capsaicin produces a short-lived burning or tingling sensation upon first contact with tissues, it does not damage them; it appears to overstimulate nerve cells so that they do not again signal pain for several hours.

A potent, naturally occurring chemical called resiniferatoxin (RTX) also stimulates TRPV1s. A cactus-like plant (*Euphorbia resinifera*) in Morocco contains high concentrations of RTX, and researchers are using its resin to develop new analgesic medications. Like capsaicin, RTX-based treatments would likely have minimal potential for abuse and few side effects. Scientists expect that injectable RTX-based medications would provide more potent pain relief than capsaicin-based topical treatments. "A single RTX local injection may offer a long-term treatment for some types of chronic pain without many of the side effects that are associated with opioid analgesics," says Dr. David Thomas of NIDA's Division of Basic Neuroscience and Behavioral Research. "Animal studies have shown that RTX alleviates both inflammatory and neuropathic pain. If clinical trials show that RTX is safe and effective, patients with certain types of pain will have a much needed, powerful alternative to current pain treatments."

results suggesting that the medication counters the opioid-induced reward sensations that can raise such patients' risk for relapse. Using an established experimental model to assess reward, the researchers repeatedly infused rats with morphine in a test chamber, then let the animals decide whether to spend time in that chamber or another.

Rats that had been pretreated with AV411 spent roughly equal amounts of time in both chambers; their lack of preference for the test chamber suggests that the morphine experience was not rewarding for them. In a related study at the University of Colorado–Boulder, spearheaded by Dr. Sondra T. Bland, AV411 suppressed—by about half—a biochemical signature of drug reward, the morphine-induced dopamine surge in rats' nucleus accumbens. This research is the first to demonstrate a relationship between glial cell activity and opioid reward.

The pharmacological action that underlies AV411's beneficial effects against pain, as well as its influence on opioid reward and tolerance, appears to be partial inhibition of a particular receptor on glial cells. This receptor, called TLR4, is a member of a family of receptors called toll-like receptors (TLRs). TLR4s sit on the surface of the glial cells and detect external dangers, such as bacteria; some also respond to indicators of tissue damage, such as bits of dead cells. Once activated, TLR4s trigger an immune response to combat the invader that caused the tissue damage. Dr. Mark R. Hutchinson's research in Dr. Watkins' laboratory suggests that TLRs are also the sites where opioids and nerve damage converge to trigger glial cell responses that amplify and maintain long-lasting pain, as well as a likely site where opioids prompt the responses by glia that enhance opioid reward and tolerance.

"Blocking TLR4 separates morphine's good and bad effects, and my colleagues and I believe that drugs that block TLR4 will prove to have great clinical utility both for making morphine better for pain control and as stand-alone drugs for treating neuropathic pain," says Dr. Watkins. In addition to working with AV411, Dr. Watkins and colleagues are collaborating with Dr. Kenner Rice of NIDA to create and test highly specific TLR4 antagonists that may maximize the potential of this pharmacological action.

"Our findings suggest that suppressing glial activation with AV411 will help relieve neuropathic pain, as well as other types of pain, and also reduce feelings of pleasure from opioids that drive craving and drug-seeking among abusers," says Dr. Watkins. "Our results, when taken together with the findings of other researchers, suggest that AV411 inhibits the cascade of molecular events sur-

rounding nerve damage in the body as well as the inflammatory response in the brain. AV411 successfully reaches the brain, whereas other inflammation inhibitors do not, which likely contributes to its beneficial effects in animals.”

Dr. David Thomas of NIDA’s Division of Basic Neuroscience and Behavioral Research notes that the results of this animal research look promising. “But clinical studies are needed to determine whether AV411 will provide relief for people with neuropathy,” he adds.

### BUILDING BETTER OPIOIDS

To create improved opioids, Dr. Portoghese and collaborators at the University of Minnesota and Louisiana State University are using newly discovered details about the receptors through which opioids exert their effects on neurons. The team’s recently designed compounds provided more pain relief than morphine, and their analgesic effect did not diminish after repeated administration in laboratory tests. Mice given the compounds also showed no behavioral signs of physical dependence.

The researchers’ design strategy is to construct single compounds that target both components of a particular dual-opioid receptor. Specifically, the compounds stimulate the

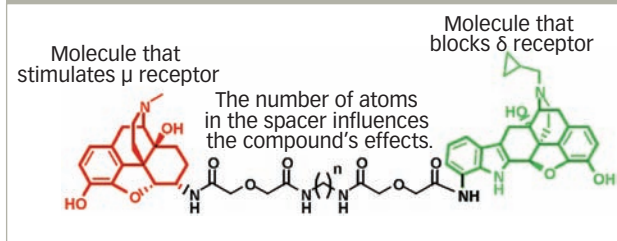
mu component and inhibit the delta component of the mu-delta dimer, or linked-pair, receptor.

Dual—or dimeric—receptors are a recent discovery. Neuroscientists long supposed that all receptors were singular entities. The finding that some receptors form pairs opened up new possibilities for medication development. Scientists observed

that when a specially designed two-part compound simultaneously engages both receptors in a linked pair, the cell reacts differently than when either receptor unit is triggered singly.

In fundamental research conducted in the 1990s, Dr. Portoghese and colleagues set the stage for the development of a new class of opioids. The results of an animal study showed that simultaneous stimulation of the mu receptor by one compound and inhibition of the delta receptor by another produced less tolerance and dependence than mu receptor stimulation alone. With the subsequent discovery of linked-pair receptors, Dr. Portoghese and colleagues recognized that targeting dimers, rather than two inde-

**DUAL-COMPONENT COMPOUNDS OPEN NEW AVENUES FOR POTENTIAL MEDICATIONS** New opioid compounds called mu-delta agonist-antagonists (MDANs) comprise two molecules tied together by a chain of atoms called a spacer (chemical structure shown). The compounds target the  $\mu$ - $\delta$  opioid heterodimer, an adjoining pair of receptors, in the neuronal membrane.

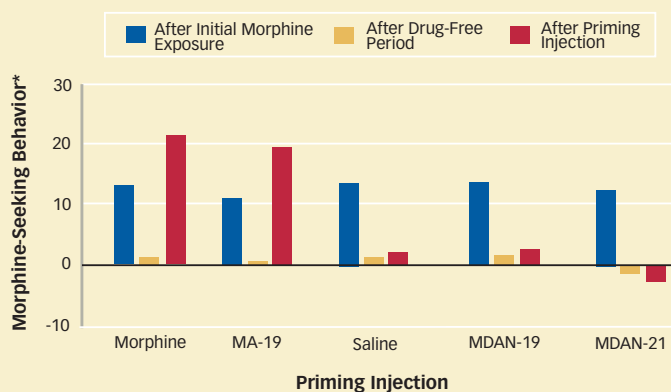


(see diagram, above). The compounds are called mu-delta agonist-antagonists, or MDANs. To maximize efficacy, the team sought spacer lengths such that once the first molecule settles into its docking area on the dual receptor, the second has just enough chain to reach its docking area. In the experiments, this criterion was met by a spacer length of 16 to 21 atoms, and the researchers designated the compounds, accordingly, MDAN-16 through MDAN-21. Taking these compounds into the laboratory, they observed that:

- *Several MDANs blunted pain.* Infusions of MDAN-16 through -21 directly into the brain increased the time that mice could withstand heat from a strong light beam on their tails. MDAN-21’s analgesic potency was 50-fold that of morphine.
- *After chronic exposure to MDAN-19, -20, or -21, the mice developed only minimal signs of physical dependence.* The researchers delivered infusions of either saline, morphine, or an MDAN into the brains of mice for 3 days and the next day pharmacologically precipitated withdrawal. The mice on MDANs exhibited fewer withdrawal symptoms (jumps) than those on morphine. Generally, mice given MDAN-19, -20, or -21 made the fewest jumps.
- *Chronic administration of MDAN-19, -20, or -21 did not induce tolerance to the compound’s pain-killing effects.* Animals maintained on these MDANs via infusions for 3

### ANIMAL STUDY SUGGESTS MDANS WILL NOT THREATEN RECOVERY

In a test of MDAN’s potential to trigger renewed drug use following abstinence, mice underwent morphine exposure to establish drug seeking followed by a drug-free period. They then received a priming injection. As expected, those primed with morphine or another compound (MA-19) that binds solely to the mu opioid receptor returned to intense drug seeking. Those who received MDAN-19 or MDAN-21, which bind at both mu and adjoining delta opioid receptors, did not.



\*Increase or decrease (percentage) in time spent in morphine-associated area as compared with baseline before initial morphine exposure.

days demonstrated pain relief from a new dose delivered 4 hours after the final infusion. In contrast, morphine's pain-killing effects dropped six-fold in the same experimental protocol.

■ *MDAN-19 and MDAN-21 show minimal potential for abuse and have little potential to induce a return to drug-seeking.* In a standard behavior test, mice increased their drug-seeking after receiving morphine but not after receiving MDAN-19 or MDAN-21. Other experiments (see graph, page 14) suggest that the new compounds are unlikely to induce relapse among people recovering from opioid addiction.

“Dual-component compounds and

dimers open up a new and exciting universe in medication development,” says Dr. Portoghese. “My colleagues and I believe that there may be other combinations of opioid receptors that when stimulated with such compounds might alleviate pain without tolerance and dependence.”

“Dr. Portoghese's results suggest that MDAN compounds could form a single drug with two functions—alleviation of pain and reduction of opiate withdrawal, tolerance, and side effects,” says Dr. Paul Hillery of NIDA's Division of Basic Neuroscience and Behavioral Research. “The MDAN compounds are also useful tools for investigators who study how stimulating the receptor complexes activates or inhibits neurons.” ■

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## BULLETIN BOARD

### NIDA Bestows Prizes at International Science Fair

To honor talented high school scientists who will produce the innovations of tomorrow and to foster their interest in addiction research, NIDA and Scholastic Corp. cosponsored the first Addiction Science Awards at the Intel International Science and Engineering Fair, the world's largest pre-college science fair, on May 11-16 in Atlanta, Georgia. Judges representing NIDA evaluated more than 50 addiction-related projects selected from the 1,500 in the fair. The winners were:

**First Place:** Kapil Vishveshwar Ramachandran, a 16-year-old senior from Westwood High School in Austin, Texas, earned the top award, which came with a prize of \$2,500. His project was titled “GluCl-alpha Ion Channel and Diazepam Binding Genes in Alcohol Addiction.” Ramachandran determined that the loss of a specific protein lessens fruit flies' tolerance to alcohol. The work provided the first link between that protein and a step in the path to addiction. In his project, Ramachandran developed a way to assess alcohol tolerance by observing flies' movement in response to light.

“The judges were particularly impressed with the winner's enthusiasm and innovative approach to exploring the neurological underpinnings of addiction,” says NIDA Director Dr. Nora D. Volkow.

**Second Place:** Ethan Garrett Guinn, a 17-year-old senior from Moore High School in Moore, Oklahoma, won the second-place award and \$1,500 for “Video Games: The Next Generation's Addiction.” The young scientist developed a survey that asked questions about gaming behavior similar to those used to assess whether drug users are addicted. It asked, for example, whether the responder chose to play video games over spending time with family

and friends or lied to others about playing. Guinn found that 69 percent of boys and 44 percent of girls showed borderline-to-severe signs of video game addiction.

**Third Place:** Shelby Marie Raye, a 15-year-old freshman from Manatee High School in Bradenton, Florida, earned the third-place award and \$1,000. Her project, “What's In and What's Out: High Schoolers' Perceptions of Coolness,” identified characteristics that affect a student's image. In Raye's survey of 389 students aged 14 to 18, both boys and girls reported that participation in athletics was “cool,” but boys said that being “funny” contributes most to coolness, while girls preferred “outgoing.”

“Our second- and third-place winners used initiative, curiosity, and good science to identify and measure relatively unstudied influences that are affecting the lives of adolescents,” says Dr. Volkow.

Scholastic and NIDA have a longstanding collaboration to provide age-appropriate educational information on the effects that drugs have on the brain, body, and behavior. For more information on the award, see <http://www.drugabuse.gov/sciencefair/>.

“We were thrilled at the quantity and quality of projects that explored addiction science, which gives us great optimism about the future of this vital field,” says head NIDA judge Dr. Lucinda Miner, deputy director of the Institute's Office of Science Policy and Communications. ■



**THREE YOUNG SCIENTISTS HONORED** From left, Dr. Lucinda Miner, of NIDA, stands with award winners Ethan Garrett Guinn, Shelby Marie Raye, and Kapil Vishveshwar Ramachandran.

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