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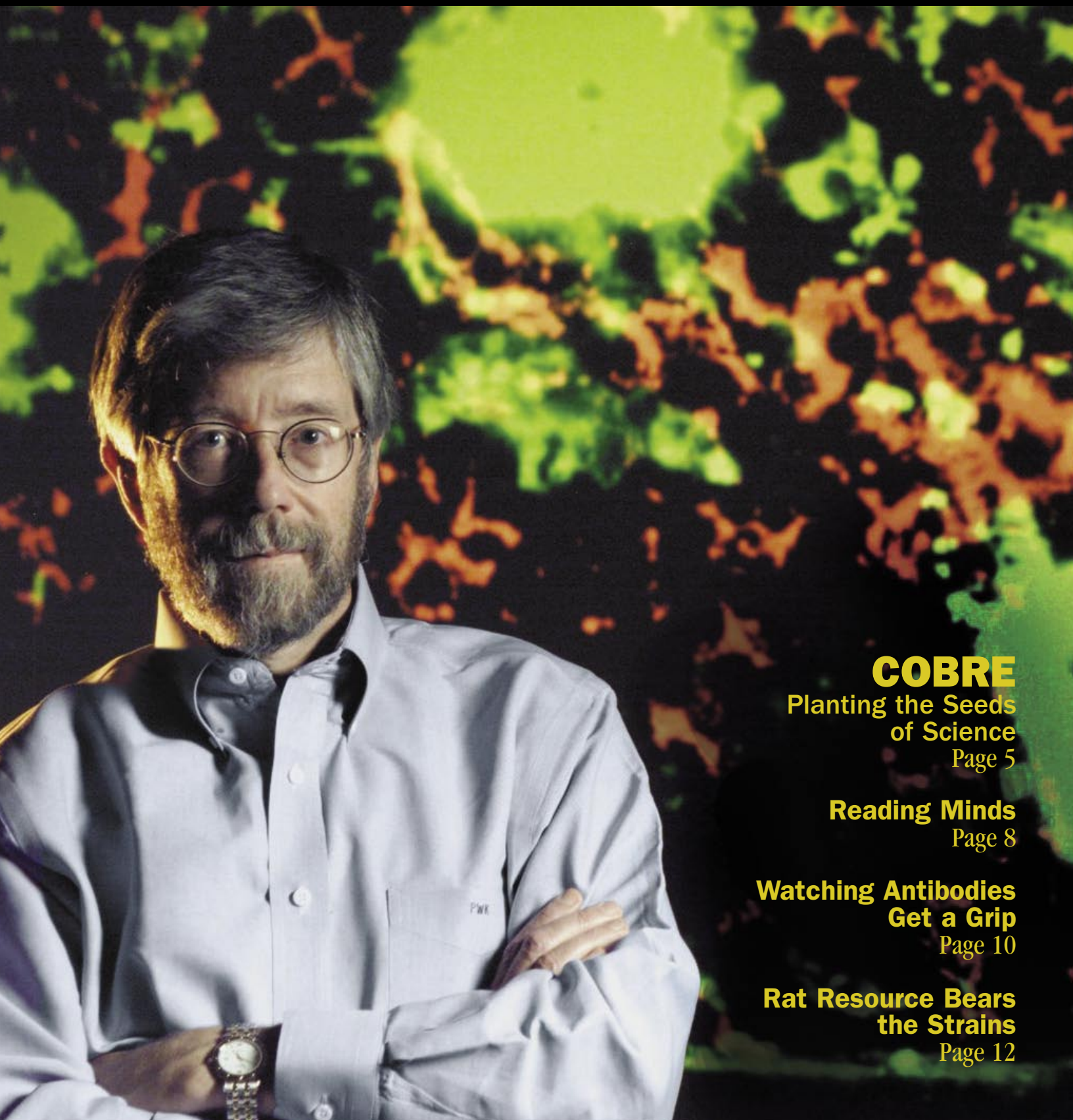

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From the Director

Forecasting for the Next Five Years

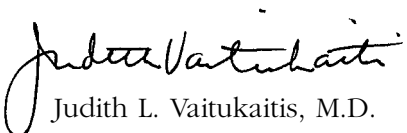
Every five years, the National Center for Research Resources (NCRR) develops a Strategic Plan to anticipate the research resource and technology needs of the NIH-supported biomedical community. This is a formidable task. To forecast future resource requirements, NCRR must track cutting-edge advances and identify emerging trends across biomedical research.

To help NCRR prepare the Strategic Plan for 2004–2008, we asked researchers from around the country to identify emerging scientific trends and make recommendations concerning research resources, biological models, and new research tools needed for future research. Researchers provided input indirectly to us via our Web site and through correspondence. That input provided a framework for discussions at our strategic planning forum held on September 10–11, 2003, in Arlington, Virginia. Biomedical scientists, high-level administrators from research institutions, representatives of scholarly organizations, and program officials from many of the categorical NIH institutes and centers, along with other interested parties, came together at this two-day session to provide their insights into the future of biomedical research. At least half of the participants at the forum had no prior history of using or developing NCRR-supported research resources. This approach gave us a fresh perspective on investigators' needs, allowing us to address new research areas.

The recommendations that were developed at the forum, along with the recommendations sent to us earlier, have now been synthesized into a document that outlines the goals and corresponding objectives that will guide NCRR programs for the next five years. The goals fall into eight areas: Clinical Research Resources and Networks; Informatics and Computational Biology; Nonhuman Models for Biomedical Research; Emerging Technologies and Instrumentation; Research Capacity Building: Resources, Networks, and Facilities; Training and Education; Research Partnerships; and Communications. The final document will be posted on our Web site at www.ncrr.nih.gov, and a limited number of hard copies will be available in the next few weeks.

It is helpful to us that our colleagues see the plan as a set of research tools to facilitate research for a wide range of NIH-supported investigations and many underlying initiatives. For instance, in describing NCRR's strategic planning process, the American Association of Medical Colleges recently reported that "the previous NCRR five-year plan anticipated many aspects of the current NIH Roadmap Initiative, including emphases on integration of clinical research and cross-disciplinary research teams. The current draft similarly complements the Roadmap, for example, by building on models such as the Biomedical Informatics Research Network, which links clinical research centers with computational and imaging facilities."

The resources and career development opportunities that you will read about in this issue of the NCRR Reporter—from building research capacity at academic institutions to providing clinical research, biotechnology, and comparative medicine resources for cutting-edge investigations—will be further developed in keeping with the 2004–2008 Strategic Plan. Stay tuned.



Judith L. Vaitukaitis, M.D.
Director, NCRR

NCRR Reporter

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Cover: Immunologist Dr. Paul Kincade, pictured before an image of stained bone marrow cells, leads one of several mentor projects that pair a senior scientist with a junior faculty member at the Oklahoma Medical Research Foundation. Funding from NCRR's Centers of Biomedical Research Excellence (COBRE) Program is strengthening the research and career development efforts in Oklahoma and other states that typically receive less federal funding for biomedical investigations. (Photo by Joseph Mills)

NCRR Reports

Research findings with NCRR support

A "Safe" Form of Mercury in Fish?

Fish are often contaminated with methylmercury, which has been linked to neurological damage and increased heart attack risk. Scientists differ as to whether methylmercury levels ordinarily found in fish are high enough to be of health concern. However, recent evidence indicates that the form of methylmercury found in fish may be less toxic than previously thought.

Using a technique called X-ray absorption spectroscopy, scientists at the NCRR-funded Stanford Synchrotron Radiation Laboratory determined that the methylmercury in fish skeletal muscle is most likely bound to cysteine, a sulfur-containing amino acid. They also showed that methylmercury cysteine is about 20 times less toxic in at least one model system—zebrafish larva—than methylmercury chloride. Assuming that methylmercury cysteine is not converted to a more toxic form when fish are eaten, these findings may be good news for diners who favor fish.

—*Science* 301:1203, 2003.

A Secret to Long Life

In an attempt to identify the biological underpinnings of exceptional longevity, scientists have found that at least one secret to a long life might be found among blood lipoproteins, which carry lipid particles.

Researchers at the Albert Einstein College of Medicine in New York City and their colleagues examined blood samples from 213 people who were, on average, a little over 98 years old, as well as more than 200 of their offspring. Examination of research participants and the processing of their blood samples occurred at the college's General Clinical Research Center, which is supported by NCRR. Compared to control groups that lacked a family history of exceptional longevity, both the elderly group and their offspring had significantly larger high-density lipoprotein (HDL) and low-density lipoprotein (LDL) particles. These lipoproteins transport cholesterol and other fatty compounds in the blood. The researchers also found

that possessing larger HDL and LDL particles was associated with a lower prevalence of age-related diseases, including hypertension, cardiovascular disease (CVD), and a constellation of risk factors for type 2 diabetes and CVD.

Why large lipoproteins would promote longevity is unknown but may involve reduced activity of an aging-associated protein known as cholesteryl ester transfer protein (CETP), which transfers cholesterol among lipoproteins. A polymorphism, or mutation, in the gene that produces CETP was significantly more common in individuals who had exceptional longevity and their offspring than in control subjects, providing a possible clue to the genetic basis of longevity. Further study may point to new strategies or therapies that promote healthy aging.

—*JAMA* 290:2030-2040, 2003.

New Source of Stem Cells

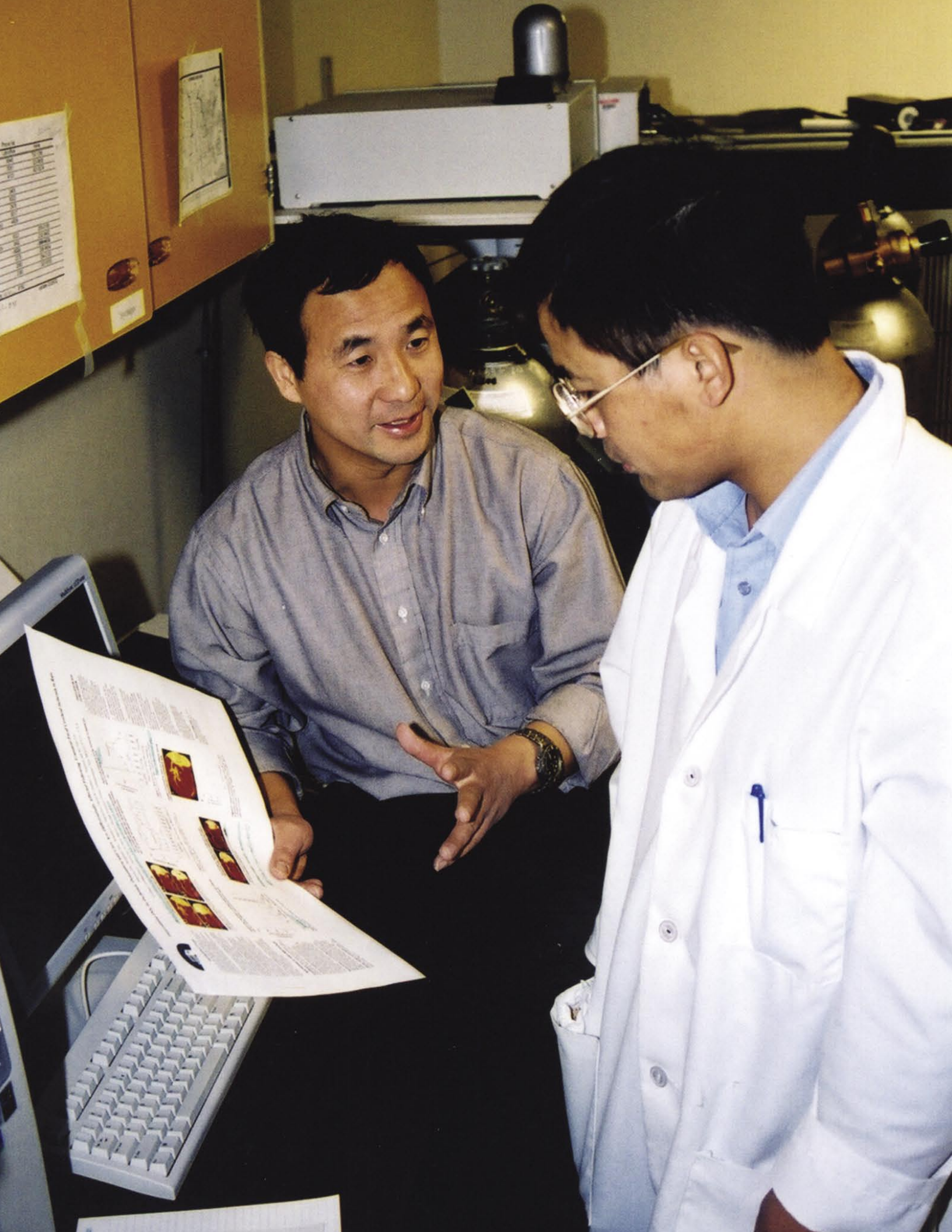
With their capacity to form different cell types in the body, stem cells have the potential to replace damaged or missing cells in patients with Parkinson's disease, diabetes, or other conditions. The problem is finding a readily available source of stem cells that can be harvested within prescribed legal and ethical provisions.

Such a source may turn out to be the pig umbilical cord. Kansas State University scientists associated with the NCRR-funded Centers of Biomedical Research Excellence Program isolated stem cells from the connective tissue inside pig umbilical cords and injected them into the brains of rats. Six weeks later, about 10 percent of the stem cells had migrated away from the injection site and lodged in another part of brain. The cells also stained positively for various neuronal proteins, indicating that the cells had taken on characteristics of neurons. No evidence could be found for an immune response against the foreign cells.

These findings indicate that stem cells derived from pig umbilical cord connective tissue might be transplanted across species. An added advantage of these cells is that they might be administered without the need for immunosuppressive drugs, which can cause serious complications.

—*Experimental Neurology* 182:288-299, 2003.

—Steven Stocker



COBRE

Planting the Seeds of Science

by Sandra J. Ackerman

Toward the end of 1999, Dr. Jim Ballard found himself considering a big change in his academic career. "I was on the verge of ending my research on anthrax because of lack of interest and support," he says. Dr. Ballard had been studying anthrax since he was a postdoctoral fellow in 1993, focusing on how the anthrax toxin gains entry to cells. But with only three people in his lab at the University of Oklahoma, and bleak prospects for hiring additional staff or obtaining the up-to-date equipment that his research required, the future of Dr. Ballard's anthrax investigations seemed doubtful. "But then the Centers of Biomedical Research Excellence (COBRE) Program was announced," says Dr. Ballard. "I thought I'd give it a try."

Drs. Ke Jian Liu (left) and Shimin Liu prepare a research poster for presentation at the November 2003 meeting of the Society for Neuroscience. Their COBRE-supported studies evaluate blood flow and oxygenation in the brain following stroke. *(Photo by Cathleen Rineer-Garber, University of New Mexico Health Sciences Center)*

The COBRE Program represents NCRR's determination to close the "biomedical research gap," the discrepancies in the amount of competitive funding that each state in the country receives from the National Institutes of Health (NIH). Nearly half the states—from Alaska to Vermont and North Dakota to Mississippi—have historically received fewer competitive research grants from NIH, in part because investigators in those states submit only a small fraction (8 to 10 percent) of the entire pool of NIH grant applications. Over the years, inadequate funding creates a vicious cycle. Institutions that receive less research support do not have access to federal support to develop the infrastructure needed for modern research laboratories, recruit top-notch scientists, or acquire state-of-the-art instrumentation and technical resources to compete successfully for NIH research grants.

Dr. Ballard's experience shows how a little financial boost can have a lasting impact. "Support from COBRE came at a crucial time, allowing me to do the experiments needed to obtain additional federal grants, get more visibility for this work, and attract researchers to my lab," says Dr. Ballard, associate professor of microbiology at the University of Oklahoma. With a five-year COBRE grant awarded in 2000, Dr. Ballard and three other lead investigators became part of the university's Center for Functional Genomic/Proteomic Analysis of Bacterial-Host Interactions, which dramatically enhanced their research environment. "We now have eight people in the lab, almost all working on anthrax," Dr. Ballard says. "We've graduated two Ph.D.s, trained a postdoctoral fellow, and obtained three competitive research grants—two from NIH and one from what is now the Department of Homeland Security."

The groundwork for this success story, and dozens of others like it, was laid by NIH a decade ago with the establishment of the Institutional Development Awards (IDeA), which assist states that have historically low success rates in competing for NIH funding. The IDeA Program, administered by NCRR's Division of Research Infrastructure, led to the creation of the COBRE grants, which support multidisciplinary research

centers in IDeA states. COBRE funding provides opportunities and resources for conducting high-quality research, helps scientists develop their talent to the fullest, and enhances the ability of junior investigators to compete independently for research support.

COBRE grants are competitive, and scientific talent is only one of several criteria considered when grants are awarded. "The principal investigator of the proposed COBRE must have a demonstrated ability in biomedical research and in mentoring, as well as in administration," says Dr. Sidney A. McNairy, director of NCRR's Division of Research Infrastructure. In addition, the proposed COBRE must have a theme that brings together a variety of academic disciplines.

The COBRE at the University of New Mexico, headed by professor of neurology Dr. Yoshio Okada, is called the Integrative Program in Central Nervous System Pathophysiology Research. This COBRE's interdisciplinary theme has concrete representation in the form of a new building, says Dr. Ke Jian Liu, associate professor of medicinal chemistry and neurology. Dr. Liu, who heads one of the four COBRE-supported research projects at the University of New Mexico, is working with colleagues to identify novel methods for integrating several neuroimaging techniques, with a primary goal of visualizing tissue oxygenation, oxidative stress, and other indicators of ischemic stroke in the brain. The building will house an array of imaging technologies not commonly seen together: magnetic resonance imaging and the newly developed electron paramagnetic resonance, electro- and magneto-encephalography, confocal microscopy, and more. "Obtaining the money to buy this state-of-the-art instrumentation has made a great difference in our research," Dr. Liu says.

Even better than the integration of technologies, says Dr. Liu, is the convergence of so many scientists with unique expertise.

But even better than the integration of technologies, says Dr. Liu, is the convergence of so many scientists with unique expertise. "We're thrilled to have all these people in one building," he says. "In the past, our researchers were scattered around the campus in different departments: psychology, arts and sciences, neurology, neuroscience, and the college of pharmacy. Now we're a close-knit group. We meet together, and we publish papers together."

The COBRE at the Oklahoma Medical Research Foundation (OMRF), dubbed "Mentoring Immunology in Oklahoma: A Biomedical Program," takes a different tack. According to principal investigator and OMRF President Dr. Donald Capra, "Among our greatest challenges are bringing a researcher to our Oklahoma institutions, providing essential resources, and getting the person to stay in this area and establish a career here." Therefore, Dr. Capra sought to develop a COBRE program that offered unique benefits difficult to match elsewhere. He decided to emphasize superb mentoring. "I paired four young faculty with four of our most outstanding senior faculty," Dr. Capra says. "I selected mentors who would agree to be responsible for getting these young investigators off and running." Each pair of senior and junior COBRE participants was asked to meet weekly, with the mentors reviewing data, helping to prepare articles for publication, demonstrating how to write NIH grant applications, and, when the junior researchers got the inevitable rejections, showing them how to respond. The early results have been positive: two of the first four participants have already been awarded R01 grants for their research, and one particularly energetic scientist has received two.

Dr. Charles Wood, director of the Nebraska Center for Virology and professor of biological sciences at the University of Nebraska, also has focused his COBRE program on training young scientists—not only those from the United States but also those from abroad. "In conjunction with the NIH Fogarty International Center, NCRR provides the resources and infrastructure that allow us to train people from other countries—physicians, healthcare providers, physician-scientists—so that they can go back home and make an impact on the health infrastructure in their own countries." In the past three years of the program, six trained physicians have returned to Zambia, although they consult often with their colleagues in Nebraska. As Dr. Wood sees it, "Once you train them, they are part of your program."

Dr. Wood works with many scientists from Zambia and conducts research there himself, studying

At the University of Nebraska COBRE, students Veenu Minhas (in back) and Saul Phiri work with human cells infected with the herpesvirus that causes Kaposi's sarcoma, a blood vessel cancer endemic in equatorial Africa. *(Photo courtesy of Dr. Charles Wood)*

how HIV infection makes children particularly susceptible to co-infection with other viruses, such as a herpesvirus that has been linked to Kaposi's sarcoma. Co-infection does not constitute a major medical challenge in the United States, but in developing countries Kaposi's sarcoma becomes a serious threat much earlier in the course of the disease and is more likely to be fatal. "Our ultimate goal is to find the factors involved in co-infection and then develop preventive strategies," says Dr. Wood. "If you can prevent HIV-infected children from getting other viruses, you improve their chances for survival." The University of Nebraska is about to extend its scope into China, with a training program similar to that in Zambia.

Back in the United States, COBRE too has extended its scope, with 62 programs now active across the country. Dr. Fred Taylor, a health scientist administrator at NCRR and director of the IDeA program, cites several success stories. As a result of a COBRE program in West Virginia, a biotechnology park has sprung up next to the university campus, a benefit that no one had anticipated. In Delaware, thousands of jobs became available, either directly or indirectly, as a result of a COBRE program. And at the individual level, in October 2003 Dr. Ballard and his colleagues published their first paper in the *Proceedings of the National Academy of Sciences*, describing the ability of treated immune cells to adapt to the anthrax toxin and survive. "This is the first example of induced resistance, not just for anthrax but for any known toxin," says Dr. Ballard.

With the first COBRE grants set to expire in 2005, the program will once again be open to applications. At the same time, says NCRR's Dr. Taylor, "we're trying to 'graduate' some COBREs to make them independent of NCRR funding." The ultimate goal, after all, is to produce more biomedical research centers that can stand on their own and compete for funding based on the quality of their work. Many COBRE-supported institutions seem to be well on their way.

The NIH Institutional Development Awards (IDeA) and the Centers of Biomedical Research Excellence (COBRE) are supported by the Division of Research Infrastructure of the National Center for Research Resources. For more information about IDeA and COBRE, see www.ncrr.nih.gov/resinfra/ri_idap.asp.

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Research Highlights

Reading Minds

How do children learn to read? It's a question that educators—especially those who work with dyslexic children—would love to see answered. Scientists believe that very young children start by focusing on the visual features of words, such the two tall lines in the middle of the word "yellow." As they mature, children begin to understand that words consist of phonemes, or units of speech that are represented by letters or groups of letters in the text.

Now a new study, conducted in part at the NCRR-supported General Clinical Research Center (GCRC) at Georgetown University Medical Center in Washington, DC, shows that this change in reading strategy is associated with a shift in neural activity from the right hemisphere of the brain to the left. Led by associate professor of pediatrics Dr. Guinevere Eden, the researchers also produced the beginnings of a brain map of reading development that may one day help to identify early signs of reading impairment or aid the evaluation of reading instruction programs.

To conduct the study, Dr. Eden and her colleagues turned to functional magnetic resonance imaging (fMRI), a noninvasive procedure ideal for examining the brain engaged in a complex activity like reading. A variation of standard MRI, fMRI provides a measure of brain activity through its sensitivity to the oxygenation status of hemoglobin, the oxygen-carrying protein found in red blood cells. Because firing neurons attract a rush of oxygen-rich blood, activated brain regions are readily detectable by fMRI.

The researchers evaluated 41 volunteers with normal reading ability, whose ages ranged from 6 to 22 years, by first conducting behavioral and reading tests in the GCRC. Participants then underwent fMRI brain scans while performing several reading activities. The goal was to isolate reading-related brain activity and observe age-related changes as reading skills mature.

The young age of many participants—more than one-third were 9 or younger—presented an unusual challenge, says Dr. Eden. Young children are notoriously fidgety, yet the brain scans required that volunteers lie completely still in a narrow, loudly clanking MRI chamber for three 20-minute sessions. Excess movement during a scan would ruin the data, and at $500 per hour, time on the MRI is too precious to waste. "We also had to be sure the children were relaxed enough that the scans would assess brain function during reading, not during anxiety," says Dr. Eden. "Obviously we want the experience to be a positive one for the children. Through this process, the children learn quite a bit about the brain and how it functions."

To address the problem, the researchers turned to an NCRR-funded MRI simulator, a type of mock instrument used increasingly in pediatric research centers. MRI simulators allow children to become more comfortable with the imaging procedures and equipment by going through practice scans and climbing on the device, which ultimately leads to more typical performance during the real fMRI scan.

A research assistant helps a study participant prepare for a trial run in the mock MRI scanner. In the real experiment, the headphones allow researchers to communicate with the subject. *(Photo by Robert Twomey, Georgetown University)*

An even greater challenge to the study design was ensuring that comparisons of brain images from younger and older participants reflected developmental changes in brain function during development of reading ability, rather than enhanced reading speed and skill acquired with age. Therefore, the researchers selected a reading activity that could be performed equally well, regardless of age and reading proficiency. Participants were shown a series of simple words interspersed with nonsense "words" made of fanciful, non-English "letters." Although subjects were not asked to read, but rather to simply identify the words with tall letters like *t* or *l* during the scan, reading is known to occur subconsciously during such activities. By comparing brain images produced when subjects viewed words vs. false-letter strings, Dr. Eden could

isolate neurological patterns that occurred during subconscious reading of single words.

The results were revealing. Older participants showed more activity in areas of the left hemisphere called the middle temporal and inferior frontal gyri, accompanied by decreased activity in certain visually oriented regions of the right hemisphere. This builds on some existing evidence that the left hemisphere handles higher order language units (such as words and phrases), while the right hemisphere works more on a letter-by-letter basis.

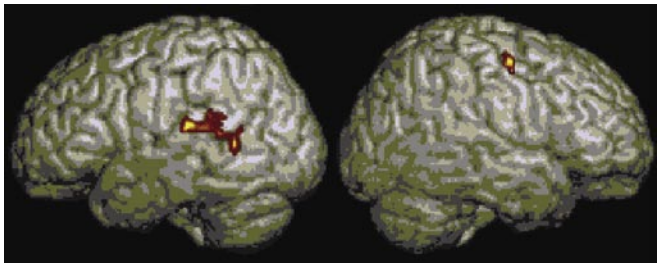
The results may point to what has gone wrong in dyslexia.

Dr. Eden is hopeful that the results will not only reveal the neural connections that underlie reading, but also point to what has gone wrong in dyslexia, which accounts for about 80 percent of all cases of reading disability. Dyslexia is characterized by difficulties with accurate and fluent word recognition and is often associated with poor spelling ability. The problems dyslexic individuals have with written language are unexpected in relation to their cognitive abilities or the effectiveness of their classroom instruction.

Getting a handle on dyslexia is difficult. Because reading has no specific genetic basis, there are no animal models or counterparts in the primate brain to guide researchers. "Our imaging studies are needed to help establish the normal signature for reading before we can study deviation of that condition in abnormal reading," says Dr. Eden.

Although all participants in her study were normal readers, some of the results provide insight into what might go awry as reading skills develop. For instance, the 26 youngest volunteers were asked to complete several skills-related tests that are commonly used to diagnose dyslexia. One is a measure of phonological awareness, or an understanding that speech is composed of units of sounds that can be represented as printed symbols. A second test challenges the subject to quickly name letters, numbers, colors, and objects on a chart. Individuals with dyslexia often score low in one test or the other, but those who do poorly on both are generally more impaired in their reading and difficult to remediate.

When Dr. Eden and her colleagues correlated the test results with the fMRI scans, different skills were



In children under 10 years old, MRI tests revealed that reading activates regions of the left superior temporal cortex (highlighted above left), as well as right-hemisphere regions (above right) that become less important as readers mature. *(Photo courtesy of Dr. Guinevere Eden, Georgetown University)*

clearly associated with distinct patterns of brain activity. Of particular note, some of the younger children who were just beginning to develop phonological awareness, a recognized prerequisite for successful reading, had enhanced brain activity in a language-related region of the left hemisphere known as the superior temporal cortex. The finding hints that an fMRI examination of this brain region might serve as an early predictor of reading outcome.

In future GCRC-supported investigations, Dr. Eden plans to perform similar scans on children with different subtypes of dyslexia to see if comparable patterns of brain activation emerge. "We want to know whether these differences in reading profiles reflect a difference in underlying brain physiology," she says.

Dr. Eden stresses that the results are still preliminary and that activation of a brain area is not absolute proof of its involvement in reading. But she hopes that confirmation will come with longitudinal studies of children, in which the same subjects are tested repeatedly over time.

—Jim Kling

This research is supported by the NCRR Division for Clinical Research Resources, the National Institute of Child Health and Human Development, and the National Institute of Mental Health.

For more information about NCRR's clinical research resources, see www.ncrr.nih.gov/clinical_rsrch.asp.

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Watching Antibodies Get a Grip

As critical components of the immune system, antibodies play a lethal game of tag with foreign substances, or antigens, flagging them for destruction. On first encounter with an alien molecule, antibody-producing immune cells known as B lymphocytes deploy a wide variety of antibodies, only some of which have the proper shape to bind tightly to the antigen. Cells that generate the best-fitting antibodies proliferate, while cells that produce less-successful antibodies die off. With repeated exposure to the antigen, B lymphocytes continuously evolve to produce even tighter-gripping antibodies. Thus the immune system hones its defenses by fraternizing with the enemy. Despite extensive study, scientists have lacked details about how this process progresses at the molecular level.

Now Dr. Roy A. Mariuzza, professor of biochemistry at the University of Maryland Biotechnology Institute in Rockville, Maryland, and his colleagues have captured the first high-resolution, three-dimensional snapshots of maturing antibodies as they perfect their ability to latch onto a specific foreign protein. Their findings lend insight into the evolution of protein-protein interactions and also may help to streamline the development of monoclonal antibodies, which are used increasingly as medications for a wide range of disorders, from cancer to allergies. The research included collaborative work with Dr. Sandra J. Smith-Gill, who isolated the antibodies at the National Cancer Institute in Frederick, Maryland.

Dr. Roy Mariuzza's structural studies of antigen-antibody pairs showed how antibodies evolve to gain a tighter hold onto foreign molecules. *(Photo courtesy of Dr. Mariuzza)*

To effectively grasp a foreign protein, the molecular shape of an antibody and an antigen must be precisely aligned, much like the oft-used lock-and-key analogy. That careful alignment can be achieved in a matter of weeks or months, driven by spontaneous mutations as B lymphocytes reproduce and multiply. Offspring cells that generate better-fitting antibodies become even more prolific, spawning daughter B cells with slight mutations that may produce even tighter-gripping antibodies. Over time, these cellular mutations collectively fine-tune the antibodies' shapes to perfectly match a specific antigen. This process of molecular evolution, called affinity maturation, often enhances an antibody's binding ability by 100-fold during an immune response.

To get a picture of the hidden mechanics of affinity maturation, Dr. Mariuzza's research team examined crystals of four different antibodies, each clutching the same antigen, in successive stages of affinity maturation. In contrast, the few crystallographic studies of affinity maturation performed in the past all analyzed antibodies bound to small molecules, even though most biological antigens are large proteins.

The scientists' detailed molecular analyses depended on the use of high-intensity X-rays available at the NCRR-supported Resource for Macromolecular Crystallography at the National Synchrotron Light Source, Brookhaven National Laboratory, in Long Island, New York. The resource, headed by Dr. Robert Sweet, develops new technologies and research methods and provides scientists with access to five synchrotron beamlines, each equipped with state-of-the-art X-ray detectors and other advanced instrumentation.

By exposing crystallized samples of antigen-antibody pairs to beams of synchrotron radiation, and then analyzing the patterns produced by the diffracted X-rays, Dr. Mariuzza and his colleagues were able to determine the three-dimensional structures of the molecules. The synchrotron-derived images had such high resolution that they revealed extremely slight differences in the alignment of the antibodies—variations that were only several atoms wide.

When the scientists compared the computer-generated images of the four antigen-antibody pairs, they could clearly see how the antibodies evolved during affinity maturation. What they found were not dramatic structural transformations, but rather minor modifications. The researchers discovered that a mere handful of amino acid substitutions in the antibody improved its fit to the antigen. "The structure of the antibody basically adjusts over time so that it fits better against the antigen. Imperfections are eliminated at the

interface," says Dr. Mariuzza. "These imperfections can be holes or cavities where things don't fit together quite well, or maybe atoms that are clashing because they are too close."

These structural changes collectively foster another major change that draws the antibody closer to the antigen—a progressive increase in the binding regions that are hydrophobic, or repelled by water. Like two drops of oil that quickly unite when dropped into a bucket of water, the hydrophobic regions of the antigen and antibody are promptly drawn to each other.

Surprisingly, the changes that occurred in the antibodies over time were not made in the binding regions, or "hot spots," in the central core of the antibody. Instead, these changes occurred in areas that surround the hot spots. "You might think that a way to improve affinity would be to make more hot spots in the center, or to make existing hot spots even hotter. But that's not what we saw," says Dr. Mariuzza. "We think that's because the central core of the antibody is about as good as it can be—it's already been optimized. To improve beyond that, changes must be engineered elsewhere in the interface, which is why affinity maturation proceeds in peripheral regions that don't quite fit together yet."

Dr. Mariuzza suspects that his findings can be generalized to explain the affinity maturation of most antibodies, and therefore might provide insights into engineering monoclonal antibodies for use as therapeutics. Monoclonal antibodies are homogenous antibodies produced by fusing a single B lymphocyte clone with tumor cells, thereby generating cells that will proliferate indefinitely and produce relatively large amounts of a single type of antibody. Currently, drug companies try to mimic natural affinity maturation by inducing random changes in antibodies until they find one that fits snugly with the antigen of interest. But Dr. Mariuzza's findings suggest more targeted approaches to improving antibody affinity, such as inducing changes only on the portions of the antibody surrounding the central binding regions.

Dr. Mariuzza began working on antibody maturation by collaborating with Dr. Cesar Milstein, who won the 1984 Nobel Prize in Physiology or Medicine, along with Dr. Georges Köhler, for developing the first monoclonal antibodies. Their technique revolutionized medical diagnostics, which uses monoclonal antibodies in tests for a range of conditions, from strep throat to pregnancy. Monoclonal antibodies also are used as new drugs for breast cancer, non-Hodgkin's lymphoma, and asthma. So it is only fitting that Dr. Mariuzza's investigations, which began in collaboration with the father of monoclonal antibodies, should offer a way to improve their development in therapeutics.

Dr. Mariuzza's findings go beyond the realm of immunology, as they provide more general insights into how proteins bind to each other. Protein-protein binding underlies many physiological activities, from the actions of hormones to the spread of cancer cells. "There's a lot of interest in designing compounds that block protein-protein binding," he says. "Designing a molecule that prevents a hormone binding to its receptor, for example, can have important pharmaceutical implications." Such studies have been difficult to conduct in the past because of limited understanding of the factors that drive protein-protein recognition. But as more investigators uncover the chemistry of protein-protein attraction, says Dr. Mariuzza, bioengineers may be able to develop a whole new class of medicines.

—Margie Patlak

The findings go beyond the realm of immunology, as they provide more general insights into how proteins bind to each other.

For more information about the NCRR-supported Resource for Macromolecular Crystallography at the National Synchrotron Light Source, visit www.px.nsls.bnl.gov. For more information about other NCRR-supported synchrotron resources, visit www.ncrr.nih.gov/ncrrprog/btdir/Synchron.asp.

This research is supported by NCRR's Division for Biomedical Technology Research and Research Resources and by the National Institute of Allergy and Infectious Diseases.

Additional Reading

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2. Sundberg, E. J., Andersen, P. S., Schlievert, P. M., et al., Structural, energetic, and functional analysis of a protein-protein interface at distinct stages of affinity maturation. *Structure* 11:1151-1161, 2003.

Critical Resources

Rat Resource Bears the Strains

Rats have long been a leading animal model in the biomedical research laboratory, contributing to the understanding of cardiovascular disease, cancer, transplantation, behavior, and pharmacology, among other areas. The laboratory mouse, in contrast, has proven to be more amenable to genetic manipulation and cloning, allowing scientists to create a remarkable array of genetically defined mice. Nevertheless, the biomedical literature still contains more peer-reviewed studies of laboratory rats than mice, although the gap has narrowed in recent years.

Rats remain more difficult to genetically engineer and clone, but progress is being made on these fronts by scientists at the Rat Resource and Research Center (RRRC), headed by Dr. John K. Critser, professor and director of the Comparative Medicine Center, and Dr. Lela K. Riley, professor of veterinary pathobiology and director of the Research Animal Diagnostic Laboratory at the University of Missouri. The resource, which opened in January 2002, is funded by NCRR and the National Heart, Lung, and Blood Institute.

Similar to NCRR's existing Mutant Mouse Regional Resource Centers, the RRRC is a multi-institutional clearinghouse for existing rat models that researchers have developed and would like to share with other scientists. RRRC investigators conduct research and provide the resources for preserving, maintaining, and distributing these specially developed rat strains that might otherwise be lost to the scientific community.

These rats were born following transfer of cryopreserved embryos. Cryopreservation prevents loss of valuable lines due to genetic instability, disease outbreaks, and other hazards. *(Photo by Yuksel Agca, University of Missouri)*

RRRC preserves rat strains that might otherwise be lost to the scientific community.

In addition to its core facilities at the University of Missouri in Columbia, the RRRC includes a research team at Northwestern University Children's Memorial Institute for Education and Research in Chicago and the Indianapolis-based Harlan-Sprague Dawley, Inc., a commercial producer of laboratory animals that provides housing and some services for RRRC strains.

The research and services provided by RRRC offer significant benefits to biomedical science. The rat is considered by many to be the best animal model for cardiovascular disease and diabetes. Its larger size makes the rat easier than the mouse to monitor when examining blood volume or certain tissues and organs.

The criteria for accepting new rat models to the RRRC are relatively straightforward. There should be some demand for the model, but it should not be available from commercial or other sources. "We are looking for high-quality models that address diseases that are the highest priorities for the NIH and biomedical investigators," says Dr. Critser. Researchers can apply to RRRC to propose that a particular rat model be preserved and distributed. RRRC has funds to grow by 10 models per year through 2006. In the first year alone, the Center received more than 40 applications.

When new rat models arrive at the Center, the animals first are purged of infectious diseases and undergo phenotype characterization. Gametes and embryos eventually are frozen and made available to other qualified researchers for a nominal fee, which helps to cover the Center's costs. RRRC scientists also are enhancing techniques for identifying and eliminating microbial pathogens in gametes, as well as improving methods for cryopreserving rat embryos and sperm.

About 20 rat strains have been accepted by the RRRC, including a spontaneous mutant that serves as a model for hereditary polycystic kidney disease and several transgenic animals. Animals generally are supplied to scientists as one or two breeding pairs. In the future, rat strains also will be available as frozen embryos and gametes or even tissue samples.

The RRRC has on-site research facilities that are available to outside scientists, primarily for collecting or preserving tissue samples. "We have everything a researcher might need to collect and process rat tissue samples," says Dr. Riley.

RRRC investigators are now tackling some of the research problems that have slowed the development of new rat strains. Rat models may be discovered as spontaneous mutants, or they can be created by inserting new genes or overexpressing existing ones. But the RRRC currently lacks one crucial type of rat model: the gene knockout, in which a specific gene is disrupted so that its protein product is nonfunctional or not produced at all. Knockouts are a "very powerful tool in mice," where they help to pinpoint the biological roles of specific genes, says Dr. Riley.

Scientists are uncertain why knockouts are so much more difficult to produce in rats than in mice. One problem is that rat embryonic stem cells (ESCs) have not yet been isolated and cultured, whereas mouse ESCs have been widely used for nearly two decades. "Rat embryos also may be more sensitive to the stresses induced by knockout techniques than are mice," says Dr. Critser. "Until we have a better understanding of the basic biology that underlies rat embryonic development, it will be difficult to know for sure."

RRRC-affiliated researchers at Northwestern University have made some progress in this arena through use of nuclear transfer. They have observed embryo development following insertion of a genetically engineered nucleus into an egg cell, although the embryos do not survive long enough to produce pups.

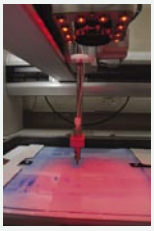
In time, Dr. Critser foresees great things for the rat. "It is a little harder to handle and to maintain, but that will soon change," he says. "I expect the rat will supersede the mouse in many types of experiments in the coming years."

—Jim Kling

For more information about the Rat Resource and Research Center (RRRC), visit www.nrrrc.missouri.edu, or contact Dr. John Critser at the University of Missouri; phone: 573-884-9469; fax: 573-884-7521; email: critserj@missouri.edu.

The RRRC is supported by NCRR's Division of Comparative Medicine and by the National Heart, Lung, and Blood Institute. To learn more about other NCRR-supported comparative medicine resources, see www.ncrr.nih.gov/comparative_med.asp.

New Resources Enhance Diverse Research



NCRR has established several new resource centers to promote investigations related to proteomics, rare diseases, and swine models of human disorders.

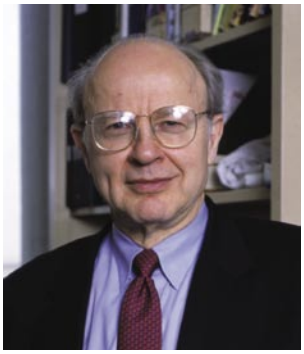
Three of the new resources develop integrated technologies that enhance the study of proteomics and glycomics, two emerging fields that seek to identify and uncover the structures, functions, and interactions of the thousands of proteins (proteomics) or carbohydrates (glycomics) found in cells. The new resources are the Proteomics Research Resource for Integrative Biology at Pacific Northwest National Laboratory, Integrated Technology Resource for Biomedical Glycomics at the University of Georgia, and the Integrated Proteome Technologies for Pathway Mapping resource at the University of Michigan, which houses the high-throughput robotic analysis system shown above right.

Another new resource was established in response to the Rare Disease Act of 2002, which directed NIH to support centers of excellence for clinical research on rare diseases. NCRR and five other NIH components established the Rare Diseases Clinical Research Network, consisting of seven Rare Diseases Clinical Research Centers (RDCRCs) throughout the country and a Data and Technology Coordinating Center, located at the University of South Florida. All RDCRCs utilize the resources and staff of nearby General Clinical Research Centers, which provide investigators with the research environment and trained personnel needed to conduct clinical research. Each RDCRC specializes in a particular group of diseases, such as rare lung diseases or urea cycle disorders.

A new comparative medicine resource, the National Swine Research and Resource Center located at the University of Missouri-Columbia, will be a national repository and distribution center for genetically modified swine. The center will house 150-250 pathogen-free swine and will cryopreserve genetic material and reproductive cells so that important swine models can be rederived as needed. Because the anatomy and physiology of pigs are remarkably similar to humans, the animals are ideal models for studying diabetes, cardiovascular disease, and obesity. The new resource also will conduct research aimed at improving cryopreservation, eliminating pathogens, and producing transgenic and knockout swine.

News from NCRR

NCRR-supported Scientists Win Lasker Award and Thomas Prize



Dr. Robert Roeder, professor and head of the laboratory of biochemistry and molecular biology at Rockefeller University, received the 2003 Lasker Award for Basic Medical Research, presented September 19 in New York City. Dr. Roeder pioneered studies of the mechanisms by which higher organisms convert the genetic information in DNA into RNA molecules, the blueprints for proteins. This process—called transcription—had previously been deciphered in bacteria, but Dr. Roeder showed that it was more complex in eukaryotic cells, which have chromosome-containing nuclei. Dr. Roeder also reproduced eukaryotic transcription in the test tube and developed techniques for identifying the proteins involved in transcription control. Over the years, his studies have drawn on several NCRR-supported resources, including the National Resource for Mass Spectrometric Analysis of Biological Macromolecules at Rockefeller University and equipment purchased through NCRR's Shared Instrumentation Grants.

Dr. Ernest Beutler, professor of hematology and chair of molecular and experimental medicine at The Scripps Research Institute, received the 2003 E. Donnall Thomas Award on December 8 at the American Society of Hematology (ASH) annual meeting in San Diego. The award, named for a winner of the Nobel Prize for Physiology or Medicine and past ASH president, recognizes pioneering research advancements in hematology. Dr. Beutler studies human genetic diseases, including hereditary hemochromatosis, Gaucher disease, and various causes of hemolytic anemia. Dr. Beutler is the principal investigator at the Scripps General Clinical Research Center.

Exhibit Marks 25th Anniversary of the Home Pregnancy Test

The Office of NIH History has produced a Web-based exhibit that tells the story of the home pregnancy test, one of the most widely used home healthcare products in the United States. The development of the test in the 1970s depended on the early research of NCRR Director Dr. Judith Vaitukaitis and Dr. Glenn Braunstein, currently chairman of medicine at the Cedars Sinai Medical Center, Los Angeles.

At the time, Dr. Vaitukaitis was a young medical resident from Boston who came to NIH in 1970 to study reproductive endocrinology. She and Dr. Braunstein, under the mentorship of Dr. Griff Ross of the National Institute of Child Health and Human Development, sought to develop a sensitive, accurate test for human chorionic gonadotropin (hCG), a hormone secreted only during pregnancy or in association with certain types of cancer. The researchers succeeded in developing an assay that specifically identified an hCG subunit and did not cross-react with other hormones. They reported their discovery in 1972, and within a few years private companies started marketing home pregnancy kits that used the assay.

The Web site (www.history.nih.gov/exhibits/thinblueline) details the history of hCG research at NIH, along with quotes from Drs. Vaitukaitis and Braunstein. The site also has a section on the history of pregnancy testing, starting with a test used in ancient Egypt more than 3,000 years ago, and a section on the impact of the home pregnancy test on popular culture.

New IOM Members Aided by NCRR

Among the 65 new members elected to the Institute of Medicine (IOM) in October are 13, listed below, who have depended on NCRR-supported resources for their research. IOM members have made outstanding contributions to health, medicine, and related fields.

Dr. Ann M. Arvin, Lucile Packard Professor of Pediatrics and professor of microbiology and immunology at the Stanford University School of Medicine, studies neonatal herpes simplex virus infections. Her research has depended on the staff and resources of the NCRR-supported General Clinical Research Centers (GCRCs) at the University of Alabama at Birmingham and Stanford University.

Dr. Rebecca H. Buckley is the J. Buren Sidbury Professor of Pediatrics, professor of immunology, and chief of the division of pediatric allergy and immunology at Duke University Medical Center. Dr. Buckley depended on the GCRC for the development and evaluation of a bone marrow transplant therapy for infants born with severe combined immunodeficiency.

Dr. Francis V. Chisari, professor of molecular and experimental medicine and director of the GCRC at The Scripps Research Institute, studies the immunobiology and pathogenesis of the hepatitis B and C viruses in humans, chimpanzees, and other animals.

Dr. Jeffrey Drazen, professor of environmental health at the Harvard Medical School and editor-in-chief of *The New England Journal of Medicine,* investigates respiratory disorders. His clinical research has drawn on the resources of the GCRC at Brigham and Women's Hospital in Boston.

Dr. Ronald M. Evans, professor and March of Dimes Chair in Molecular and Developmental Biology and Howard Hughes Medical Institute investigator at the Salk Institute for Biological Studies, has benefited from several NCRR resources, including laser and microscopy resources at the University of California campuses in Irvine and San Diego.

Dr. Jeffrey S. Flier is the George C. Reisman Professor of Medicine at Harvard Medical School and chief academic officer at Beth Israel Deaconess Medical Center, where he has relied on the GCRC to study the pathways that regulate metabolism and body weight and the role these pathways play in diabetes, obesity, and other conditions.

Dr. Ashley T. Haase is Regents' Professor, head of microbiology, and professor of medicine at the University of Minnesota Medical School. His studies of HIV pathogenesis and sexual mucosal transmission have depended in part on the resources and expertise at the NCRR-supported National Primate Research Centers in Wisconsin and New England.

Dr. Thomas R. Insel, director of the National Institute of Mental Health, studies the neurobiology of complex social behaviors in animals, including maternal behavior, pair bond formation, and aggression. He served as director of the Yerkes National Primate Research Center from 1994 to 1999.

Dr. Cynthia J. Kenyon is Herbert Boyer Professor of Biochemistry and Biophysics and director of the Hillblom Center for the Biology of Aging at the University of California, San Francisco. Her molecular studies of aging relied on specific strains of the roundworm *Caenorhabditis elegans,* obtained from the NCRR-supported Caenorhabditis Genetics Center at the University of Minnesota.

Dr. Margaret Pericak-Vance, James B. Duke Professor of Medicine and director of the Center for Human Genetics at Duke University Medical Center, studies factors that contribute to complex disorders like Alzheimer's disease. Her research has drawn on diverse NCRR-funded resources, including the Duke GCRC and the Human Genetic Analysis Resource at Case Western Reserve University.

Dr. Neil R. Powe is director of the Welch Center for Prevention, Epidemiology, and Clinical Research and professor of medicine, epidemiology, and health policy and management at Johns Hopkins University. He has used GCRC resources for his studies of patient outcomes, technology assessment, and cost-effectiveness analysis in many clinical areas.

Dr. Hugh A. Sampson, professor of pediatrics and director of the GCRC at the Mount Sinai School of Medicine, is one of the world's leading experts on food allergies. In the early 1990s, Dr. Sampson served as director of the NCRR-supported Pediatric Clinical Research Center at Johns Hopkins University.

Dr. Alan F. Schatzberg, professor and chair in the department of psychiatry and behavioral sciences at the Stanford University School of Medicine, studies the psychopharmacology of anxiety and depressive disorders. His studies of cortisol in anxiety and depressive disorders depended on GCRC support.

NCRR Releases New Fact Sheets

NCRR has issued two new fact sheets that provide overviews of the General Clinical Research Centers (GCRCs) and Biomedical Technology (BT) Resource Centers throughout the country. The *General Clinical Research Centers* fact sheet describes the history of the program, which began in 1959 to provide clinical investigators with the specialized research staff, instruments, and facilities needed to conduct sophisticated patient-oriented research. The fact sheet also outlines the GCRC management structure, the members of the research team and their roles, clinical research training programs, and instructions on gaining access to GCRCs for research.

The *Biomedical Technology Resource Centers* fact sheet describes the more than 40 specialized BT Resource Centers that develop and provide the scientific community with access to state-of-the-art instruments, methodologies, and computational tools that are not broadly available. Staffed by scientists who have expertise in technology and biology, the centers also create new tools for biomedical research and identify applications for these tools. The fact sheet summarizes the research, service, training, and dissemination components of the centers and also provides information about gaining access to or establishing new centers.

These and other fact sheets are available on NCRR's Web site at www.ncrr.nih.gov/publications.asp and can be obtained free-of-charge from the Office of Science Policy and Public Liaison, NCRR/NIH, 6701 Democracy Boulevard, Room 978, Bethesda, MD 20892-4874; phone: 301-435-0888; fax: 301-480-3558; e-mail: info@ncrr.nih.gov.

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