



NCRR: Catalyst for Discovery

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The Globalization of Research Resources

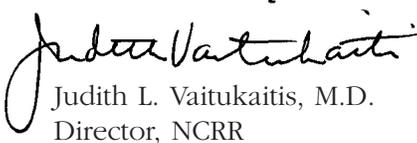
Some of the world's most pressing health concerns can best be addressed through international research efforts that integrate resources, technologies, and expertise from around the globe. An international approach is of critical importance in the fight against HIV/AIDS, malaria, hepatitis, and many other diseases and health disorders. The National Center for Research Resources (NCRR), with its cross-cutting mission, is ideally positioned to advance this evolving global approach.

One of the key elements in the international quest for an AIDS vaccine is an adequate supply of nonhuman primates to carry out testing. To help meet the need, the NCRR-supported Washington National Primate Research Center in Seattle is working in collaboration with the Primate Research Center at Bogor Agricultural University, Indonesia. Extensive breeding programs, research collaborations, and training programs benefit investigators in both countries. A similar arrangement—a Collaborative International Program in Primatology—is under development in Nepal. In addition, a joint project with NIH's Office of AIDS Research supports the training of scientists from India and China at several of the NCRR-supported National Primate Research Centers.

Another example of integrating research internationally involves the mouse model, which has gained new prominence in biomedical laboratories now that scientists can readily modify the animal's genome to create transgenic and "knockout" models of human disease. In 1999, NCRR established the Mutant Mouse Regional Resource Centers to expand the nation's capacity for preserving specialized mice and distributing them to biomedical researchers. (See "The Mouse Genome: Let the Sequel Begin," page 5.) Because of the program's success and value to the scientific community, NCRR now plans to extend the scope of the mouse resource to an international level. Collaborations will be established with Mutant Mouse Resources at sites in Europe and Japan, thereby minimizing unplanned duplicative efforts on a global scale.

In another effort to advance international research, NCRR, in concert with the Office of Science and Technology Policy, is sponsoring a meeting with the Office of Economic Cooperation and Development (OECD), an organization that assists countries with the economic, social, and governance challenges in a globalized economy. This spring, a subgroup of the OECD Working Party on Biotechnology, involving 40 countries, will develop recommendations for standards to assure the quality and maintenance of materials preserved in the global network of Biological Resource Centers.

These are just a few examples of how NCRR-supported resources are helping to advance the global research approach. The paradigm of the researcher working alone in an isolated lab has shifted to large interdisciplinary teams working together—across laboratories and now across national borders. NCRR will continue to bring researchers and resources toward a common international goal—enhancement of human health.


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

NCRR Reporter

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Cover: Drs. Leah Rae Donahue (left) and Muriel Davisson examine mice carrying a new spontaneous mutation. Although genetically engineered mice are often in the spotlight, mice with naturally occurring mutations remain valuable to biomedical studies. For 25 years, The Jackson Laboratory has housed the NCRR-funded Mouse Mutant Resource, devoted to investigating these animals. (Photo courtesy of The Jackson Laboratory)

••• NCRR Reports

••• *Research findings with NCRR support*

Exercise Reduces Dangerous Fat Levels

Fat deposited around the organs within the abdomen, so-called intra-abdominal fat, is associated with hypertension, cardiovascular disease, and type 2 diabetes, among other disorders. It is also the predominant type of fat that women tend to accumulate after menopause. Fortunately, a new study shows that regular exercise can reduce the levels of intra-abdominal fat in postmenopausal women.

For the study, which depended on the resources of the NCRR-supported General Clinical Research Center at the University of Washington, scientists at multiple sites recruited 173 sedentary, overweight, postmenopausal women. Half of the women were randomly assigned to an exercise group, which engaged in moderate-intensity exercise five days a week for a year, and half to a control group, which attended weekly stretching sessions for a year. Both groups were asked to maintain their usual diet.

After a year, the exercise group lost substantially more weight, total fat, and intra-abdominal fat than did the stretching group. The reduction in intra-abdominal fat was considerable and dose-dependent in the exercise group: the more diligently the women adhered to the fitness program, the more intra-abdominal fat they lost.

—*Journal of the American Medical Association* 289:323-330, 2003.

New Brain Circuit Regulates Hunger

Discovered just four years ago, the hormone ghrelin quickly gained attention because of its ability to

induce both animals and humans to consume large quantities of food. This and the fact that ghrelin levels in the blood rise right before mealtime have led to the theory that ghrelin may be part of the hunger signal that triggers eating. Produced predominantly in the stomach, ghrelin also is generated in the brain, although the hormone's production sites and functions in the brain were unknown.

Now NCRR-supported researchers at Yale University and the Oregon Health and Science University have identified ghrelin-containing cells in the rat brain in a structure called the hypothalamus, close to areas known



to be involved with feeding and metabolism. The investigators found that ghrelin not only promotes the release of neuropeptides that trigger eating but also inhibits the release of neuropeptides that suppress eating. Because the ghrelin-containing neurons are located in an area that receives input from brain regions that mediate circadian rhythms, the scientists suspect that the ghrelin circuit may serve as the interface between the brain's circadian clock and regions that regulate food intake and energy expenditure.

—*Neuron* 37:649-661, 2003.

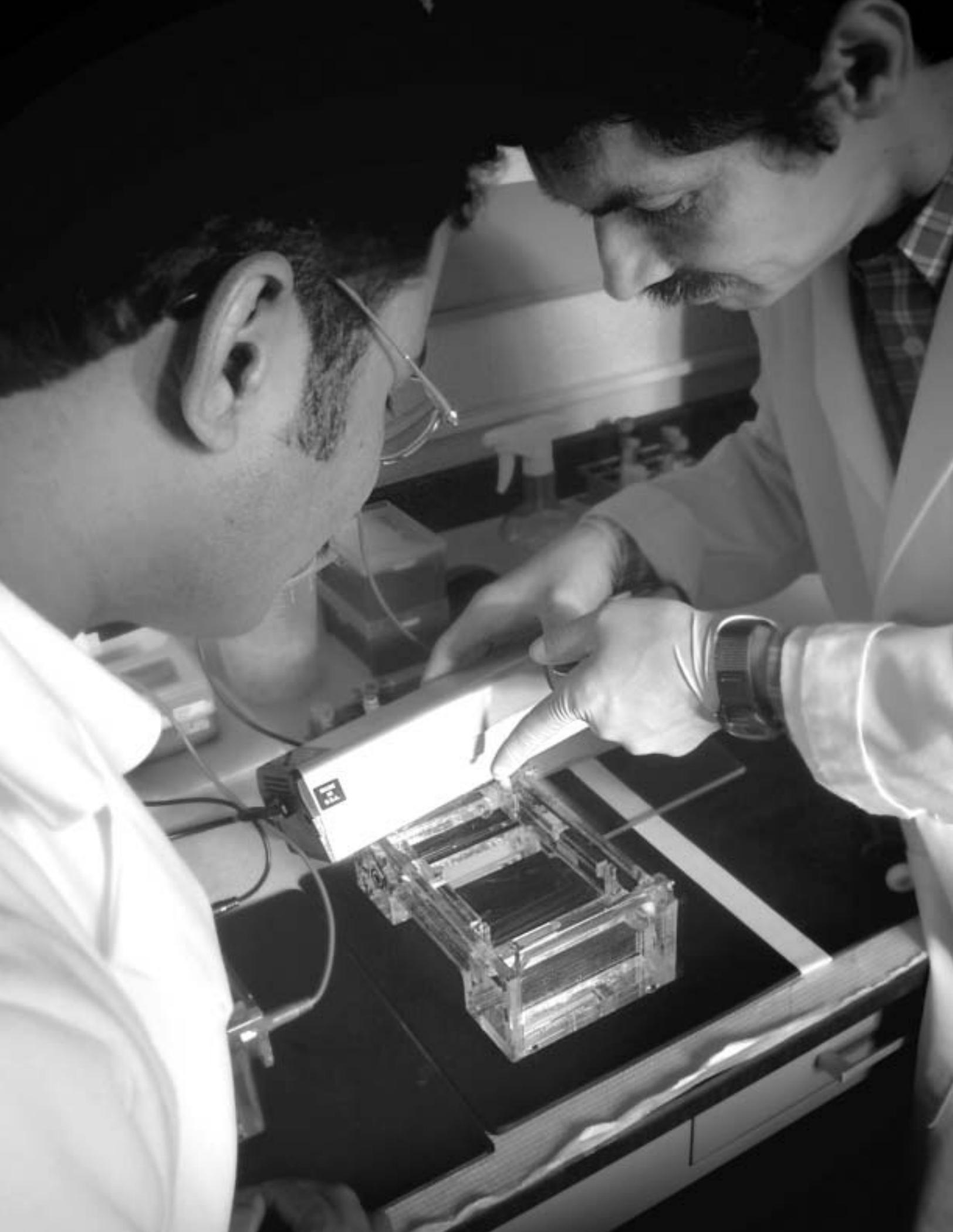
Antibodies Bend To Fit

Until recently, scientists assumed that an antigen attached to its corresponding antibody in a rigid manner, like the proverbial key fitting into a lock. However, a study from The Scripps Research Institute and the University of California at San Diego (UCSD) suggests that some antibodies may be more flexible when binding than previously thought.

The researchers used a technique called photon echo spectroscopy to study the binding-site dynamics of three antibodies that bind the same antigen—a chromophore named fluorescein. In this technique, a laser beam is used to excite fluorescein bound to an antibody, thereby knocking the antigen-antibody complex out of equilibrium. By measuring fluctuations in the light frequencies emitted by fluorescein as the system reestablishes equilibrium, scientists can determine how the antibody's shape changes during the process. Data analyzed at the NCRR-supported National Biomedical Computation Resource at UCSD showed that two of the three antibodies were extremely malleable as they interacted with fluorescein, while the third was relatively rigid. The findings provide support for the theory that antibodies may bind to more than one antigen by changing their shape to fit the antigen. This may help explain how a limited number of antibodies in the body can respond to a virtually unlimited number of foreign antigens on the surfaces of microbes, pollen, and tissue transplants.

—*Proceedings of the National Academy of Sciences USA* 100:92-97, 2003.

—Steven Stocker



The Mouse Genome

Let the Sequel Begin

by Tina Adler

In the 50 years since Drs. Francis Crick and James D. Watson announced the chemical structure of DNA to their colleagues at a local pub, geneticists have gone on to associate thousands of DNA mutations with human disease, in some cases improving therapies based on this knowledge. Behind many of these discoveries and accomplishments is a humble creature, the laboratory mouse, often lauded as the most important animal model for illuminating human biology. Last year, the Mouse Genome Sequencing Consortium published the first high-quality draft of the animal's genome in the December 5 issue of *Nature*. Researchers now are putting these hard-won data to use and, in doing

As the scientific demand for genetically engineered mice increases, so does the need for experienced mouse pathologists. An NCRR training program allows students like Dr. Prasad Nadella (left) to gain experience under the mentorship of a veterinary pathologist. Here he works with visiting assistant professor Dr. Ramiro Toribio (right) to examine an electrophoresis gel of DNA from genetically modified mice. (Photo by Jerry Harvey, Ohio State University)

so, are getting help from new and established mouse resources.

The draft genome made clear that, at the genetic level, mice and humans have a lot in common, including many more shared non-protein-coding sequences than previously expected, explains Dr. Eric S. Lander, one of the authors of the *Nature* paper and director of the Whitehead Institute's Center for Genome Research in Cambridge, Massachusetts. Knowing which genes are conserved in humans and mice "transforms the human sequence from simply a long laundry list of bases to the beginning of an annotated text," Dr. Lander asserts. The sequence also sheds light on gene structure, which will help scientists predict the consequences of creating mutations.

"Having the mouse sequence will allow us to eventually identify more of the genes that function both in humans and in mice," agrees Dr. Muriel Davisson, senior staff scientist at The Jackson Laboratory in Bar Harbor, Maine. "The next step is to figure out what each of those genes is doing, what its function is, and how that relates to human disease," she says.

To pursue all of these lines of inquiry, researchers will be enlisting

mutant mouse strains and the expertise of mouse pathologists, who can diagnose the effects of variant genes. They will also be looking for mouse specialists to house and phenotypically evaluate their new genetic resources.

NCRR's Division of Comparative Medicine has long supported mouse resources, including The Jackson Laboratory, which annually distributes millions of mutant mice to the scientific community. The laboratory was founded in 1929 by Dr. Clarence Little, the first researcher to create inbred strains of mice. He developed the C57BL/6J mouse strain, which was used for the genome sequencing. The Jackson Laboratory has grown considerably since Dr. Little's day, but it remains a highly controlled environment. To ensure the high health status of animal inhabitants, the laboratory houses the 80 or so new strains of mice donated to the laboratory every year in an "importation facility." Only their offspring enter the regular breeding rooms.

In 1992, in response to concerns from the scientific community regarding the cost, health, and distribution of genetically engineered mice, NCRR established funding for the Induced

Mutant Resource (IMR) at The Jackson Laboratory. Now home to almost 1,000 mutant mice, IMR selects, imports, cryopreserves, maintains, and distributes induced mutant strains. For 25 years, Jackson also has housed the NCRR-funded Mouse Mutant Resource, a repository for strains carrying spontaneous mutations. These mice either are donated by outside research groups or are culled from The Jackson Laboratory's own extensive breeding colonies, which can turn up about 20 mice with naturally occurring mutations each month. Jackson also has an NCRR-funded Special Mouse Strains Resource, which breeds animals for use in genetic analysis of complex traits.

Finally, The Jackson Laboratory houses the Informatics Coordinating Center, which runs an electronic network linking NCRR's Mutant Mouse Regional Resource Centers (MMRRCs) at the University of North Carolina at Chapel Hill; the University of California at Davis; Taconic Farms in New York; and Harlan Sprague Dawley, Inc., in collaboration with the University

of Missouri. MMRRCs, begun in 2001, are repositories for spontaneous and induced mutant mouse lines. They operate similarly to The Jackson Laboratory, in that researchers can donate their mutant strains for distribution, as well as acquire them. Each MMRRC facility is equipped to cryopreserve embryos or gametes,

techniques and results. They are also donating the strains to The Jackson Laboratory. "There has been quite a bit of interest expressed in Dr. Nadeau's strains," says Dr. Davisson.

Dr. Nadeau's team has succeeded in using these mouse strains to study the genetic susceptibility to testicular

• ***"There is a desperate need for both veterinarians and pathologists—experts who wouldn't mistake a mutant phenotype for a disease."***

rederive strains, and characterize the genetic and phenotypic makeup of the mutants. (For more information about the MMRRCs, see the Summer 2001 *NCRR Reporter*, pages 12-13.)

Another NCRR-supported resource, based at Case Western Reserve University in Cleveland, Ohio, specializes in developing mouse models of genetically complex human diseases. Headed by Dr. Joseph Nadeau, a professor in the department of genetics, with assistance from Dr. Lander, the resource has been instrumental in developing 21 strains of chromosome substitution mice, produced when the scientists swapped whole chromosomes between inbred strains. The resulting mouse "is just like the original strain except that it is inbred and homozygous for the substituted chromosome," explains Dr. Nadeau. Because the mouse differs in only one chromosome, researchers can trace any phenotypic differences between the animals to genes on that one chromosome, greatly simplifying their search for particular genes, Dr. Nadeau explains. He and his colleagues currently are writing a paper that describes their breeding

cancer, and they expect to have initial results soon for resistance to obesity. "We have identified the chromosomes that are involved, and now we are doing the crosses to localize and identify the genes," Dr. Nadeau says.

In addition to funding mouse researchers and repositories, NCRR provides grants to train mouse pathologists. "There is a desperate need for both veterinarians and pathologists—experts who wouldn't mistake a mutant phenotype for a disease," says Dr. Davisson. "The shortage is becoming more serious." Pathologists diagnose diseases, including the unexpected or subtle changes that occur when genes have been knocked out, mutated, or otherwise manipulated. Pathologists also help ensure the health of mouse facilities.

Since 2001, NCRR has awarded 10 grants for mid-career scientists to mentor young researchers—such as students, postdoctoral fellows, and junior faculty—in mouse pathology, says Dr. Franziska Grieder, a health scientist administrator in the NCRR Division of Comparative Medicine. These nonrenewable grants cover



Dr. Eric Lander helped propel the mouse genome sequence into the pages of *Nature* and into the hands of scientists.
(Photo by Justin Allardyce Knight)

50 percent of the grantees' salaries to free up their time for research and mentoring.

One recipient, Dr. Thomas Rosol, a veterinary pathologist at the Ohio State University in Columbus, has six graduate students studying mouse models of human disease. While his entire department provides the students' professional training, he oversees the students' research training. In one recent study, published in the February 27 issue of *Nature*, Dr. Rosol and colleagues at several institutions found an unexpected link between a mutation in a tumor-suppressor gene (Rb), resulting in an inherited form of eye cancer, and abnormal development of the placenta in mice. Dr. Rosol's group imaged and assessed the pathology of the placentas. Dr. Rosol is currently working with his students on three additional projects, each of which have direct implications for human health, including developing a mouse with the parathyroid hormone-related protein (PTHrP) gene knocked out in the mammary gland and a mouse model of hypercalcemia malignancy. A further project is examining how breast and prostate cancer metastasize to bone.

Besides his Mid-Career Investigator Award in Mouse Pathobiology Research from NCRR, Dr. Rosol also

used by him and his colleagues to track cancer cells in mice. "Now we can examine and measure bone metastases in living mice," Dr. Rosol says. To image cells, the device's camera captures light emitted from an enzyme, luciferase, that the cells are made to express.

Coming on the heels of the mouse sequence will be the release this spring of data revealing the sequence of the chimpanzee genome, which Dr. Lander anticipates posting to the Whitehead web site. Other animal genomes will follow. But researchers expect that the mouse will remain key to understanding what causes—and cures—human disease. Comparative genomics is the power tool of choice for unlocking the human genome, the Mouse Genome Sequencing Consortium asserts in its *Nature* paper, and one of the most powerful starting points for comparison is none other than the reliable laboratory mouse. Over the next



Dr. Thomas Rosol is training future mouse pathologists to work on mouse models of human disease. (Photo by Jerry Harvey, Ohio State University)

• **Comparative genomics is**
• **the power tool of choice for**
• **unlocking the human genome.**

relies on an NCRR training grant to provide stipends to veterinary students studying in his lab. Critical support for his efforts also comes from an NCRR Shared Instrumentation Grant, which enabled the purchase of an in vivo imaging system,

few years, with the mouse genome sequence in hand, Dr. Lander predicts that researchers will be able to "extract all the functional information from the genome, and have a wiring diagram of the cell."

The mouse resources described in this article are supported by the Division of Comparative Medicine of the National Center for Research Resources.

For more information about funding opportunities and resources supported by the Division of Comparative Medicine, see www.ncrr.nih.gov/comparative_med.asp.

Additional Reading

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Research Highlights

WISE Study Defines Women's Heart Disease

Heart disease, the leading cause of death in the United States, impairs both men and women, but its modus operandi differs in the two sexes. The disease can manifest similarly in both genders—for example, as symptoms of chest pain during exercise. The standard battery of cardiac tests does a good job of pinning down the cause in men: 80-90 percent prove to have ischemia, or inadequate blood flow due to narrowing of the coronary arteries. But in women, the cause of chest pain is more elusive. Only 60-70 percent of women with chest pain have significant narrowing of the coronary arteries, leaving 30-40 percent without an explanation for their symptoms—or treatment to relieve them.

“Research has done a good job of outlining heart disease in men. But we’re 50 years behind in actively studying women and heart disease,” observes Dr. Noel Bairey Merz, medical director and endowed chair of the Women’s Health Program, Preventive and Rehabilitative Cardiac Center, at Cedars-Sinai Medical Center in Los Angeles. Dr. Bairey Merz also is scientific chair for the Women’s Ischemia Syndrome Evaluation (WISE) study, a landmark investigation that seeks to close the gender gap in heart research.

Begun in 1996 with primary funding from the National Heart, Lung, and Blood Institute (NHLBI), the first phase of the WISE study enrolled almost 1,000



Multicenter studies led by Dr. Noel Bairey Merz identified aspects of heart disease unique to women. (Photo by Alan Braus, Cedars-Sinai Medical Center)

women between the ages of 20 and 60. Research was carried out at four sites—University of Pittsburgh; University of Florida, Gainesville; University of Alabama, Birmingham; and Allegheny General Hospital in Pittsburgh. Now entering its second phase, the WISE study has already generated more

than 40 publications on diverse topics, including the genetics of women’s heart disease; differences in electrocardiography between men and women; and the effects of phytoestrogens and “yo-yo” dieting on women’s heart health.

The WISE study reflects a trend in medical research that is expanding the definition of women’s health beyond reproductive health. According to Dr. Bairey Merz, “Studies of women’s health should look at women comprehensively and ask, Is it important to take the sex of the subject into consideration when it comes to screening, diagnosis, and treatment of disease? Pretty much everywhere we’ve looked, the answer has been yes.”

Developing better diagnostic tools is one of the goals of the WISE study. But Dr. Bairey Merz notes two further objectives. First, scientists hope to determine if women with symptoms of inadequate blood flow, but no actual coronary artery blockages, have a poorer long-term prognosis than other women. The second objective is to study the influence of reproductive hormones on the health of women’s hearts.

The WISE study enrolled women who had experienced symptoms of ischemia and were scheduled for, or already had undergone, coronary artery angiography, the gold standard for assessing coronary artery blockages. During the study, women also underwent additional diagnostic tests, including blood tests. The NCRR-supported General Clinical Research Center (GCRC) at Cedars-Sinai Medical Center “was really key for the WISE enterprise, serving as the administrative core for the WISE studies,” explains Dr. Bairey Merz. “It accepted all the blood samples, catalogued them, stored them, and sent them out. The GCRC allowed us to do core lab work for lipids, homocysteine, reproductive hormones, androgen hormones, and other blood markers studied by the WISE investigators.” The GCRC at the University of Alabama, Birmingham, also lent clinical support to the WISE study.

This type of support was essential to many WISE research discoveries, including the first evidence that low estrogen levels in premenopausal women may be a risk factor for heart disease, a finding that may help explain a traditional observation. “Overall, young women don’t have as many blockages as young men, but those few women who do have blockages have them in spades, and they have a higher mortality rate than young men,” says Dr. Bairey Merz. Noting that heart research has traditionally focused on women over 40, she adds, “One of the things we’re learning is that we cannot ignore even the 18-year-old. If she’s having chest pain, we have to pay attention.”

A second WISE effort examined how reproductive hormones are affected by the widely used cholesterol-lowering drugs known as statins, a concern because cholesterol is the body's chemical precursor for estrogen, progesterone, and testosterone. The researchers found that statins did not significantly alter levels of these hormones.

• ***“We’re 50 years behind in actively studying women and heart disease.”***

In another line of investigation, the researchers employed innovative magnetic resonance (MR) spectroscopy techniques to reach one of the key conclusions of the WISE study—that many women with ischemic symptoms, but no coronary artery blockages, do in fact have heart disease. The MR studies, led by Dr. Gerald Pohost, chief of the division of cardiovascular medicine at the Keck School of Medicine at the University of Southern California, employed technologies developed during the 1990s. As then-principal investigator of an NCCR-supported MR resource at the University of Alabama, Birmingham, Dr. Pohost used an experimental high-field (4.1-tesla) clinical MR system to develop noninvasive techniques for assessing heart health and function. The NCCR-supported studies also “paved the way for the commercial evolution of higher-field MRI systems, such as the 3-tesla MR scanners now entering clinical use,” says Dr. Pohost.

For the WISE investigations, MR spectroscopy was used in combination with one of the standard diagnostic methodologies, the cardiac stress test. In conventional stress testing, patients exercise on a treadmill while the heart is monitored by electrocardiography (EKG) and/or nuclear medicine perfusion scanning. In addition, an ultrasound technique—echocardiography—can be used in conjunction with pharmacologically induced stress caused by infusion of an agent that simulates exercise. “But these traditional diagnostic methods have been adopted largely from the changes one sees in men,” comments Dr. Pohost.

MR spectroscopy of phosphorus-31, a naturally occurring element, allowed researchers to measure the concentrations of two phosphorus-containing compounds in the heart muscle—phosphocreatine and adenosine triphosphate, critical molecules in the body's energy-storing biochemical pathways. Research with MR

spectroscopy had previously shown that the relative concentrations of these two compounds changed in a predictable way during exercise in women with coronary artery blockages. In the WISE study, MR spectroscopy was used to study women with ischemic symptoms but no blockages. “About 20 percent of these women had a significantly positive response, identical to the response of women with established coronary artery disease,” says Dr. Pohost. “This suggests there is inadequate blood supply during exercise.”

Follow-up WISE research lent further support to this theory. “Women with positive MR spectroscopy results were hospitalized significantly more frequently than those with negative results,” says Dr. Pohost. One explanation, he adds, is that these women may have an inadequate blood supply, but at a smaller caliber of arteries (the microvasculature) than can be visualized by traditional angiography.

Dr. Bairey Merz notes that characterization of microvascular dysfunction has become one of the main goals of the WISE study, now a year into its second phase. “Is this an early form of plugged-up arteries, or is this a totally different process? To treat it, we may need to identify a whole new line of receptors, because preliminary studies suggest that it's not related to cholesterol and cigarette smoking.” Dr. Bairey Merz comments that the results of the WISE study are helping to demonstrate the value of women's health research. “There's a gradual awareness that it's not that complicated, that we can do the research, and that, yes, it's worth it.”

—*William Oldendorf*

This research is supported by the Division of Clinical Research of the National Center for Research Resources and by the National Heart, Lung, and Blood Institute.

For more information about the NCCR Division of Clinical Research, see www.ncrr.nih.gov/clinical_rsrch.asp.

Additional Reading

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East Side Story: Encouraging Minority Scientists in Manhattan

Dr. Erich Jarvis says that when he used to visit high schools in New York City to talk to minority students about careers in biomedical science, the students would tell him that he didn't look like their idea of a scientist. "To them, a scientist was someone much older, gray, white, and with glasses," he says. Dr. Jarvis was, at the time, in his 20s, and his skin is dark. But he has been known to wear glasses.

∴ ***"The RCMCI Program has made a big impact on research at Hunter College."***

Today, Dr. Jarvis is 37 years old and an assistant professor in the department of neurobiology at Duke University in Durham, North Carolina, where he is a world-renowned authority on the neurobiology of bird song. And he is still defying people's notions about scientists. He is just as likely to be found in the dense forests of Brazil listening to the raspy squeaks of hummingbirds as in his laboratory measuring gene expression in the brains of songbirds.

As an undergraduate, Dr. Jarvis studied at Hunter College on the east side of Manhattan, where he used the resources of the college's Center for Study of Gene Structure and Function. The center is one of 19 such centers around the United States and Puerto Rico funded through NCCR's Research Centers in Minority Institutions (RCMI) Program. RCMI grants are provided to expand research capacity in colleges and universities that award doctoral degrees in the health professions or related sciences and have a 50 percent or greater enrollment of students belonging to ethnic groups underrepresented in biomedical sciences. Underrepresented groups include African Americans, Hispanics, Native Americans, Alaskan Natives, Native Hawaiians, and Pacific Islanders.

RCMI funding to Hunter—which last year totaled nearly \$2 million—is used to support eight core research facilities, such as the Sequencing and Separations Facility used by Erich Jarvis when he was a student in the laboratory of Dr. Rivka Rudner. "My project required DNA sequencing," says Dr. Jarvis. "Before the gene center was established, I was using old equipment, which was slowing me down. But after the gene center was set up, I was able to utilize new equipment at the Sequencing

and Separations Facility. My research just blossomed after that, and I was able to publish six papers as an undergraduate." At the time, his experiments usually involved ribosomal RNA (rRNA) genes in bacteria, which were the focus of Dr. Rudner's research. From this research came the discovery that the structure of rRNA genes could be used to differentiate bacterial species. "Ribosomal RNA genes are highly conserved," explains Dr. Rudner. "If you see a deviation in sequence, then you know there's been a real leap in evolution, because the ribosomal RNA genes are the last to change."

Dr. Rudner and her students proposed that knowledge of rRNA gene structure might be used not only for clarifying bacterial taxonomy but also for diagnosing bacterial diseases and studying bacterial epidemiology.

After leaving Hunter College, Jarvis traveled several blocks east to

Rockefeller University to work with Dr. Fernando Nottebohm, who studies vocal learning in birds. Vocal learning involves imitating the sounds of other creatures—for example, a songbird imitating the song of a neighboring songbird or a parrot imitating the words of its human owner. "There was a great amount of



Dr. Erich Jarvis holds a parabolic microphone, which he uses to record the songs of wild birds. By correlating singing behavior with gene expression patterns in different brain regions, Dr. Jarvis is helping to uncover the mechanisms of vocal learning in birds. The results may shed light on the brain mechanisms of language learning in humans. (Photo by Chris Hildreth, Duke University)

understanding of the brain systems involved in vocal learning but very little molecular biology,” says Dr. Jarvis. “So I decided to apply the techniques that I had learned at Hunter to vocal learning in birds, with the idea of one day eventually studying primates or humans.”

Using molecular biology, he and his colleagues were able to study gene expression in the brains of birds as they learned and produced songs. Measuring the expression of one gene in particular led to the identification of seven structures present in the brains of vocal-learning birds but lacking in birds incapable of such learning, such as chickens or pigeons. Dr. Jarvis thinks that similar structures may also exist in the brains of vocal-learning mammals—which include bats, cetaceans (whales and dolphins), and humans—and he speculates that studying vocal learning in birds may shed light on language learning in humans. “I no longer intend to quit studying songbirds, because we’re still learning so much from them. They’re really not so far away from mammals as one might think,” he says.

Last year, Dr. Jarvis received the Alan T. Waterman Award—an annual award from the National Science Foundation to a young scientist or engineer—for his work on bird song gene expression and neuroanatomy. Shortly after, Dr. Jarvis returned to Hunter College to speak at the first international symposium on bird song neurobiology, aptly titled “Singing in the Brain.” The conference was the 16th in a series of annual symposia held by the gene center and sponsored by NCRR.

Scientists have been paying increasing attention to the subject of bird song neurobiology since the discovery by Dr. Nottebohm and his colleagues that adult songbird brains can produce new neurons, contrary to the dogma that neurogenesis ceased at the end of brain development. This discovery has implications for repairing brains damaged by stroke, injury, and drug abuse, all of which are more prevalent among minority populations.

Dr. Robert Dottin, director of the RCMI program at Hunter, says he is actively trying to attract more minority students to the gene center, hoping to produce more graduates like Dr. Jarvis. Dr. Dottin also is making efforts to recruit a more diverse faculty. “We’re trying to make our faculty reflect the undergraduate student population, which is 50 percent minority, and so far, we’ve been very successful,” he says. RCMI funds help to recruit new scientists by partially financing the building of new laboratories for their research and providing salary support for up to 2 years. The funds also support pilot projects for all faculty, which may help them obtain research grants from other NIH institutes and centers, and other Federal and private sources.



Dr. Rivka Rudner discovered that the structure of ribosomal RNA genes could be used to differentiate bacterial species, which has implications for diagnosing bacterial diseases and for studying bacterial evolution and epidemiology. As an undergraduate, Erich Jarvis learned molecular biology in Dr. Rudner's laboratory. (Photo by Ana Golici, Hunter College)

“On the whole, the RCMI Program has made a big impact on research at Hunter College,” says Dr. Dottin. “Without it, it would have been impossible for us to have our outstanding research program, which has attracted the attention of scientists worldwide.”

—*Steven Stocker*

For more information about the Center for Study of Gene Structure and Function, visit the web site at <http://sonhouse.hunter.cuny.edu/genecenter> or contact Dr. Robert Dottin at Hunter College, City University of New York, 695 Park Ave., New York, NY 10021; phone: 212-772-5171; fax: 212-772-5466; e-mail: dottin@genectr.hunter.cuny.edu.

For more information about the Research Centers in Minority Institutions Program, see www.ncrr.nih.gov/resinfra/ri_rcmi.asp.

Critical Resources

MRI Provides New Views of the Brain

Since its debut a quarter century ago, magnetic resonance imaging (MRI) has become a remarkably versatile technology for studying the brain. Using various MRI techniques, scientists can see brain structures, identify areas of neuronal activity, or study regional differences in brain chemistry. At the Resource for Quantitative Functional MRI, located at the Kennedy Krieger Institute and Johns Hopkins University in Baltimore, scientists are using different types of MRI to investigate diseases ranging from the rare disorder X-linked adrenoleukodystrophy to the more common cerebral palsy. They also are developing innovative new methods to visualize axonal pathways, display MR images, and detect patterns of brain activity.

Producing MR images involves establishing magnetic fields around a patient and then introducing radiofrequency pulses, which cause hydrogen atoms in water molecules to emit their own detectable radio waves. Computers then translate these emitted waves into images. By varying parameters of the pulses and magnetic fields, scientists can create images that highlight different brain constituents. “There are thousands of different ways to produce MR images, and each method sensitizes different aspects of the tissue,” says Dr. Peter van Zijl, principal investigator of the NCRN-supported resource.

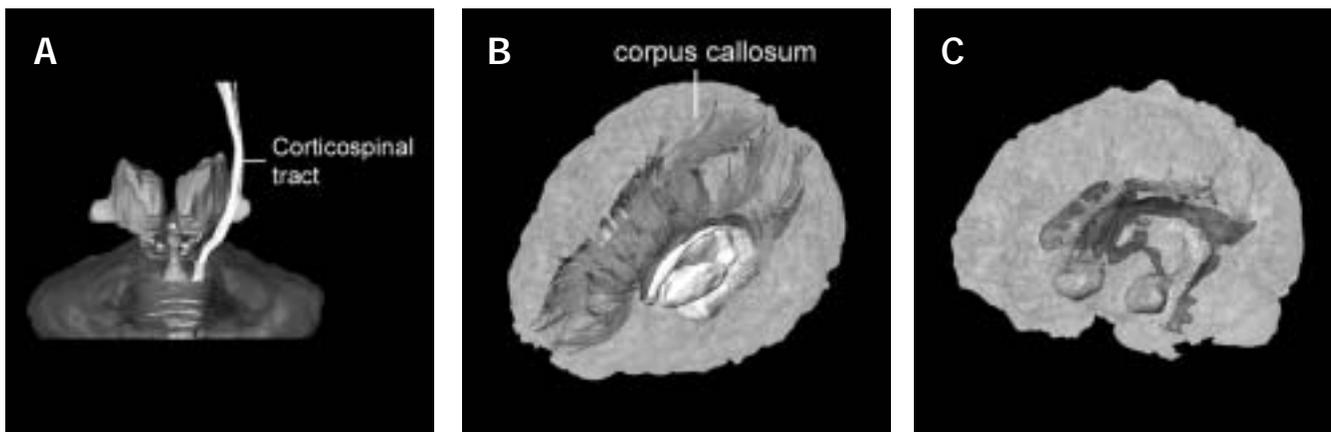
For example, functional MRI (fMRI) highlights blood oxygenation changes that accompany neuronal firing,

In the fMRI division of the resource, led by Dr. James Pekar, scientists are using a new data analysis method called independent component analysis (ICA) to help make sense of fMRI signals produced when the brain is processing complex information. With ICA, researchers are able to identify brain regions that operate together to perform certain functions—in some cases, detecting active regions missed by the conventional method for analyzing fMRI data.

“The conventional method basically just tests hypotheses,” explains Dr. Pekar. “But that approach is like going out in the woods and only looking for deer. There also might be moose or elk or lions or tigers walking by, but you won’t see them because you’re entirely focused on finding deer. In contrast, ICA asks, ‘What’s in the data?’ It’s like going out in the woods without any preconceived notions about what you’ll find, and that lets you see the lions and tigers and all the other animals, including the deer.”

Recently the scientists used ICA to determine which brain regions became active when volunteers played a video game of simulated driving. Among the regions that lit up were those that maintain vigilance, detect errors, and inhibit behaviors. Surprisingly, as volunteers drove faster in the video game, activity in these regions decreased. Dr. Pekar says that one interpretation of this finding is that to perform a risky behavior, such as driving fast, a person has to inhibit those parts of the brain that say, “Don’t do that! It’s risky!”

Another form of MRI used at the resource is diffusion-tensor imaging (DTI), which highlights water diffusion in the brain. Dr. Susumu Mori, who heads the DTI division,



With diffusion tensor imaging, scientists can produce brain images that highlight specific axonal pathways, almost as if they had been dissected from the brain. Figure A shows the corticospinal tract, which transmits motor information from the cerebral cortex to the spinal cord, in a brain image lacking all of the cerebral cortex and most of the brainstem. Figure B shows the corpus callosum, which transmits information between the two cerebral hemispheres. Figure C throws into relief the limbic system, which mediates emotions. (Courtesy of Dr. Susumu Mori, Johns Hopkins University)

and his colleagues discovered that the technique can be used to visualize major axonal pathways because many of the water molecules in the brain diffuse longitudinally inside axons. This is the first technique to noninvasively examine axonal tracts in living people. Recently Dr. Mori and other scientists used DTI to examine axonal pathways in the brains of two children with cerebral palsy. They found a reduction in the number of fibers that transmit

• ***Diffusion-tensor imaging is the first technique to noninvasively examine axonal tracts in living people.***

sensory information to the cerebral cortex, indicating that deficits in sensory processing may be partly responsible for the abnormal movements and increased muscle tone characteristic of this disease.

A third resource division, led by Dr. Peter Barker, uses MR spectroscopy and a related technique called MR spectroscopic imaging (MRSI) to obtain estimates of the levels of certain metabolites, such as the neuronal marker N-acetylaspartate. Among the diseases being studied at the resource using MRSI is X-linked adrenoleukodystrophy (X-ALD), a rare genetic disorder in which very long chain fatty acids (VLCFAs) accumulate in the blood and later the brain, causing myelin breakdown. Symptoms typically appear by age 5 or 6 and include impairments in cognition, vision, and movement. The MRI studies are being conducted in conjunction with a clinical trial of Lorenzo's Oil, an oil mixture that normalizes VLCFA blood levels. The study, led by Dr. Hugo Moser, is designed to test whether the oil therapy and a restrictive diet can prevent symptom onset in boys carrying the X-ALD gene by preventing VLCFA buildup in the brain. The NCCR-supported General Clinical Research Center (GCRC) at Johns Hopkins University is lending critical resources to the clinical trial, including the GCRC personnel who prepare the specialized meals, monitor the boys' growth and development, and conduct tests of body composition.

Trial results to date have shown that the treatment and diet regimen reduced the chances of developing neurological symptoms by two-thirds. The researchers also discovered that MRSI could detect developing brain deformities before they became visible in structural MRI images. According to Dr. Moser, MRSI might prove useful for identifying potential candidates for bone marrow transplantation, which can be an effective

therapy for X-ALD if performed early enough in the disease.

In a fourth resource division, Dr. Michael Miller is developing standardized methods for displaying MR images so that ready comparisons can be made between brains of different sizes and between brains from different species. The methods also will allow comparisons among brain images taken in various laboratories that

perform MRI in different ways.

In developing these methods, Dr. Miller is working with other researchers associated with the Biomedical Informatics Research Network (BIRN), of which the Resource for Quantitative Functional MRI is the newest member. BIRN is

an NCCR-funded project that links biomedical laboratories around the country in a high-performance computer network that enables the rapid sharing of data, including MR images. This allows scientists at universities nationwide to collaborate on studies almost as easily as if they were in the same location.

To enhance its MRI capabilities, the resource recently obtained a 3-tesla MR scanner, using funds from the resource grant and an NCCR Shared Instrumentation Grant. Compared to the 1.5-tesla scanner that the resource has been using, the new 3-tesla scanner induces stronger signals from the body, which translate to better spatial resolution in the images. It also enables images to be obtained in less time, which is particularly useful with very young patients, since children often have difficulty lying motionless in the scanner for long periods.

Prospective users of the resource are asked to submit a resource utilization form, which is reviewed by a committee of resource staff. "If the proposal is high quality and fits in well with what we are doing, we generally accept it," says Dr. van Zijl.

—*Steven Stocker*

For more information about the Resource for Quantitative Functional MRI, visit the web site at mri.kennedykrieger.org or contact Dr. Peter van Zijl at the Kennedy Krieger Institute and Johns Hopkins University, 707 North Broadway, Baltimore, MD 21205; phone: 443-923-9500; fax: 443-923-9505; e-mail: pvanzijl@mri.jhu.edu.

The Resource for Quantitative Functional MRI is supported by the Division of Biomedical Technology of the National Center for Research Resources. To learn more about other NCCR-supported biomedical technology resources, see www.ncrr.nih.gov/biomedical_tech.asp.

SPF Monkeys To Support AIDS Research

To ensure that NIH-funded scientists have access to the animal resources needed for AIDS research, NCRR has awarded six new cooperative agreements to primate centers in the United States and Puerto Rico. As part of the agreements, the institutions will breed specific-pathogen-free (SPF) macaque monkeys that are free of three common retroviruses that can interfere with AIDS experiments—simian immunodeficiency virus (SIV), Type D simian retrovirus, and simian T-lymphotropic virus. The monkeys will also be free of herpes B-virus, which on rare occasions has infected and killed human caretakers. In addition, some of the macaques will carry genes for certain major histocompatibility complex (MHC) molecules that have been shown to intensify the immune response to SIV. (For more information about the MHC molecules, see the *NCRR Reporter*, Fall 2000, pages 8-9.)

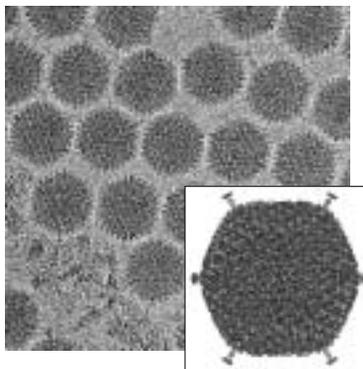
The new awardees are the Caribbean Primate Research Center Program in Puerto Rico and the National Primate Research Centers (NPRCs) in California, Georgia, Louisiana, Oregon, and Washington. Besides breeding SPF monkeys, some centers will also conduct research on breeding and diagnostic screening methods. For example, the California NPRC is developing a single blood assay for detecting multiple viruses, which will enable high-throughput screening at reduced cost.

New Resource Deciphers Macromolecular Structures

Cryo-electron microscopy is useful for determining the structures of large macromolecular assemblies like ribosomes, but the technique is time consuming and labor intensive. To speed up the process and make it easier to use, the new National Resource for Automated Molecular Microscopy (NRAMM) has been established at The Scripps Research Institute in La Jolla, California. The goal of this NCRR-supported resource is to provide an integrated system that generates detailed three-dimensional (3-D) reconstructed images, including molecular-scale details, through a largely automated process.

To achieve this goal, the resource is launching four core projects. The specimen-handling project will develop a robot to transfer specimen grids to and from the

microscope, a system to monitor cryostage temperature, and a system to automatically replenish cryogens in the cryostage during long-term operation. A second project involves automating the process of image acquisition. For this project, the researchers have developed a prototype system that acquires images of helical filaments; they are now extending this system to automate data collection for a wide variety of specimens. A third project, which centers on information handling, involves designing a database capable of storing images and parameters. The database will be available on the NRAMM web site, along with a library of tools for querying the database. The fourth project focuses on developing a system to automatically identify a segment in a high-magnification image of an object (such as a macromolecular complex or virus), analyze that segment, and then produce a detailed 3-D reconstruction of the object. For example, the larger picture shown below is a high-magnification cryo-electron micrograph of multiple adenovirus particles, while the insert is a computer-generated rendering of an average of these particles, showing its structure in greater detail than that seen in the micrograph.



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NRAMM welcomes applications for both collaborative and service projects. As part of its training mission,

NRAMM will be hosting an intensive course on cryo-electron microscopy from November 12 to 20, 2003. More information about the resource can be found on the NRAMM web site at <http://nramm.scripps.edu>.

Diabetic Mouse Repository Established

Developed in 1980 in Japan, the nonobese diabetic (NOD) mouse strain has proven to be an excellent animal model for insulin-dependent, or type 1, diabetes mellitus. NOD is the only mouse strain that spontaneously develops type 1 diabetes, complete with certain characteristics of the human disorder, including autoimmune responses against pancreatic islets and a reduction in the number and size of islets. Several

strains of genetically engineered mice have been derived from the original NOD strain for the purpose of studying diabetes-related genes. To cryopreserve strains that are important for diabetes research but are not used extensively, NCCR has joined with the National Institute of Diabetes and Digestive and Kidney Diseases to establish a Type 1 Diabetes Repository at The Jackson Laboratory in Bar Harbor, Maine.

"Some strains are in enough demand by researchers that it's cost-effective to keep breeding them. The other strains are important to individual scientists but are unlikely to be requested in sufficient numbers to make breeding them cost-effective. The repository was created to ensure that about 150 of those strains are cryopreserved," explains Dr. David Serreze, staff scientist at The Jackson Laboratory and coscientific supervisor of the repository.

Periodically, the repository brings strains with related mutations out of cryopreservation and breeds them for live distribution in what are called "functional waves." For example, the current wave consists of mice with different NOD chromosomal regions that affect diabetes susceptibility. "If the demand for any strain becomes large enough when we release it in a functional wave, then The Jackson Laboratory will continue to make the strain available for live distribution," says Dr. Serreze. More information about the Type 1 Diabetes Repository can be found at www.jax.org/t1dr/about.html.

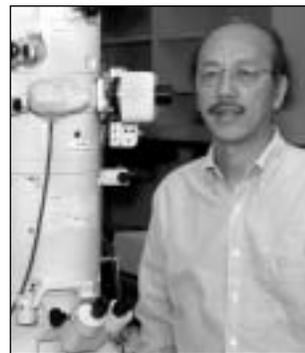
Four Appointed to NCCR Advisory Council

The National Advisory Research Resources Council has gained four new members, each a leader in the scientific community, who will advise NCCR on policies and programs and perform second-level peer review of grant applications. The new appointees are:

Dr. Robert J. Beall, president and chief executive officer of the Cystic Fibrosis Foundation in Bethesda, Maryland. Dr. Beall is known for his innovative approach to research and education, which has guided the foundation's research development program.



Dr. Wah Chiu, Alvin Romansky Professor of Biochemistry at the Baylor College of Medicine in Houston, Texas. Since 1985, Dr. Chiu has directed the NCCR-supported National Center for Macromolecular Imaging, which develops cutting-edge computational and visualization techniques.



Dr. Catherine C. Fenselau, professor of chemistry and biochemistry at the University of Maryland. An expert in mass spectrometry and its biological applications, Dr. Fenselau focuses her current research on proteomics and the detection of biological warfare agents.

Dr. Joan S. Hunt, University Distinguished Professor in the Department of Anatomy and Cell Biology and the Department of Pathology and Laboratory Medicine at the University of Kansas Medical Center in Kansas City. Dr. Hunt is the principal investigator for the NCCR-supported Kansas Biomedical Research Infrastructure Network, a program that links research universities, undergraduate campuses, and a tribal college in an effort to inspire students across the state to pursue careers in biomedical research.



NCRR on the Move

As of **April 14**, all NCRR offices formerly located at 6705 Rockledge Drive in Bethesda, Maryland, have relocated. Our new address is:

**One Democracy Plaza
6701 Democracy Boulevard MSC 4874
Bethesda, MD 20892-4874**

Staff members' e-mail addresses, phone numbers, and fax numbers remain unchanged.



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