INVESTING IN THE FUTURE OF SCIENCE

The First Five Years of the Career Awards Program

May 2000

BURROUGHS WELLCOME FUND

The Burroughs Wellcome Fund would like to acknowledge the contributions of Tom Burroughs in developing the idea for this publication and of Dennis Meredith for research and writing support.

The Fund would also like to thank all of the Career Award recipients who gave of their time to provide information about themselves and their research projects. Their enthusiasm and willingness to help were indispensable in producing this report.

The Fund acknowledges Georgine M. Pion, Ph.D., Vanderbilt University, and Martin Ionescu-Pioggia, Ph.D., Program Officer, for their work in evaluating the progress of career awardees.

Finally, Rolly Simpson, Program Associate; Carr Agyapong, Senior Program and Communications Officer; Martha Peck, Vice President for Programs and Communications; and Enriqueta Bond, Ph.D., President, provided information, feedback, and suggestions that immeasurably improved the final product. Thank you.

Karyn Hede, Communications Officer
Editor

Cover:
Depicted in BWF's logo, the eye of the ancient god Horus is widely considered a symbol of health.

About the Burroughs Wellcome Fund

The Burroughs Wellcome Fund is an independent private foundation dedicated to advancing the medical sciences by supporting research and other scientific and educational activities. It conducts the majority of its grantmaking through competitive programs designed to support the career development of young scientists and to build capacity in research areas BWF believes to be undervalued or in need of targeted support. The Fund's current assets are in excess of $700 million, with about $47 million in grants awarded in 1999. BWF has no affiliation with any corporation. For more information about the Fund, or to view this publication on-line, visit our Web site at www.bwfund.org.

Burroughs Wellcome Fund
21 T. W. Alexander Drive
Post Office Box 13901
Research Triangle Park, NC 27709
Telephone (919) 991-5100
Fax (919) 991-5160

Acknowledgments

Table of Contents

Career Awards Advisory Committee

Mina J. Bissell, Ph.D.
Director, Life Sciences Division
Lawrence Berkeley National Laboratory

Pamela J. Bjorkman, Ph.D.
Associate Investigator, Howard Hughes Medical Institute
Associate Professor of Structural Biology
California Institute of Technology

Patricia K. Donahoe, M.D.
Chief, Pediatric Surgical Services
Director, Pediatric Surgery Research Laboratories
Massachusetts General Hospital

Uta Francke, M.D.
Investigator, Howard Hughes Medical Institute
Professor of Genetics
Stanford University Medical Center

Elaine Fuchs, Ph.D.
Investigator, Howard Hughes Medical Institute
Amgen Professor of Basic Sciences
University of Chicago

Phil Gold, M.D., Ph.D. (co-chair)
Executive Director, Clinical Research Center
Montreal General Hospital

Ashley T. Haase, M.D.
Professor and Head, Department of Microbiology
University of Minnesota Medical School

Eric R. Kandel, M.D.
Senior Investigator, Howard Hughes Medical Institute
University Professor, Department of Psychiatry
Columbia University College of Physicians and Surgeons

Stanley J. Korsmeyer, M.D.
Director, Program in Molecular Oncology
Dana-Farber Cancer Institute
Sidney Farber Professor of Pathology and Professor of Medicine
Harvard Medical School

Martin M. Matzuk, M.D., Ph.D.
Stuart A. Wallace Professor of Pathology
Baylor College of Medicine

I. George Miller, M.D. (through 3/2000)
John F. Enders Professor of Pediatric Infectious Diseases
Professor of Epidemiology and Molecular Biophysics and Biochemistry
Yale University School of Medicine

Mary Lou Pardue, Ph.D. (through 3/2000)
Boris Magasanik Professor of Biology
Massachusetts Institute of Technology

Donald W. Pfaff, Ph.D. (co-chair)
Professor of Neurobiology and Behavior
Rockefeller University

Jerome F. Strauss III, M.D., Ph.D.
Luigi Mastroianni Jr. Professor
Director, Center for Research on Reproduction and Women's Health
University of Pennsylvania Health System

Ian A. Wilson, D.Phil.
Professor of Molecular Biology
Chair, Structure and Chemistry Affinity Group
Scripps Research Institute

James M. Wilson, M.D., Ph.D.
Director, Institute for Human Gene Therapy
University of Pennsylvania Health System

Career Awards Program Officers

Martin Ionescu-Pioggia, Ph.D.
Program Officer

Rolly Simpson
Program Associate

Burroughs Wellcome Fund Board of Directors

Mary Ellen Avery, M.D.
Thomas Morgan Rotch Distinguished Professor of Pediatrics, Emeritus
Harvard Medical School

Enriqueta C. Bond, Ph.D.
President
Burroughs Wellcome Fund

Gail H. Cassell, Ph.D.
Vice President, Infectious Diseases
Drug Discovery Research and Clinical Investigation
Lilly Research Laboratories
Eli Lilly and Company

Stephen D. Corman, C.P.A.
Founder, Former Chair and Chief Executive Officer
Pharmalink Inc.

Henry G. Friesen, M.D.
President
Medical Research Council of Canada

David M. Kipnis, M.D. (chair)
Distinguished University Professor of Medicine
Washington University School of Medicine

I. George Miller, M.D.
John F. Enders Professor of Pediatric Infectious Diseases
Professor of Epidemiology and Molecular Biophysics and Biochemistry
Yale University School of Medicine

Joseph S. Pagano, M.D.
Lineberger Professor of Cancer Research
Director Emeritus, Lineberger Comprehensive Cancer Center
University of North Carolina-Chapel Hill School of Medicine

Mary-Lou Pardue, Ph.D.
Boris Magasanik Professor of Biology
Massachusetts Institute of Technology

Jerry L. Whitten, Ph.D.
Professor of Chemistry
North Carolina State University

Jean D. Wilson, M.D.
Charles Cameron Sprague Distinguished Professor of Biomedical Science
University of Texas Southwestern Medical Center-Dallas

Career Awards Advisory Committee/Program Staff/ BWF Board of Directors

In the 1950s, Arthur Kornberg was a young biochemist with an insatiable curiosity about an obscure substance, which we now know as DNA. At the time, DNA was a novelty. Scientists knew it was involved in heredity, but had no idea how it carried genetic information. For a beginning scientist, working on DNA was exciting, but risky. It certainly didn't seem to have practical benefits. But Kornberg was onto something big. Within a few short years, he discovered several key proteins that make exact copies of DNA, a feat no one could have imagined when Kornberg started his work. The discovery garnered him a Nobel Prize in 1959 at the age of 41 and helped launch the age of biotechnology.

These were heady days, when young molecular biology students learned at the hand of an experienced mentor and could then expect to move on rapidly to their own laboratories. Scientists like Kornberg and the generation of young scientists starting their careers at that time could go as far as their intellect and imaginations took them, with the help of ample research grants provided by the federal government.

Today, the biomedical research enterprise is strong and prolific, having built upon the foundation laid by scientists of Kornberg's generation. But times have changed for today's rising scientists. No longer can a talented scientist necessarily expect to become independent simply by completing the academic requirements for the doctoral degree, applying for grants, and publishing research papers.

As Kornberg himself pointed out in a recent speech, "The overriding issue in biomedical science, as I see it, is how to give our abundant scientific talent the resources to exploit the extraordinary new technologies to advance knowledge. Currently, a pervasive mood among productive biomedical scientists makes them fear for continued grant support, persuades them to choose safe and practical projects over the untried and adventurous, and tempts their interest in commercial ventures. This is clearly a state that discourages young people from entering science and drives others to abandon science."[1]

At the Burroughs Wellcome Fund (BWF), we have always believed that to maintain the strength of the biomedical research enterprise, we must build for the future by investing in people. Our long-time board members Dr. George Hitchings and Gertrude Elion, who shared the Nobel Prize in Physiology or Medicine in 1988, maintained that a small amount of money given at the right time to young researchers can have a catalytic effect on their scientific productivity and their careers.

Our philosophy has always been that we are funding a person, not just a project.

[1] Kornberg, Arthur. March 13, 1997. "Basic Biomedical Research Support." Speech given at the Conference on the Future of Biomedical Research sponsored by Federation of American Societies for Experimental Biology, The Brookings Institution, and the American Enterprise Institute.

INTRODUCTION: *Encouraging Research Careers*

BWF has dedicated itself to supporting the women and men who will be the next generation of George Hitchings, Gertrude Elions, and Arthur Kornbergs – scientists who will chart the course to a healthier nation and world.

Thus, in 1995 BWF launched its Career Awards in the Biomedical Sciences to identify highly talented scientists during their formative periods and provide them the support they need to become independent investigators.

The Career Awards Program offers up to six years of salary and research support for biomedical scientists during their advanced postdoctoral training and initial faculty years. Each year we award about 25 grants of about $500,000 each. (Further details may be found on our Web site at wwww.bwfund.org.)

We view this funding as venture capital that gives scientists early in their careers the freedom to choose their own research directions and to demonstrate the promise of those new directions to prospective employers and funding agencies.

This report highlights some of the insights and experiences of our awardees and staff in a program now approaching five years of operation.

In this report we:

- share the insights we have gained in administering the program in its first five years;
- introduce the innovative work of a cross-section of our career awardees;
- highlight some of the career issues that these scientists face.
- provide a summary of career management topics selected by awardees themselves and addressed at the most recent convocation of Career Awards recipients.

In 1999, we reached the milestone of 100 career awardees and $50 million in grant awards. Many of these scientists attended our Career Awards meeting held in Coronado, Calif., July 22 - 24, 1999. Such meetings, held every two to three years, offer our awardees a unique, supportive group environment in which to ask questions, network with one another, and receive mentoring advice from senior scientists.

Early career support is critical, in particular, for physician-scientists, given the many clinical obligations that create a real test of commitment to find time for basic research.

In talking to awardees, we found that most are comfortable and confident in their scientific work. Many, however, are missing a close, productive mentoring relationship, or if they are lucky enough to have found guidance from an experienced scientist, are still lacking key information about how to handle critical issues as their careers progress.

Our philosophy has always been that we are funding a person, not just a project. To reflect this belief, we designed our award policies to be highly flexible. Recipients may change projects or institutions during the period of their awards and may apply their funds to equipment, supplies, travel, or personnel, as they deem appropriate. We see the awards as complementing, not competing with, other support these scientists might attract.

In addition to providing funds, we help to restore some of the mentoring that so many young scientists report is lacking in today's research environment. Besides providing access to board members and advisory committee members, we also help them learn critical mentoring skills early in their

careers, so they in turn can support and nurture the next generation of scientists. By convening awardees, we provide an opportunity to build a network of professional connections that can provide support, advice, and scientific collaborations. To extend that access beyond the boundaries of a meeting, we provide funding for career development projects such as Science's Next Wave, a Web site dedicated to providing career development assistance and information to large numbers of post-docs and young faculty worldwide (www.nextwave.org).

Such early career support is critical, in particular, for physician-scientists, given the many clinical obligations that create a real test of commitment to find time for basic research. Given the patient-centered training of physician-scientists, we believe they can make substantial contributions to both basic science and to the translation of fundamental discoveries to clinical applications. Career Awards protect 80 percent of their time during the first three years of a faculty appointment, allowing physician-scientists to get a jump-start on research early in their careers, and making continuing on a research track a more viable career option. We also fund a select number of young researchers each year in reproductive science to encourage what we feel is a woefully under-emphasized field relative to its scientific and clinical importance and in which funding is becoming more difficult to obtain.

We help to restore some of the mentoring that so many young scientists report is lacking in today's research environment.

Our Career Awards Program is relatively modest, given the enormous scope of the nation's research enterprise and its great need for future leaders. However, we believe we have significantly aided the early careers of an important cadre of highly promising scientists, as well as learned valuable lessons about the policies and programs that are most effective in promoting career development.

Do we have another Arthur Kornberg, Gertrude Elion, or George Hitchings in our midst? Only time will tell, but for a select group of scientists, we can say that BWF is providing the means to find out. ■

Combining Forces to Foil Brain Cancer

James Olson, M.D., Ph.D. and Matthias Gromeier, M.D.

The childhood brain tumors called medulloblastomas remain frustratingly enigmatic — that is, oncologists cannot predict whether a given cancer will prove to be relatively slow-growing or aggressively malignant.

However, modern gene technology is beginning to reveal genetic differences among tumors that can predict how they might behave or respond to therapy.

Dr. James Olson and patient.

James Olson is applying that technology to studying medulloblastoma, and his discoveries are raising the possibility of more accurate prognosis and even potential new treatments for a cancer that kills the vast majority of children that it strikes.

Olson is using silicon wafers much like the processor chips in computers, but instead of electronic circuits these wafers, dubbed "gene chips," hold an array of DNA. When genetic material derived from a tumor is applied to the chip, the genes stick to their complementary gene sequences on the chip, and indicator molecules allows identification of those genes. The gene chips can measure the activity of thousands of medulloblastoma tumor genes at once, quickly detecting which ones are turned on in the cancer cells.

"Before these micro-arrays, it might take a year to measure a small number of genes in a dozen tumors," says Olson, who is at the University of Washington School of Medicine. With the micro-arrays Olson was able to survey 6,800 genes in a dozen medulloblastoma tumor samples and identify a group of genes that appeared to be important in the cancer's renegade growth.

In particular, Olson chose tumors that make a particular transcription factor, a type of molecular switch that turns on other genes. The gene that encodes this switch, called NEUROD3, is usually active in normal developing nerve cells called stem cells.

"Several years ago we showed an association between NEUROD3-positive tumors and tumors that were metastatic, or progressed rapidly," says Olson. He reasoned that other active genes typical of primitive nerve cells would also be

found in these tumor cells. This primitive state, they believed, would partially explain the aggressive behavior of these tumors.

Further analyses of the active genes, however, gave a puzzling result. One of the active genes encoded a protein named CD155. What puzzled Olson was that CD155 is known as the molecule that poliovirus uses to enter brain cells. It is a receptor that sits on the surface of cells and responds to chemical signals from the surrounding environment.

"I had difficulty interpreting this finding, because it didn't make any sense to me how that would be biologically significant," recalls Olson.

However, fellow Career Award recipient Matthias Gromeier, M.D., noticed CD155 listed in Olson's poster display of research results at a Burroughs Wellcome Fund meeting of Career Award recipients and immediately recognized its significance.

Says Olson, "Matthias explained that CD155 is a receptor that's turned on in developing primitive brain cells, and that it allows polio virus to cause polio in older kids."

> ***"By identifying genes like CD 155 that are preferentially expressed in the NEUROD3-positive tumors, we might be able to identify new forms of therapy that are directed at the most aggressive medulloblastomas."***

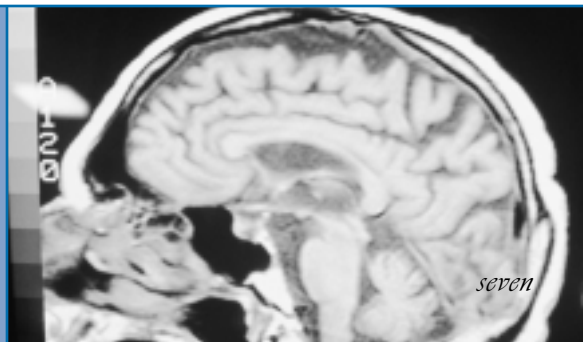
Particularly exciting was that Gromeier's research dovetailed with his own. Gromeier had genetically altered a poliovirus to target and kill CD155-positive cancer cells. He had removed from the poliovirus the dangerous genes that allow it to damage normal nerve cells, replacing them with genes from the common cold virus. The resulting engineered virus was able to kill glial brain tumor cells that expressed the CD155 receptor. However, the prospect of using the engineered poliovirus against the deadly medulloblastoma offered an extraordinary prospect for a new treatment.

"We sent Matthias a medulloblastoma cell line that was expressing CD155; he treated it with his poliovirus, and the virus rapidly killed the cells," says Olson.

So now Gromeier, at Duke University, is planning pre-clinical trials to test whether the virus kills human medulloblastomas implanted into mice. If such experiments prove the treatment's effectiveness, Gromeier and his colleagues will advance the altered poliovirus to clinical trials in patients.

Olson hopes to identify more molecules, like the CD 155 receptor, that could translate into treatments.

"By identifying genes like CD 155 that are preferentially expressed in the NEUROD3-positive tumors, we might be able to identify new forms of therapy that are directed at the most aggressive medulloblastomas," he says. ■



How Cholera Gets Its Lethal Weapon

David Karaolis, Ph.D.

Sometimes actual science is stranger than science fiction and that's certainly the case in David Karaolis's research on how cholera, usually a benign inhabitant of freshwater lakes and streams, suddenly becomes virulent. It appears that the cholera bacterium acquires its ability to cause massive bouts of diarrhea in people by itself becoming infected with two cooperating viruses, one of which is a new virus that Karaolis discovered in the course of his research. Together, the two form a sort of viral conspiracy to infect and destroy.

When he began his exploration, Karaolis knew that many of the bacteria that make people sick differ from their benign cousins only in that they possess clusters of genes known as "pathogenicity islands." Besides the cholera bacterium, pathogenicity islands have also been found in some varieties of *E. coli*, *Salmonella*, the ulcer-causing *Helicobacter pylorus*, and the bacteria that cause dysentery and bubonic plague. Despite the importance of pathogenicity islands, scientists know very little about their origin, how they are transferred to bacteria, or the role their genes play.

"The ability to cause disease afforded by the pathogenicity island confers an advantage to that particular strain, because it enables that strain to increase its numbers in the environment," explains Karaolis, who is at the University of Maryland School of Medicine. "A harmless cholera bacteria living in water will have relatively limited numbers. But if it can infect humans and produce toxins that cause diarrhea and secretions, millions and millions of bacterial cells will be excreted back into the environment."

While many bacteria appear to have acquired their pathogenicity islands as permanent residents in the distant past, says Karaolis, some appear to move between bacterial strains through bacterial viruses, or bacteriophages, that Karaolis has dubbed "pathophages." Karaolis wanted to find out if cholera was a target of a pathophage.

"We had previously identified a pathogenicity island that was necessary for the disease process in epidemic cholera strains, and that region was absent

in nonpathogenic strains," says Karaolis. "In particular, one gene cluster in this pathogenicity island coded for hair-like pilus structures on the bacterial surface that were essential for the bacteria to colonize." This pilus also served as a docking point and entry for the bacteriophage that carry the toxin genes into the cholera bacteria.

Karaolis's studies showed that some of the pathogenicity island genes closely resembled viral genes and the pathogenicity island tended to insert itself in the same place in the bacterial chromosome.

Then Karaolis and his colleagues discovered a strange strain of cholera bacterium that possessed the cholera toxin, but not the pathogenicity island genes.

"That suggested to me that maybe this pathogenicity island region is mobile, and this strain had somehow lost it. It also led me to think that maybe this region could itself be a bacteriophage."

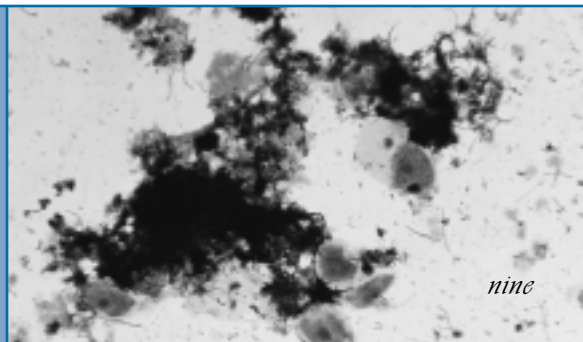
Sure enough, the scientists were able to isolate a new bacteriophage from the bacterial cultures and to show that it contained the pathogenicity island DNA. The hair-like appearance of the phage and its DNA suggest it is a type of filamentous phage, found Karaolis.

"Since the structure and measurements of these filamentous phages were identical to the pili on the surface of the bacteria, we wondered whether these weren't really pili but the outer protein coats of viruses. So, maybe the whole pathogenicity island in cholera may be an active bacteriophage – a pathophage."

The theory was proved when the scientists were able to infect cholera strains with the "pili" pathophage and to transfer the pathogenicity island into the bacterium. What's more, it appears that a "damaged" or mutated pathophage can no longer transfer the pathogenicity island. Furthermore, their analysis of the major coat protein in the phage revealed it to be identical to the coat protein of the pilus. So the phage that carries the cholera toxin actually enters the cholera bacterium through this newly discovered filamentous phage.

Karaolis and his colleagues are now exploring the details of the newly discovered pathophage, its methods of infecting cholera bacteria, and the role of its genes in epidemic disease. They have already discovered that epidemic cholera strains that differ in their virulence also have differences in their pathogenicity islands, as well as their ability to make and produce the pathophage.

Such studies, they believe, will yield important insights into the intricate biological strategies of one of the world's deadliest microbial killers. ■



A Surprise "Hero" in Heart Attacks

Jeanine D'Armiento, M.D., Ph.D.

Most of the seven million people annually who feel the sudden chest-wrenching grip of a heart attack never saw it coming. Then on that fateful day, for reasons no one knows, a tiny fatty globule called plaque breaks loose from an artery wall and clogs an already narrow coronary artery, starving heart muscle of oxygen-carrying blood and killing it.

When Jeanine D'Armiento set out to understand what triggers such potentially deadly plaque rupture, she believed she had at least one culprit in her sights – an enzyme commonly found in plaque called MMP-1. But as frequently happens in science, her experiments brought her a surprise finding. The supposed villain enzyme may actually be a hero that reduces plaque size and number, a finding that may have important implications for drug treatments to prevent heart attacks.

Her quarry, MMP-1, short for matrix metalloproteinase-1 (MMP-1), belongs to a family of enzymes that degrade proteins outside of cells, in the "extracellular matrix."

"The MMP enzyme family numbers more than a dozen, and all of these enzymes probably play a role in a variety of diseases," she says. Thus, D'Armiento believes that her research concentration on the MMP

"Since previous published studies had shown that the metalloproteinases are present in plaque, many people assumed they played a role in rupture, because they degrade the extracellular matrix," explains D'Armiento, who is at the Columbia University College of Physicians and Surgeons. Her previous research on the MMP enzyme family had shown a role for MMP-1 in emphysema, and she decided to use a similar approach to explore MMP-1's role in plaque rupture.

D'Armiento began by locating a type of mouse that lacks an important gene for removing cholesterol from the bloodstream. This mutation makes the mouse susceptible to atherosclerosis, or clogging of the arteries. She genetically engineered the mouse so that it would contain the human gene for MMP-1. In

addition, she added a control signal that caused the MMP-1 enzyme to be produced in immune system cells called macrophages, which eat away plaque build up.

After feeding the animals a high-fat diet, D'Armiento and her colleagues carefully studied the plaque lesions in the animals, using computer image analysis to measure their size.

"We were very surprised to find a significant decrease in lesion size," she says. "The lesions had decreased matrix formation, and most importantly, we found no plaque rupture." As a result of their studies, D'Armiento and her colleagues have come to see MMP-1 in a more favorable light.

"We're beginning to think that the MMP-1 in the macrophage may actually be contributing to beneficial remodeling of plaque, and may not cause rupture. This was very exciting to us, because it was the first time that anyone had shown a beneficial effect of these enzymes."

D'Armiento theorizes that MMP-1 might decrease plaque formation by breaking down the protein collagen that forms the basic scaffolding of plaque, reducing the aggregation of fatty lipids. Or, the MMP-1 enzyme might alter macrophages that attack plaque; or affect the muscle cells' ability to migrate into the plaque deposits.

Whatever the reason for MMP-1's action, D'Armiento sees her findings as offering a cautionary note to companies developing plaque-fighting drugs.

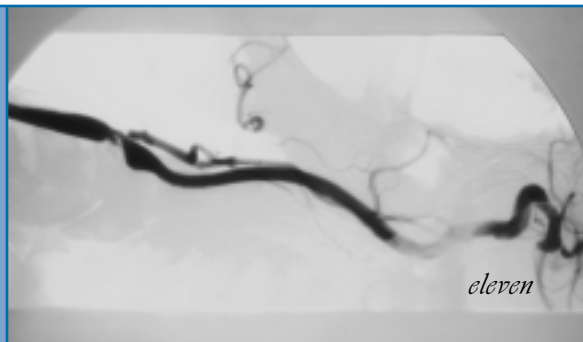
"Many pharmaceutical companies are testing MMP inhibitors to treat atherosclerosis or prevent further heart attacks. I think these findings indicate that they should be cautious, because this enzyme may play a protective role," she says.

D'Armiento is also using transgenic mice to study the effects of another enzyme, MMP-9, which may indeed prove to be a villain in plaque rupture.

"MMP-9 is a much more aggressive enzyme, and in our initial studies, it appears that the lesions in MMP-9 look more unstable. The enzyme appears to eliminate the protective fibrous cap on plaque deposits, which suggests that those deposits might have more potential to rupture," she says.

Since plaque rupture involves more factors than just the enzymes, she is also using the mice to explore whether inflammation destabilizes the plaque deposits. And, by attempting to produce a mouse that does show unstable plaque that leads to rupture, she is examining more closely the biological differences between humans and mice that might explain why mice haven't shown plaque rupture.

In general, she sees the MMP enzyme family as a fertile research field. Additional studies of the role of MMP-1 in lung disease explore why the enzyme seems to be associated with smokers who suffer emphysema. ■

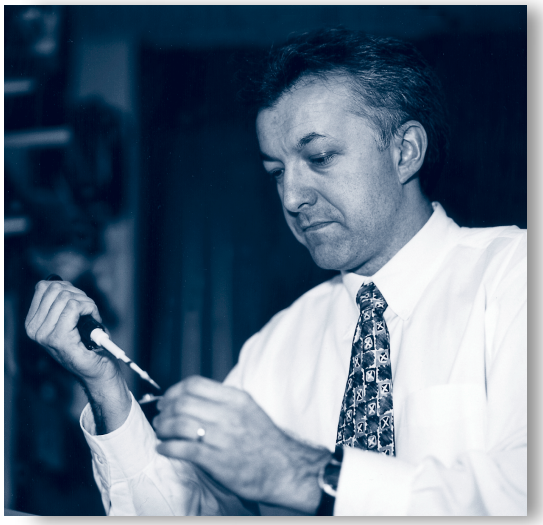


Untangling the Biochemical Web of a Leukemia

George Daley, M.D., Ph.D.

Sometimes it is difficult to see progress in the war on cancer. But in the case of leukemia, scientists have made tremendous progress. The five-year survival rate for leukemia has tripled in the last 38 years. Part of the reason is that scientists have been able to identify the specific genetic changes that cause some forms of leukemia, such as chronic myelogenous leukemia, or CML. Scientists know enough about this cancer's basic biological malfunction to help unlock the dark secrets of other malignancies.

In 1969, two physicians studying chromosomes in cancer cells noticed that a chromosome in the white blood cells of CML patients was shorter in length than that of the same chromosome in normal cells. The researchers pinpointed the precise cause of the shortened chromosome: two chromosomes, numbered 9 and 22, are incorrectly snipped apart and restitched. In this errant rearrangement, a gene called ABL becomes linked to a gene called BCR. While ABL is normally a well-behaved gene that controls cell proliferation, BCR corrupts it, adding a permanent "on-switch" that causes the renegade BCR/ABL duo to churn out a malfunctioning protein enzyme that triggers uncontrolled blood cell proliferation.



This basic understanding of CML has enabled George Daley, of MIT's Whitehead Institute for Biomedical Research, to study the basic machinery of a class of cancers called myeloproliferative diseases, using CML as a model.

"In working toward cancer therapy, what we're really trying to do is to find the targets that drive the cancer cell to grow," he says. In CML, we have the rare privilege of knowing that the precise target is BCR/ABL. So, I'm exploring the metabolic pathways that BCR/ABL activates to drive abnormal cell growth."

For example, the BCR/ABL gene transforms white blood cells so that they ignore the signals that trigger normal cell death, called apoptosis. Daley's studies

"In working toward cancer therapy, what we're really trying to do is to find the targets that drive the cancer cell to grow."

have shown which metabolic pathways may be involved in this phenomenon.

"It is quite a tantalizing hypothesis to consider that even a very subtle defect in the programmed death of blood cells could lead to the tremendously high white blood counts we see in myeloproliferative syndromes," says Daley.

Normally, cells will grow only when they receive chemical signals known as growth factors, which plug into receptors on the cell surface. However, studying bone marrow cell cultures, Daley has found that BCR/ABL thwarts this orderly process.

"BCR/ABL lurks inside the cell and makes the cell believe there are growth factors around, when in fact they aren't."

By tinkering with the BCR/ABL gene to delete various parts of it by mutation, he has discovered that in order to promote cell growth, the BCR/ABL enzyme needs its kinase activity — the ability to switch on other enzymes by adding a phosphate to them.

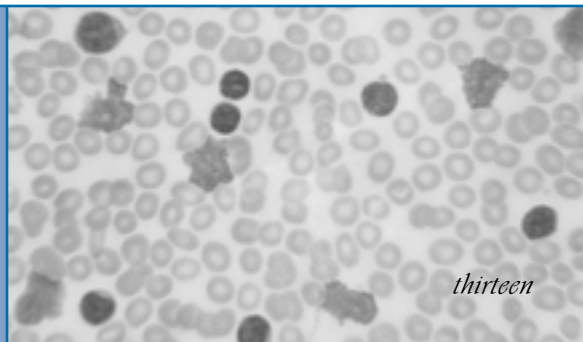
"What's exciting is that the drug company Novartis has developed a compound that inhibits the kinase activity of the BCR/ABL protein," says Daley. "So far, in early clinical trials, it seems to be working beautifully to treat CML."

While drug treatments kill cancer cells, they may also be highly toxic. So, Daley is moving toward understanding how to target BCR/ABL cells immunologically, to eliminate them from the body. Such therapy would be far safer, Daley points out, than the current treatment of destroying the patient's immune system by giving patients high-dose radiation or chemotherapy, then attempting to restore it with a bone marrow transplant. The key to immune therapy for CML might come from studying the immune-system booster interferon, believes Daley.

"Patients who aren't candidates for curative bone marrow transplants are treated with interferon, and many attain startlingly durable remission. People haven't figured out yet how interferon works," says Daley. "However, we have just discovered that a particular gene called Interferon Consensus Sequence Binding Protein, which is turned on by interferon, appears to stimulate an immune response against leukemia cells. We're now working to harness that gene as a more effective way of doing what interferon does, and stimulating patient's immune response against CML cells. In all these efforts, we're emphasizing translating basic knowledge about BCR/ABL mechanisms into novel treatment strategies that are less toxic and potentially curative.

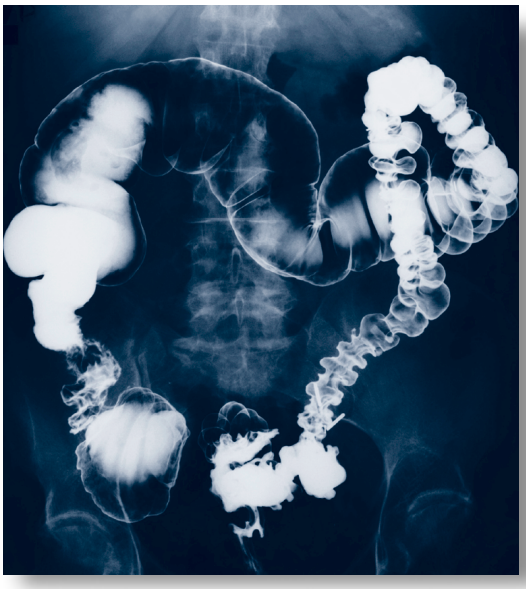
What's more, he and his colleagues are already using their knowledge of CML to search the blood cells of patients for the genetic malfunctions behind other myeloproliferative diseases, including polycythemia vera, essential thrombocythemia, and myeloid metaplasia/myelofibrosis.

"These diseases, like CML, are probably caused by mutations in signaling pathways that make them independent of growth factors. We don't know the genes driving these other diseases, but CML definitely gives us a paradigm to look for them," says Daley. ■



Exploring the "Poison Pills" of Bacteria

Meta Kuehn, Ph.D.



Most people would never have heard of *E. coli*, a bacterium that usually resides benignly in the intestine, except for the fact that certain strains can become highly virulent, secreting a toxin that causes traveler's diarrhea and even death from dehydration of children in developing countries.

Meta Kuehn is trying to uncover the secret of this virulence by studying tiny transparent spheres that bud off the bacterial cell surface. Microbiologists once dismissed these protein-lipid "blebs" as mere random fragments sloughed off by the bacteria. But biochemist Kuehn's research at Duke University Medical Center has shown that these blebs may, indeed, serve as key toxin-carrying "poison pills" in the organism's attack on its host. What's more, the toxins in these vesicles trigger a severe immune reaction, sometimes leading to toxic shock, when antibiotics are used to overcome the infection.

Kuehn and her colleagues separate the vesicles from the other components of the bacterium for detailed study. The experiments have clearly revealed the distinct character of the vesicles, she says.

"For example, our research has found that in the bacterial outer membrane and the vesicles, the protein profiles are not the same," says Kuehn. "One might think that there was a purpose for such differences; the cell does not waste that much energy making lipid and protein for nothing."

Another clue to the vesicles' key role in the cell is that pathogenic strains produce tenfold more vesicles than do harmless strains, she has found. Her studies have also revealed hints about the vesicle proteins' functions.

"We have found that proteins on the vesicle surface resemble those in other bacteria that may have an adhesive or invasive function, so they can actually interact with epithelial cells or with immune cells and thereby stimulate responses," she says

Kuehn is also mapping the geography of the bacterial outer membrane.

"We always think of *E. coli* as a big bag of enzymes, but it's much more sophisticated than that," she says. "The possibility is that there are 'hot spots' for vesiculation that contain the particular proteins that we see in these vesicles; and that would imply a kind of compartmentalization of the bacterial membrane."

Perhaps most important is her recent discovery that the vesicles carry an enriched concentration of toxins compared to the rest of the bacterium.

"This finding is important because it implies that there is a 'selection' process that leads to the inclusion of more toxin than would randomly be incorporated into such a membrane fragment," says Kuehn. "Thus, we think we have some exciting work ahead of us, trying to discern the genetic basis for vesiculation and cargo incorporation into vesicles."

What's more, Kuehn and her colleagues are discovering hints that the bacterium doesn't automatically erupt vesicles on its surface, but varies the number and type of vesicles according to growth conditions.

"We're now at the beginning stages of looking at this phenomenon in a systematic way," she says. "We're trying to determine whether vesicle production is controlled by environmental signals and what they are."

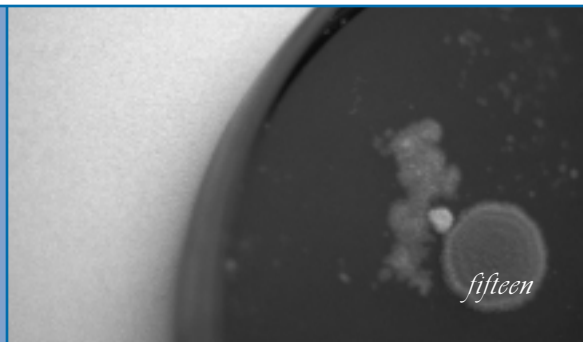
> ***"We always think of* E. coli *as a big bag of enzymes, but it's much more sophisticated than that."***

Besides characterizing the vesicles, Kuehn is seeking to understand their functions. Working with Duke pathologist Soman Abraham, she has found that the vesicles stimulate immune cells known as mast cells to secrete a substance called TNF alpha that induces the body's inflammation response to invaders. She is also exploring the vesicles' effects on macrophages – specialized immune cells that roam the bloodstream attacking invaders.

"Too much of a good thing is a bad thing," she explains. "Our bodies need to make TNF alpha at reasonable levels to be protective, to ensure that there is recruitment of the appropriate immune cells. But too much TNF alpha leads to septic shock. It seems that this overproduction somehow benefits the bacterium in its infective process."

Kuehn expects such insights to help give humans a greater edge over not only virulent strains of *E. coli*, but also other vesicle-producing bacteria, such as the gonococcus bacterium, which causes the sexually transmitted disease gonorrhea.

"We're aiming to increase our knowledge about the enemy," she says. "This includes how these bacteria interact with our immune cells and what their weapons are. If we can discover the components of the vesicles that over-activate our immune system, we may be able to develop treatments to block that process." ■



Understanding the Biological Clock of Birth

Louis Muglia, M.D., Ph.D.

Of the multitude of profound biological mysteries presented by life's processes, to Louis Muglia, understanding the onset of birth constitutes a particularly resonant scientific problem.

"In reproductive biology one of the most important and fundamental questions is what controls the timing of birth," says Muglia, who is particularly committed to exploring this mystery because of its profound medical implications. "About eight to ten percent of all pregnancies are complicated by preterm labor, so it's a very common problem," he says. "Particularly frustrating is that medical efforts over the past 20 or 30 years have not impacted the problem at all, and newborn nurseries are still overflowing with premature babies."

Working at Washington University School of Medicine, he and his colleagues have tackled the problem by using genetic and chemical methods to explore the birth-triggering system in mice. While studying mice isn't the same as studying humans, says Muglia, the two share basic physiology, including some aspects of the birth process.

For example, in both mice and humans labor is triggered by a rise in substances called prostaglandins, whose production in turn is triggered by critical control enzymes called cyclooxgenases. These enzymes include two forms, abbreviated COX-1 and COX-2, and while both definitely play a role in triggering birth, as well as many other body processes, they remain complex and little understood, with differences between humans and mice that complicate comparisons, says Muglia.

In studies to define the role of these enzymes in labor, Muglia used "knockout" mice that lacked the gene for making COX-1, and discovered that the onset of their labor was prolonged so much that their offspring could not survive.

"These findings showed us that the requirement for COX-1 is entirely confined to the mother, and the fetus does not have to make it," says Muglia.

Since in mice, it's the rise in prostaglandins – which causes the hormone

progesterone to drop – that seems to trigger labor, Muglia reasoned that he could restore the mice to normal by removing their ability to make a key hormone that during pregnancy might maintain progesterone levels – namely, oxytocin.

"So when we made animals that are deficient both in oxytocin and COX-1 we no longer needed prostaglandin made by COX-1 to cause the fall in progesterone," says Muglia. Indeed, he discovered that animals deficient in both COX-1 and oxytocin showed normal timing of labor onset.

> ***"What we've done so far is define the changes in the uterus that make it more likely to go into labor."***

In studies that aimed directly at understanding the role of COX-1 and COX-2 in premature labor, Muglia and his colleagues used mice that they could induce to begin labor by giving them the bacterial toxin, endotoxin.

In these mice, the drug indomethacin, which blocks both COX-1 and COX-2, blocked pre-term labor. Similarly, recently developed drugs that only blocked COX-2 also blocked such labor. And when Muglia explored which of the two enzymes was actually present in the uterus during pre-term labor, he discovered increased COX-2, but not COX-1. The findings could have important clinical implications for preventing preterm labor in humans, says Muglia.

"We believe that COX-2-specific inhibitors, at least in inflammatory models of pre-term labor, may be very useful," he says. "The drug that is primarily used now for such cases is indomethacin, which has been associated with significant toxicity to both the mother and fetus." These problems include hemorrhaging in the fetal brain and inflammation of the gastrointestinal tract.

Thus, Muglia is testing the new COX-inhibiting drugs for toxicity in mice and is involved in a clinical study testing a COX-2-inhibiting drug to prevent pre-term labor. He emphasizes that his current studies represent only the beginning of his efforts to understand the biology of the birth process.

"I think what we've done so far is define the changes in the uterus that make it more likely to go into labor," he says. "We really don't know the signals that start the whole process. The COX studies have given us a marker for induction of labor, and we will use that to discover what happens in the mother and fetus to induce this enzyme. We'll then work our way back to define the fundamental mechanisms.

"I'd really like to define what the clock mechanism in parturition is, and where that clock mechanism resides. For example, is the clock entirely confined to the maternal system, or is it in the fetus? Or, is there cross-talk between mother and fetus? Nobody has yet defined the events behind this critical process." ■

Detecting How a Jekyll-and-Hyde Parasite Lurks

Laura Knoll, Ph.D.

The insidious *Toxoplasma gondii* parasite has mastered the art of hiding from the body's defenses, with evidence of its success in the fact that up to half the world's population carries antibodies indicating exposure to the organism, although most people don't even realize they have been infected. Chief among this parasite's evasive strategies is its ability to transform itself from a fast-growing form called a tachyzoite to a dormant variation called a bradyzoite that encases itself in a protective cyst.

The genetic machinery that *Toxoplasma* uses in the transformation process is a profound scientific mystery, says Laura Knoll of Stanford University. "Although we don't really know what causes *Toxoplasma* to go into the cyst form, most likely it's the body's immune response. We do know that cyst formation makes sense evolutionarily, because if the parasite stayed in the fast-growing form, it would kill its host within about 30 days by destroying all the muscle and brain tissue."

Especially important to the parasite, says Knoll, is that the hardy cyst is the infective form when people eat undercooked infected meat. People can also contract the disease – which causes swollen glands and fever – from contact with the feces of infected cats, through changing cat litter or when gardening. So tough are these cysts, she says, that no drugs can touch them. Particularly vulnerable are pregnant women, because infection can be passed to the developing fetus and cause birth defects, and people with AIDS and transplant patients, whose immune systems are suppressed.

"But if we understood the genetic switches that cause the tachyzoites to become bradyzoites, we might be able to develop drugs to either keep them in the cyst form or to turn them into the fast-growing form which can be killed by current therapy," she says.

Using genetic screening techniques, Knoll is searching for the genes that are turned on when the parasite first begins the switch to the dormant form.

Studying *Toxoplasma* cells in culture, she uses a "gene trapping" method to track these genes. The technique basically involves inserting a marker into the genes of the parasite cells that make them susceptible to specific drugs if the marked gene is turned on. By applying the drug to marked cells, she can isolate the cells that are in the process of switching, thereby distinguishing genes that are turned on early in the transformation from tachyzoite to bradyzoite.

So far, she has isolated several bradyzoite-specific genes, as well as evidence that the parasite uses a sort of genetic jump-start to speed its Jekyll-Hyde transformation. Called "post-transcriptional regulation," the jump-start method involves having certain genes always prepared to be translated into proteins that will perform the cyst transformation.

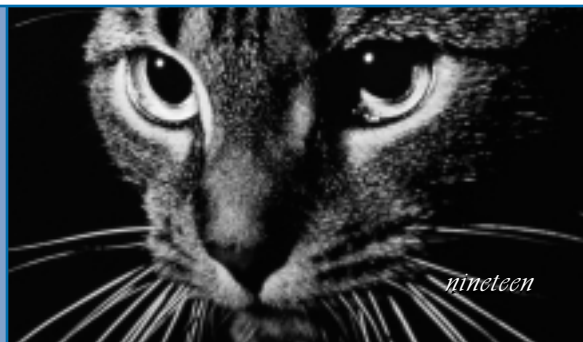
"It's a common mechanism that organisms use when they need to respond quickly to stress, in this case to switch to a protected cyst," says Knoll. "Now, we're trying to understand exactly how the mechanism works in *Toxoplasma*."

In a second major effort, Knoll and her colleagues are creating mutant forms of *Toxoplasma* in an attempt to understand its switching mechanism. In a technique called "signature-tagged mutagenesis," she has randomly inserted bits of gene-wrecking DNA into the *Toxoplasma* genome. Each of these bits carries a unique signature-tag, a sequence that scientists can use to identify the DNA.

"I have a library of about 5000 of these mutants, and I'm going to be testing how they develop cysts in mouse brain," says Knoll. By identifying via the tags those mutants that fail to form cysts, Knoll can work backward to determine how the mutants are genetically defective and pinpoint which genes are critical for cyst formation.

Using such powerful techniques, Knoll hopes to better understand, not only *Toxoplasma*, but also its insidious cousins that cause malaria and *Cryptosporidium* infections.

"As parasites go, *Toxoplasma* is really the most genetically tractable," says Knoll. "So, one long-term goal is to see if we can use what we learn from studying it to understand development and cyst formation in these other parasites." ■



Origins of Anxiety: Clues from the Mouse

Edward Brodkin, M.D.

Panic attacks, divorce, alcoholism, drug dependence, inability to work, even suicide – these are only a few of the consequences of anxiety disorders, suffered by one in four people in the United States at some point in their lives. However, despite the profound impact of anxiety disorders, researchers still have little understanding about what causes them. For Edward Brodkin, this gap in knowledge presents a challenge that has launched him on a scientific odyssey to begin mapping the genetics of anxiety.

"There definitely seems to be a genetic component to anxiety," he says. "Many studies of families, twins, and adopted children have shown an important genetic predisposition to several anxiety disorders, although environmental factors also play a role. But the identity of those genes is the really big question." Progress has been slow in understanding anxiety disorders not only because of the complex interplay of heredity and environment, but because genetic experiments are obviously impossible to do on humans.

However, Brodkin believes that in studying mice he can forge an important pathway to scientific understanding.

"With mice, there are inbred strains that are completely genetically homogeneous," he explains. "So within a particular inbred strain every single mouse of the same sex is genetically identical to every other. It's also well-established that inbred mouse strains differ significantly on tests of anxiety."

Brodkin's work at Princeton University combines these behavioral differences and genetic analysis of mouse strains to reveal the hereditary basis of anxiety. He has begun by screening some of the 100 or so mouse strains to find large differences in predisposition to anxiety.

"One might wonder how it is possible to study an emotion like anxiety in mice," he says. "In fact, mice put in anxiety-provoking environments – like open high places or brightly lit environments – show a consistent set of measurable behavioral responses such as freezing in place or stretching out toward the ground." Thus, Brodkin will use two well-established behavioral tests, exposing mice to a maze that places them on an elevated platform or in a dark-light box that exposes them to bright light.

Once the tests have revealed clear differences among strains, Brodkin will use a method called quantitative trait locus (QTL) analysis to seek the genetic origins of the differences.

QTL analysis constitutes a sort of "aerial reconnaissance" of chromosomes to seek the chromosomal regions, called loci, that contain genes affecting a behavioral trait. Basically, the technique involves crossbreeding mouse strains that differ maximally in an anxiety trait and measuring the level of that trait in offspring. By correlating anxiety levels in crossbred mice with genetic analysis of those mice, Brodkin can locate the chromosomal locations associated with inheritance of anxiety traits.

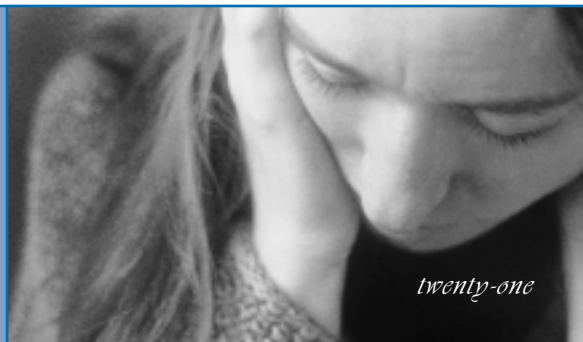
"These traits are usually influenced by many genes, and QTL analysis is a method for identifying these multiple loci," says Brodkin. "Once you find the loci, you seek to identify the relevant gene within that locus." Pinpointing such genes is like detailed ground exploration of terrain scouted using aerial reconnaissance, and it is a challenging task, says Brodkin.

"A great advantage of studying mice is that if you can identify a gene that affects anxiety, you can quickly begin testing humans..."

"However, I'm optimistic about locating such genes, because techniques for fine genetic mapping are getting better all the time, and also because of enormous progress in the human and mouse genome-mapping projects," he says. "It used to be that when a researcher mapped a locus and then looked in the genetic databases for genes within that locus, there weren't many identified. But thanks to the genome projects, over the next several years we will be able to identify a host of genes within a given locus." Critically important, emphasizes Brodkin, is that the international gene-mapping projects include both mouse and human genomes.

"For virtually every human gene there is a mouse homologue, and vice versa," he says. "So, a great advantage of studying mice is that if you can identify a gene that affects anxiety, you can quickly begin testing humans to see if the same gene is correlated with anxiety disorders."

Since anxiety diseases likely involve many genes exerting subtle effects, Brodkin's scientific journey will be long and arduous. He will spend countless hours breeding colonies of hundreds of mice, minutely observing their behavior and carefully analyzing their DNA. However, the payoff could be tremendous, opening a new pathway into understanding anxiety disorders that affect millions of lives. ■



Yeast Yields Budding Science

Sylvia Sanders, Ph.D.

©Copyright Justin Allardyce Knight

An ordinary baker's yeast cell viewed under a low-power microscope is a model of simplicity – a rather uniform sphere with no discernable organization. But when it begins to create daughter cells it creates an entire architecture of scaffolds to help it divide. Sylvia Sanders believes this architectural system holds important clues to some of the most fundamental questions about how cells organize themselves to grow. She has set out to tease apart the biochemical details of how these simple one-celled organisms create their architecture, seeking insights into this fundamental biological process that underlies all animal development.

"We're interested in how cells and organisms are able to build structures at the right time, at the right place, and make them in the right shape," says Sanders, who is an assistant investigator at the Howard Hughes Medical Institute at Massachusetts Institute of Technology. From bacteria to growing neurons, all cells must organize their internal structures to position molecules critical for growth, she says.

In such organization, all cells seem to establish a "targeting patch" on their surface as a focal point for reproduction. In yeast cells, this patch is the site at which the cell creates a bud in order to divide.

"Conceptually, yeast budding is very similar to cells of higher organisms," explains Sanders. "When the yeast decides to enter the cell division cycle, it grows a bud in a particular place. And it organizes its internal cytoskeleton asymmetrically, which allows polarized secretions necessary to build the bud.

"The 'daughter' bud continues to grow at the expense of the 'mother,' until it becomes nearly as big as the mother, when cytokinesis – cleavage into two cells – occurs. The cell's cytoskeleton, which is built mainly of the protein actin, controls cell shape, organization, and movement," she says.

"We're attempting to understand the subtle, complex interplay of genes to create proteins that control budding, cytoskeleton architecture, and actin

production," she says. "More generally, we're trying to understand why yeast cells may want to have a particular budding pattern at all."

Yeast cells may choose between two basic budding patterns, based on their circumstances, explains Sanders. They may bud "axially," with cells budding adjacent to where they budded previously. Or, they may show "bipolar" budding, in which a cell buds opposite from where it previously budded.

Sanders studies mutant yeast cells that, while otherwise perfectly viable, are unable to pick appropriate sites for polarized growth.

"We're trying to use such mutants to understand how the cell uses the spatial information dictated by the bud proteins," she says. "I really would like to know how the bud spatial information impacts on the cytoskeleton dynamics and the secretory machinery. If we could isolate a complex of factors, including bud factors, that could stimulate actin formation, that would be very exciting."

Studies by Sanders and her colleagues have focused on the action of proteins produced by genes called BAD, for "bud-site selection axial determinant." For example, she has isolated a mutant yeast called BAD15 with the peculiar effect that the mother cells bud axially, but their daughters show bipolar budding. Sanders' studies have revealed details of the malfunctioning BAD15 cellular machinery that can produce such a paradoxical result.

Sanders is especially interested in whether the machinery of yeast budding is connected with the process by which cells cleave, called cytokinesis.

"The factors that specify bud position might have redundant roles with other components involved in essential processes, like cytokinesis, so under certain conditions bud factors would be essential," she says. "We already have genetic evidence for this connection, showing that the budding factors may help stabilize cytokinesis, perhaps providing the cell some backup."

Sanders and her colleagues are extending their work to explore the effects of aging on yeast budding and how the timing of budding is controlled. She also plans to study other cells, such as neurons in the ear, which must grow in the right time, place, and shape to produce a functional auditory system.

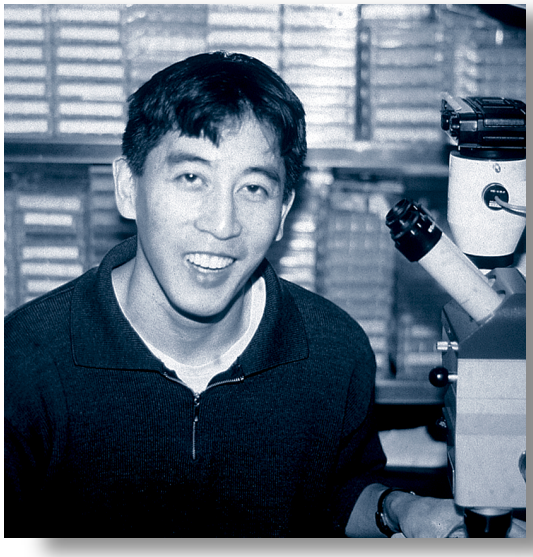
"We do know that a protein called formin — just like one involved in polarity decisions in yeast — is required for hearing," she says. "So, I believe that understanding how this molecule functions in model systems could definitely shed some light on the mechanism of the hearing defects in patients with inherited hearing disorders." ■

Exposing the True Face of HIV

Peter Kwong, Ph.D.

HIV is a stunningly insidious virus, constantly, subtly shifting its structure to avoid being targeted by the immune system, even as it destroys that system. Peter Kwong of Columbia University has dedicated himself to opening new medical pathways to attack HIV by obtaining the most detailed knowledge yet of its structure.

One key element of HIV is its machinery for attacking T lymphocytes, the immune cells that are the virus's main target in the body. That machinery includes a protein called gp120, which festoons the surface of the virus, and which it uses to latch onto a receptor on the T lymphocyte called CD4, in the initial step in penetrating the cell.



Many researchers have concentrated on gp120 as a possible target for anti-HIV drugs or vaccines. But such treatments had not worked well in the past, and to understand why, scientists wanted to get a clearer look at the structure of gp120. The challenge was perfect for Peter Kwong, who specializes in the powerful analytical technique of X-ray crystallography. Basically, this technique involves projecting X rays through a crystalline form of a protein. By analyzing the complex pattern of diffracted X rays produced by the protein, scientists can deduce its structure.

For Kwong and the other team members, the trick was to crystallize the gp120 protein attached to CD4. The problem was gp120's notoriously "floppy" irregular structure. But Kwong used a new approach. The research team constructed and tested slightly altered forms of the protein, looking for one whose molecular shape lent itself to crystallization. Finally, after two years, the researchers found such a molecule, and Kwong was able to obtain the gp120-CD4 structure. In June 1998, he was the lead author on the scientific paper in *Nature* announcing the new X-ray structure. For Kwong, the achievement was only a means to his major goal.

"I'm really interested in medical application of this knowledge," he says. "The problem is it's not clear how the huge body of knowledge that's contained in the structure can be applied to something medically related. While this information does show you how clever HIV is in evading the immune system, knowing its defenses doesn't necessarily mean you can see a route to a drug or vaccine."

For example, the virus has evolved its gp120 protein so that the critical connections with its target – those that must remain constant to work – are shielded from view, so that the immune system cannot use its defenses against this segment of the virus. However, says Kwong, the detailed structure of the gp120-CD4 combination reveals intriguing cavities which drug molecules might be able to plug into to jam the infective machinery.

Kwong and his colleagues are using their structural information, working with the drug company SmithKline Beecham to explore small molecules that might jam the infective machinery. However, Kwong points out, even if such drugs are developed, they would be expensive.

> ***"If you're really going to make a broad medical impact, I think you have to find new approaches to a vaccine."***

"In terms of treating the worldwide pandemic, there is no way developing countries can afford these drugs. So, if you're really going to make a broad medical impact, I think you have to find new approaches to a vaccine." Unfortunately, he says, the gp120 has not proven a good vaccine target.

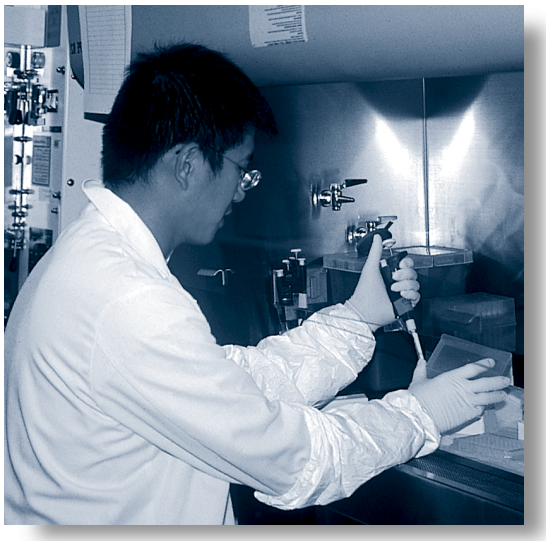
"HIV has evolved all kinds of immune-evading defenses in which it avoids the antibody contacts that the immune system uses to attack viruses," he says. However, armed with the new high-resolution picture of gp120, Kwong believes that he and his colleagues may find a way to induce the immune system to attack the virus.

"The answer, in part, might be that we concentrate on regions of gp120 that can't change, such as the part that binds CD4. I call this the true face of HIV, which most of the time hides deep within its structure, and I think there are ways to expose this true face of gp120 to the immune system." ■

Pinpointing the Prion Protein

Surachai Supattapone, M.D., D.Phil.

The idea of the prion, short for "proteinaceous infectious particle," was so utterly alien that when biochemist Stanley Prusiner and his colleagues first proposed it in the early 1980s, most researchers greeted the concept with disbelief. How, they asked, could a disease possibly be transmitted with no genetic material, neither DNA nor RNA? But after nearly two decades of research, science has largely accepted that an aberrant, infective protein does, indeed, cause such fatal brain-destroying human diseases as Creutzfeldt-Jakob disease and kuru, and the animal diseases scrapie and bovine spongiform encephalopathy, or "mad cow disease."



Now, the scientific challenge is to understand how the usually harmless cellular brain protein, called PrP^{C}, changes its molecular shape to become a pathogenic scrapie-producing form, called PrP^{Sc}. During infection, pre-existing PrP^{Sc} recruits normal PrP^{C} to transform it into the new PrP^{Sc}.

Surachai Supattapone, working in Prusiner's laboratory at the University of California at San Francisco, has set out to understand the workings of the prion protein by creating mutant versions of the normal PrP^{C}. His objective is to understand how normal protein turns into the aberrant PrP^{Sc}.

"Normally PrP^{C} is a 208-amino-acid molecule, and my project was to examine what would happen if we created deletions of various regions of PrP^{C}, based on structural analysis of the protein," explains Supattapone. To his surprise, a mutant form he created that included only 106 of the protein's amino acids allows infection by full-length prions.

"PrP106 is only half the size of full-length PrP. Nevertheless, it is capable of supporting scrapie infection," says Supattapone. In his experiments, Supattapone and his colleagues used mice whose normal PrP^{C} gene had been deleted, making them resistant to scrapie infection. However, when the scientists introduced the gene for the shortened PrP106 protein into the mice, and infected them with full-length prions, the mice developed neurological symptoms and

accumulated PrP^{Sc}106. Brain extracts from infected PrP106 animals were infectious only to other PrP106 mice, but not to mice expressing full-length PrP^{C}, found Supattapone. While it took 300 days for PrP106 animals to show infection from full-length PrP^{Sc}, it took only 66 days for them to become infected by PrP106. This unexpected artificial transmission barrier between PrP106 and full-length PrP^{Sc} resembles the naturally occurring prion transmission barrier between different species of animals.

Also important, says Supattapone, is that the truncated PrP106 protein appeared to have a structure very different from full-length PrP^{C}. For example, he says, the shorter protein is partially resistant to degradation by an enzyme called proteinase K, while the full-length prion version is totally susceptible to such attack; and the shorter PrP106 was partially soluble, whereas the longer protein was fully soluble.

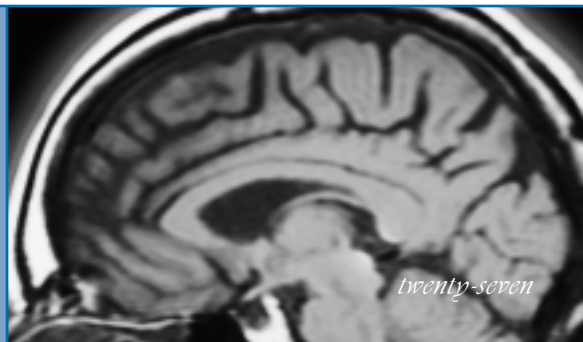
"So, we think that PrP106 might represent a kind of intermediate between PrP^{C} and PrP^{Sc}," says Supattapone. The shortened version might already be partially unfolded, in essence on its way to the process of transforming into PrP^{Sc}.

If so, Supattapone's finding might support a theory called the "template assistance model" of how the prion protein wreaks its havoc. In this theory, normal PrP^{C} molecules bind to an unidentified "template" molecule, which unfolds PrP^{C} into a globule that is vulnerable to conformational change. The infectious PrP^{Sc} helps this globule refold itself to propagate the aberrant form, which accumulates and kills.

"So PrP^{Sc} is like an enzyme, catalyzing the formation of an intermediate state," says Supattapone. "And we think that PrP106, because of its deletion, is already unfolded and doesn't need to interact with the template. It's ready to go, so when PrP^{Sc} comes along, PrP106 is ready to interact quickly and propagate."

Supattapone is continuing analysis of the shortened PrP^{C} protein to understand even more about the strange infectious process of prions. He is also whittling the protein down even further to find the absolutely necessary pathogenic core of the PrP^{C} protein. Importantly, his work has also hinted at a possible pathway to treatment for prion diseases.

"Almost accidentally, I stumbled on the fact that highly branched polyamine compounds appear to clear pre-existing PrP^{Sc} from cells at non-cytotoxic concentrations," he says. "When a cell culture is treated with the compound and then cultured in a compound-free medium, the cells remain PrP^{Sc}-free a month later, so these compounds may have potential as therapeutic agents." ■



As if understanding life's machinery wasn't complicated enough, biomedical scientists must also master a myriad of professional skills – from grant writing to interviewing for faculty positions – to give themselves the best chance to succeed as research scientists. These practical skills should be acquired during graduate school and postdoctoral training, but too many scientists emerge from this training period without the career savvy that can be as important as scientific acumen to building a successful and productive scientific career

We at the Burroughs Wellcome Fund recognize the critical importance of helping young scientists cope with professional and personal challenges. The Fund's Career Awards Program, in particular, has been designed to give, not only financial support, but also practical information about a broad range of professional skills. Our program officers and associates, themselves experienced scientists and grant administrators, often assist awardees with issues such as negotiating a faculty position and leveraging start-up funding. Through our experiences in talking with awardees, we recognized the dearth of available information on topics such as negotiating a start-up package and inquiring about pension and retirement incentives.

To help remedy the situation, the Fund has developed a career development booklet to provide basic information on career topics. However, we soon realized that many more people could reached by through the Internet. Together with the Howard Hughes Medical Institute and the American Association for the Advancement of Science, we developed a free online Career Development Center (nextwave.sciencemag.org/feature/careercenter.shtml) as part of *Science* magazine's Next Wave (www.nextwave.org), a Web site for advanced postdoctoral scientists and new faculty. The Career Development Center includes information on topics such as grant writing and management and the state of the academic job market, as well as a wealth of links to online resources to aid postdoctoral and junior faculty.

The Fund has also joined with the National Academies' Committee on Science, Engineering, and Public Policy (COSEPUP) to produce a guide to the postdoctoral experience that will be available in September 2000. For more information, contact Dr. Deborah Stine at 202-334-3239 or visit their Web site (www4.nationalacademies.org/pd/cosepup.nsf).

We also realized that our career awards meetings offer a rare opportunity for awardees to meet and learn from one another, forge collaborations, and seek advice from senior scientists on our board and advisory committees. But perhaps most importantly, we try to provide an environment in which awardees feel free to ask questions, to give constructive feedback on our awards, and to seek advice about career topics, something that is absent at most meetings young scientists attend. At each career awards meeting we offer several sessions on topics that the awardees themselves have chosen.

What follows are synopses of the 1999 convocation sessions on:

- mentoring
- laboratory management
- issues facing women scientists and dual-career couples
- academic-industrial partnerships.

Other issues that are being considered for future meetings include collaborative research, grantsmanship, presentation skills, communicating science to the public, negotiating faculty positions, and understanding the tenure process.

Shaping a Successful Career With Mentoring

Joan Reede, M.D., M.P.H., Associate Dean for Faculty Development and Diversity, Harvard Medical School, spoke about the importance of mentoring – both as a giver and receiver of guidance – as an important interpersonal skill. She emphasized that the mentor's primary role is to help the mentored person to achieve career goals, rather than for that person to serve merely as an apprentice. Becoming an effective mentor, she said, rests on establishing an open environment for cooperation and communication, in which creativity and problem-solving can flourish. Listening, rather than talking, is the most important part of the mentoring process.

Effective mentoring includes understanding the mentored individual's motivation, goals, and values, building

Construction Skills for Building a Career

a climate of trust, and negotiating the responsibilities of both sides of the mentoring relationship. The prospective mentored individual, she said, should talk to people who were previously mentored by the senior scientist to gain a thorough knowledge of the outcome of a mentor's previous relationships.

Both parties to the mentoring relationship must also be prepared for their relationship to evolve as the mentored person moves toward independence, achieves career goals, or sets new goals. The relationship may also evolve with changes in department leadership and the funding environment.

As women and minorities in the scientific enterprise increase, they are being particularly sought after as mentors and for committee membership. Although these are important responsibilities, people in these positions must consider carefully whether additional responsibilities will compromise their time and in turn the quality of their research.

Listening, rather than talking, is the most important part of the mentoring process.

Reede suggested that mentors should limit the number of mentoring relationships in order to have enough time to devote to each mentoring relationship.

Finally, both parties should be prepared to end unproductive relationships, such as those in which the mentor or the person mentored may be passive, manipulative, inaccessible or negative. However, she said, such exits should be as graceful as possible, leaving bridges to a further relationship firmly intact.

Further reading on the subject of mentoring: *Adviser, Teacher, Role Model, Friend: On Being a Mentor to Students of Science and Engineering* (National Academy Press, Washington, D.C., 1997). Available online at (books.nap.edu/books/0309063639/html/1.html). In addition, many universities now operate an office that manages mentoring issues, usually the office of faculty development.

Laboratory Management Issues

The 1999 meeting also featured discussions of laboratory management issues. Several speakers emphasized that managing a productive research laboratory is central to a successful research career. Such management requires a combination of scientific, interpersonal and political skills that can be learned.

For example, a key to successful laboratory management, said Thomas Davis, Ph.D., Professor of Pharmacology, University of Arizona College of Medicine, is that the head of the lab demonstrate personal leadership through maintaining a constant daily presence in the laboratory. This presence should include close consultation about experimental progress and technical issues, including a continuing dialogue about principles of the scientific method, research integrity, institutional standards and policies, and responsible research practices.

Among these practices is the maintenance of laboratory notebooks. One reference for further reading is *Writing the Laboratory Notebook* (American Chemical Society, Washington, D.C. 1985). Among the criteria for a good notebook Davis cited is that it be a bound, page-numbered volume, handwritten in ink, with entries signed, dated and witnessed. All data in original hard-copy form should be inserted into the book. Computer files, he said, do not constitute a legitimate laboratory notebook. All who work in the laboratory should understand that they do not own the data; the data are the property of the laboratory.

The laboratory notebook, he said, should be well organized, with headings, and should contain exhaustive accounts of all research and related issues that occur in the laboratory. Scientists should "think in your lab notebook," making it a comprehensive diary of ideas, observations, detailed experiments, and notes taken at laboratory meetings. The last point is critical, he said, since too many brilliant ideas in laboratory meetings have been lost because notes were not taken in laboratory notebooks. Also, exhaustive records of experiments make it unequivocally clear who should receive credit, including co-authorship on scientific papers, for contributions to a research project.

A detailed research protocol book – also *not* kept primarily as a computer file – is also a key document in any well run research laboratory. This book should contain a highly detailed account of all procedures used, with each change dated and signed. Many laboratories, he said, find it useful to post such procedures on their Web sites, particularly for sharing with other laboratories. Again, however, the information must be absolutely current, with changes signed and dated, including notice of copyright, where appropriate.

Carefully maintaining protocol records enormously aids the conduct of precise, reproducible experiments, the training of new personnel, the maintenance and use of equipment, and the preparation and publication of papers and reports, he said.

Reporting results should include following accepted guidelines. For example, guidelines for statistical reporting in medical journals can be found in the *Annals of Internal Medicine,* 108:266-273, 1988. According to these guidelines, scientific authors must describe their statistical methods in detail, quantify

their findings, and present them with appropriate indicators of measurement error or uncertainty. Authors should also discuss eligibility of experimental subjects and include in their papers details about randomization, treatment complications, number of observations, losses of subjects to observation, and computer programs used.

Managing Issues Facing Women

Women have achieved major progress over the last decade in successfully entering scientific careers. Indeed, the Fund strongly supports in its programs proportionate representation of women in science.

Women contribute distinctive professional talents and personal attributes that are critical to scientific advancement, emphasized speakers Mary-Lou Pardue, Ph.D., Boris Magasanik Professor of Biology, Massachusetts Institute of Technology and Suzanne Pfeffer, Ph.D., Professor of Biochemistry, Stanford University School of Medicine. Women have made great strides in learning to manage competing demands that are often very different from those faced by men in building research careers. And, they have learned to mentor both women and men.

Yet women still face hurdles to achieving positions of leadership in the scientific community. Several career advisors at the convocation sessions recommended especially that women cultivate the habit of confidently and assertively advocating on their own behalf by seeking fair compensation, adequate facilities for their research, or even nomination for an award. In negotiating for their initial academic positions, women should be willing to ask for not only fair salaries and other benefits, but also a clear understanding of how childbearing, if planned, will affect timing of the tenure process. Typically, say administrators, maternity leave of more than a year may pose difficulties in remaining on a tenure track.

Women must cultivate the habit of confidently and assertively advocating on their own behalf.

Women may also want to think strategically about advancing their career through non-research activities. Service on key academic committees, such as search committees that will guide future recruitment, may help career progress. However, some speakers emphasized that women should feel absolutely justified in declining service on committees or other administrative tasks whose time demands could interfere with their research progress.

Mentoring among women and collegial relationships with peers may have a somewhat different dynamic than for men, according to one speaker. Women must try to overcome any possible tentativeness about developing professional relationships with male mentors and colleagues. They should actively seek the best mentors, whether male or female, and should engage male colleagues on a collegial level when circumstances warrant.

Managing Dual Careers

Two-career couples – both dual-academic-career couples, and those in which one partner is outside academe—enjoy many advantages in career-building, but they also face challenges, say experienced advisors. Certainly, the mutual support couples can give one another in their careers is a definite advantage. A major initial difficulty, however, may come when one member begins negotiating for a permanent position, or a subsequent higher position, while the other may not be ready to move. Such cases can often be handled by clear and open communication between the partners to reach agreement as to the best solution for both, said Jane Loeb, Ph.D., of the University of Illinois at Urbana-Champaign, and co-editor of *Academic Couples: Problems and Promises.* eds. Marianne A. Ferber and Jane W. Loeb (University of Illinois Press, Chicago. 1997).

Clearly, requesting that a university commit to hiring two people must be handled with diplomacy, but such negotiations can have a very positive outcome.

For dual-academic-career couples, if the "trailing" partner is willing to move, advisors recommend that during negotiations the partner being recruited raise the issue of a second academic position when appropriate – generally somewhere between the opening interview and final negotiations. Also, when the institution is not willing or able to offer a second position, it may be willing to help the partner secure a position at a nearby institution. In some cases, the institution may be willing to consider offering the couple two part-time shared positions, with the potential for dual tenure tracks. Clearly, requesting that a university commit to hiring two people must be handled with diplomacy, but such negotiations can have a very positive outcome for both the couple and the institution, she said.

For two-career couples who have or are contemplating having children, a contemplated move to a new institution might include contacting the institution's human resources office to explore whether the institution is "family friendly."

While research collaborations between dual-researcher-couples might seem to be easily managed, couples were counseled to enter such partnerships with care. For example, dual-researcher couples – especially where one member is senior to another – may face insinuations that the senior member is the major contributor to a project. One solution to such assumptions is to make quite clear each partner's contribution to proposals, presentations, and papers. In some cases, it might be the wisest course to postpone collaborations between partners until both have established their own independent reputations.

Forging Productive Academic-Industrial Connections

Developing useful relationships with industry is another important area that early-career basic scientists must manage carefully and cautiously. Given that basic biomedical discoveries these days frequently attract commercial interests, researchers often face the possibility for industrial involvement far earlier in their career than in past decades. Working with an industrial collaborator will not appeal to every researcher. Such partnerships might be too complex and time-consuming for a scientist interested primarily in basic laboratory investigation. However, if a researcher does decide that academic-industrial partnerships are of interest, he or she must be prepared to develop skills and expectations in working with industrial colleagues that are sometimes quite different than those needed in the academic research laboratory, according to meeting speakers.

Surprisingly, industrial relationships often do not bring large sums in research support, contrary to some academic scientists' expectations. Statistics show that most industrial research support agreements bring significantly less than $100,000 a year in funding, with only a very few percent bringing in $500,000 a year or more. Overall, industrial support represents about five to ten percent of academic biomedical research budgets, compared to federal and other funding resources. Industrial contracts are also usually short, typically lasting no more than a year. While academic researchers might seek basic research funding and in-kind support, corporations are more interested in supporting applied research that will yield a patented product.

A corporate relationship may take many forms besides direct research support, ranging from consultation to participation by the scientist in launching a new company. The most common and simplest relationship is consulting, in which a scientists agrees – usually for a fee of about $1,000 per day – to provide his or her expertise on a specific problem. Here speakers emphasized that all industrial relationships raise a wide variety of important professional issues, including the expectations of the two parties regarding the relationship's outcome, publication rights, academic independence, effects on the tenure process, ownership of intellectual property, handling of proprietary information within an academic laboratory, and impact on patient care.

Given the complexity of such issues, the speakers also strongly advised new researchers to seek counsel and information in advance from experienced senior researchers and importantly, the university's technology transfer office. The beginning researcher should fully understand all institutional rules and regulations governing industrial relationships, especially those involving data ownership, intellectual property, conflict of interest, confidentiality, and indirect costs. Critically important, the researcher should sign nothing without having it reviewed by the technology transfer office, the speakers said. Open communication among all parties regarding contractual arrangements and other commitments should be the rule, with the researcher regularly informing all involved, both in the corporation and in the university of the expectations and developments regarding the relationship.

A corporate relationship may take many forms besides direct research support, ranging from consultation to participation by the scientist in launching a new company.

Technology transfer professionals at the meeting also cautioned junior academic researchers to prepare themselves to cope with sometimes heavy demands on their time from industrial partners. Researchers must make careful decisions about whether they can meet these demands, while still fulfilling the university's expectations for research, teaching, publication, and service. However, they also agreed that involvement with corporations can bring many worthwhile benefits, including access to advanced equipment, new ideas, and the enormous satisfaction of seeing one's basic discoveries benefiting society. ■

As the profiles in this report amply demonstrate, there are many talented investigators capable of making significant contributions to science if given opportunities at the beginning of their careers. Indeed, the future of the entire biomedical research enterprise depends upon this next generation of researchers. That's why we at the Burroughs Wellcome Fund have made a commitment to identifying and nurturing the research careers of the next generation of scientists.

We recognize that career prospects of biomedical scientists today are substantially different from what they were 20, or even 10 years ago. Today's life scientist is slightly older and takes more than two years longer to obtain the Ph.D. degree. A newly minted Ph.D. today will enter the swelling ranks of postdoctoral fellows waiting to obtain permanent positions. The average life scientist today is likely to be 35 to 40 years old before getting a permanent job.[1]

In today's tough job market, our Career Awards Program offers a rare opportunity for a measure of independence for young scientists conducting postdoctoral research. We seek to identify the highest-quality scientists while they are still postdoctoral fellows and give them financial independence to conduct scientific projects of their own design in anticipation of obtaining a faculty position. Our hope is that getting a career award, at a transitional phase of training when developing autonomy is critical, will help these scientists obtain an independent position and "hit the ground running" with the security of initial financial support.

After five years of administering the Career Awards Program, we have begun an evaluation process to determine if we are indeed developing independent scientists and to assess if our awards are indeed making a difference in their careers. We completed a survey of awardees in 1998 and 1999 to gather their impressions about the impact of the program on their careers and to assess their progress. Preliminary data indicate that the program is on the right track.

Of 101 awardees who received grants since 1995:

- All of the 64 awardees who were eligible for faculty positions have received tenure-track appointments.
- The institutions that hired awardees provided "start-up" funding that averaged $327,000, with one awardee garnering a $1.1 million start-up package.
- During 1998 and the first half of 1999, 78 percent of those with new faculty positions have published one or more articles in peer-reviewed journals.
- Of these publications, nearly two-thirds were in highly prestigious journals such as *Science* and *Nature*.

Overall, our awardees continually emphasized that career awards were key to giving them the independence to strike out along promising new research pathways. Awardees who had already identified their desired research directions told us that the awards enabled them to develop momentum, in the form of solid data and research plans, that make additional funding easier to obtain. Awardees who obtained faculty positions told us they are able to spend an average of 84 percent of their time on research.

Also helpful, we have learned, is that our awardees bring to the academic negotiating table the leverage of existing funding – critically important, since academic appointments today bring with them high expectations of early research accomplishments and self-sufficiency in grant support. This performance pressure means that most new faculty must spend their first three years writing grant proposals, whereas our awardees have the ability to concentrate on developing pilot data, that is crucial to long-term funding success rates.

In part because of this jump-start, our awardees have been quite successful on their initial grant applications and in obtaining support from other funding sources. Of the career awardees who have obtained faculty positions, 75 percent have already submitted grant applications to various sources and 56 percent had received awards that averaged $133,000 in annual direct costs.

[1] *Trends in the Early Careers of Life Scientists* Committee on Dimensions, Causes, and Implications of Recent Trends in Careers of Life Scientists, National Research Council, 1998.

On Nurturing Research Careers

While employment, funding and publication data give an indication that our awardees are doing well, our experience is that there is considerable art, as well as science, to nurturing research careers. For example, although we cannot scientifically graph the increased confidence we believe our career awards have given recipients, we believe that it is considerable.

The distinction of being a career award recipient – along with the other early-career honors and awards by foundations and government agencies — helps to ensure that their talent is not overlooked by prospective employers. Indeed, experience shows that employers view independent, peer-reviewed awards such as career awards as a "stamp of approval," and individuals who bring independent funding are typically highly attractive job candidates.

Also central to the art of nurturing scientific careers has been our efforts to foster a sense of community through regularly convening awardees. This brings our awardees into supportive relationships with fellow scientists who share a bond of being at the same stage in their career, as well as the opportunity to interact with senior scientists and policymakers who are willing to share their own career experiences. Our awardees tell us that the benefits of such a community include the opportunity to share experiences in career-building areas from negotiating faculty positions to grantsmanship to laboratory management. As our cadre of awardees grows, we hope that new recipients will be more likely to have senior awardees at their own institutions who hold faculty positions, as well as contacts made through awardee convocations.

Especially gratifying is that our community of awardees has already given rise to highly promising collaborations. An excellent example is the collaboration between James Olson and fellow awardee Matthias Gromeier, as discussed in the profile on page 6 on Olson's work. At a recent career awardee meeting, Gromeier discovered upon seeing Olson's poster display that a genetically altered poliovirus that Gromeier had developed to target and kill certain cancer cells might prove an effective treatment for the deadly childhood brain cancer, medulloblastoma. This collaboration also illustrates how the broad range of research areas represented by our awardees affords them a remarkable opportunity to meet colleagues doing work quite far afield from their own, and whom they would not likely encounter in the narrowly focused scientific meetings they normally attend.

While overall we have found much to be gratified in the Career Awards Program, we have also identified a few areas to expand future efforts.

For example, even though the awards do give our awardees a valuable boost in starting their careers, many remain inexperienced at negotiations for their first position. They may be willing to accept barely adequate start-up funding when they could command a more generous package that would give them a better chance at success. Also, first-time recruits seem not to push for ancillary benefits such as longer periods of guaranteed salary and relief from teaching loads in order to focus on research.

Attracting sufficient women and minority applicants to our program is also a continuing concern. Over the five years of the program, the number of women in our candidate pool has been good, but somewhat lower than their general representation among postdoctoral fellows, and we are working with the nominating universities to ensure that they include their best female and minority candidates in their nominations.

Finally, we continue to worry about whether we are reaching all appropriate administrators and senior faculty with information about the Career Awards Program. We want to reach the full range of institutions, from the major research universities to the smaller institutions, where talented young scientists might be found. We encourage postdoctoral fellows to apply to the Career Awards Program. Likewise, we encourage academic administrators, department chairs, and senior faculty to nominate their most talented postdoctoral fellows for career awards.

It is our hope that this report has demonstrated the value of providing funding and support for our most promising young biomedical researchers; for the future of the biomedical research enterprise, and indeed our nation's health, depends on the contributions of women and men such as those we have introduced here. ■

Ravi Allada, M.D.
Brandeis University
Molecular and genetic analysis of the circadian rhythm gene, Clock, in Drosophila

Oscar M. Aparicio, Ph.D.
Massachusetts Institute of Technology
Understanding the relationship of DNA replication to cell cycle control of cellular proliferation and chromosomal organization

Nenad Ban, Ph.D.
Yale University
Determination of the high resolution structure of the large ribosomal subunit

Gregory J. Beitel, Ph.D.
Northwestern University
Mechanisms that control and execute the cell movements and shape changes underlying metazoan morphogenesis

Mark Bix, Ph.D.
University of Washington
Effector CD4+ T cell development: evidence for the stochastic generation and clonal distribution of a combinatorial cytokine repertoire

Azad Bonni, M.D., Ph.D.
Harvard Medical School
Regulation of glial fate specification in the central nervous system

Edward S. Brodkin, M.D.
Princeton University
Genetic analysis of anxiety-related behaviors in mice

Paul S. Buckmaster, D.V.M., Ph.D.
Stanford University School of Medicine
Mechanisms of temporal lobe epilepsy

David C. Chan, M.D., Ph.D.
California Institute of Technology
Structural and mechanistic studies of virus-mediated membrane fusion

Thomas R. Clandinin, Ph.D.
University of California-Los Angeles School of Medicine
Dissecting neuronal target selection in the Drosophila visual system

Michael K. Cooper, M.D.
Johns Hopkins University School of Medicine
Modulation of sonic hedgehog signal transduction by cholesterol homeostasis

Anita H. Corbett, Ph.D.
Emory University School of Medicine
Components of the nuclear transport system

Brendan P. Cormack, Ph.D.
Johns Hopkins University School of Medicine
Virulence determinants of *Candida glabrata*

David E. Cummings, M.D.
University of Washington School of Medicine
Studies of spermatogenesis and metabolism using mutant mice

George Q. Daley, M.D., Ph.D.
Whitehead Institute for Biomedical Research and Harvard University School of Medicine
Probing the pathogenesis of chronic myelogenous leukemia

Jeanine D'Armiento, M.D., Ph.D.
Columbia University College of Physicians and Surgeons
The role of matrix metalloproteinases in disease

Gregory C. DeAngelis, Ph.D.
Washington University School of Medicine
Neural mechanisms underlying perceptual feature binding

Paul De Koninck, Ph.D.
Stanford University Medical Center
Decoding rhythms in the nervous system

Jeffrey S. Diamond, Ph.D.
Oregon Health Sciences University
Mechanisms underlying silent synapses: implications for synaptic plasticity in the hippocampus

Aaron DiAntonio, M.D., Ph.D.
Washington University School of Medicine
Genetic analysis of synapse formation, growth, and plasticity

Tamara L. Doering, M.D., Ph.D.
Weill Medical College of Cornell University
GPIs in fungi: biosynthesis and function

Michael J. Eck, M.D., Ph.D.
Harvard Medical School
Structural analysis of protein interactions in signal transduction

Guowei Fang, Ph.D.
Stanford University
Mechanism of the spindle assembly checkpoint control

Steven Fiering, Ph.D.
Dartmouth Medical School
Mutational analysis of the beta-globulin locus control region by homologous recombination

Elizabeth A. Finch, Ph.D.
Duke University Medical Center
Postsynaptic calcium signaling by inositol trisphosphate in neuronal dendrites

Reiko Maki Fitzsimonds, Ph.D.
Yale University School of Medicine
Distributed properties of synaptic plasticity in neural networks

Robert C. Flaumenhaft, M.D., Ph.D.
Harvard Medical School
SNARE proteins in platelet alpha-granule secretion

Catherine A. Fox, Ph.D.
University of Wisconsin Medical School
On the formation of domains of gene expression in eukaryotic chromosomes

Daved H. Fremont, Ph.D.
Washington University School of Medicine
Structural studies of antigen presentation and T cell activation

Timothy P. Galitski, Ph.D.
Massachusetts Institute of Technology
Genetic networks

Jeffrey S. Glenn, M.D., Ph.D.
Stanford University School of Medicine
Prenylation and viral assembly

Joseph A. Gogos, M.D., Ph.D.
Columbia University College of Physicians and Surgeons
Genetic analysis of connectivity in the mammalian olfactory system

Todd R. Golub, M.D.
Harvard Medical School
Models for the molecular pathogenesis of human leukemia

Patrick A. Grant, Ph.D.
Pennsylvania State University
Analysis of histone acetyltransferase/ transcriptional adaptor complexes in the regulation of gene expression

Michael Graziano, Ph.D.
Princeton University
From eye to hand: sensory-motor integration in the primate brain

Rachel Green, Ph.D.
Johns Hopkins University School of Medicine
Defining the peptidyl transferase center of the ribosome

Matthias Gromeier, M.D.
Duke University Medical Center
Principles of polio neuropathogenesis: exploiting poliovirus for the treatment of brain cancer

Gyorgy Hajnoczky, M.D., Ph.D.
Thomas Jefferson University Jefferson Medical College
Control of cellular function by calcium oscillations

Carrie Haskell-Luevano, Ph.D.
University of Florida College of Pharmacy
Determination of the physiological and pharmacological role of the brain specific agouti-related transcript

Zhigang He, M.D., Ph.D.
University of California-San Francisco School of Medicine
Signaling mechanisms mediating the repulsive effects on developing and regenerating axons

Xianxin Hua, M.D., Ph.D.
Massachusetts Institute of Technology
Identification and characterization of novel components in the TGF-ß signaling pathway

Alan J. Hunt, Ph.D.
University of Michigan
Role of microtubule dynamics in mitotic chromosome movement

Jeffry S. Isaacson, Ph.D.
University of California-San Diego School of Medicine
Biophysical properties of presynaptic terminals in the central nervous system

Raymond H. Jacobson, Ph.D.
University of California-Berkeley
TBP-related factor and selectivity factor I: probing TBP function in alternative contexts

David K. R. Karaolis, Ph.D.
University of Maryland School of Medicine
Pathogenicity islands in the emergence of epidemic and pandemic cholera

Suzanne L. Kirby, M.D., Ph.D.
University of North Carolina-Chapel Hill School of Medicine
Proliferative advantage for therapeutic bone marrow

Laura J. Knoll, Ph.D.
Stanford University Medical Center
Molecular genetic approaches to investigate developmental regulation in *Toxoplasma gondii*

Scott C. Kogan, M.D.
University of California-San Francisco School of Medicine
Use of a transgenic mouse model of acute promyelocytic leukemia to elucidate disease pathogenesis and to improve therapy

William L. Kraus, Ph.D.
Cornell University
Biochemical analysis of estrogen receptor activity with chromatin templates

Margarethe J. Kuehn, Ph.D.
Duke University Medical Center
Secretory protein sorting in yeast and bacteria

Peter D. Kwong, Ph.D.
Columbia University College of Physicians and Surgeons
An atomic-level investigation of the structure and associated biology of the HIV-envelope protein, gp120

Bruce T. Lahn, Ph.D.
University of Chicago
Systematic investigation of mammalian spermatogenesis

Jeh-Ping Liu, Ph.D.
Columbia University College of Physicians and Surgeons
Molecular mechanisms in neural crest specification

Hiten D. Madhani, M.D., Ph.D.
Massachusetts Institute of Technology
Specificity of signal transduction during dimorphic development of yeast

Mala S. Mahendroo, Ph.D.
University of Texas Southwestern Medical Center-Dallas
Characterization of fecundity and parturition defects in mice deficient in steroid 5 alpha-reductase type 1

Zachary F. Mainen, Ph.D.
Cold Spring Harbor Laboratory
Optical studies of synaptic plasticity mechanisms

Kelsey C. Martin, M.D., Ph.D.
University of California-Los Angeles School of Medicine
Communication between the synapse and the nucleus during long-lasting synaptic plasticity

Andrea I. McClatchey, Ph.D.
Harvard Medical School
The roles of NF2 tumor suppressor gene product in embryogenesis and tumorigenesis

Robert Menard, M.D., Ph.D.
New York University Medical Center
Genetic analysis of malaria sporozoite virulence

Chandra Mohan, M.D., Ph.D.
University of Texas Southwestern Medical Center-Dallas
B-cell tolerance and humoral autoimmunity

Kelle H. Moley, M.D.
Washington University School of Medicine
Glucose transport and apoptosis in blastocysts from diabetic mice

Louis J. Muglia, M.D., Ph.D.
Washington University School of Medicine
Adrenal and behavioral responses to stress

Charles E. Murry, M.D., Ph.D.
University of Washington School of Medicine
Muscle regeneration after myocardial infarction via gene therapy with MyoD

Inke S. Nathke, Ph.D.
University of Dundee
Role of APC/catenin complexes in colon cancer

James Olson, M.D., Ph.D.
University of Washington School of Medicine
NeuroD abrogation in neuroblastoma

Karen M. Ottemann, Ph.D.
University of California-Berkeley
Chemotaxis in *Helicobacter pylori* and its role in pathogenesis

George A. Oyler, M.D., Ph.D.
University of Maryland School of Medicine
Signaling pathways of neuronal apoptosis

Martin S. Pavelka Jr., Ph.D.
University of Rochester
Biosynthesis of the mycobacterial cell wall

Thomas Perkins, Ph.D.
Stanford University
Measurements of single DNA-based molecular motors

Martin R. Pollak, M.D.
Harvard Medical School
Mouse molecular genetic studies of the extracellular Ca^{2+}-sensing receptor

Salman T. Qureshi, M.D.
McGill University Faculty of Medicine
Genetic analysis of innate resistance to bacterial pathogens

Jill Rafael, Ph.D.
Ohio State University School of Medicine
The role of muscle proteins in synaptic structure and neuromuscular disease

Ilaria Rebay, Ph.D.
Massachusetts Institute of Technology
Investigation of yan function in Ras1-mediated signal transduction

Matthew Redinbo, Ph.D.
University of North Carolina-Chapel Hill
Structural and functional characterization of human topoisomerase I and the Werner syndrome helicase

David J. Robbins, Ph.D.
University of Cincinnati College of Medicine
Dissecting the hedgehog signal transduction pathway

Aimee K. Ryan, Ph.D.
University of California-San Diego
Analysis of inductive events responsible for specification and differentiation of the anterior pituitary gland

Sylvia L. Sanders, Ph.D.
Massachusetts Institute of Technology
Analysis of cell-type differences in bud-site selection in *Saccharomyces cerevisiae*

Maria A. Schumacher, Ph.D.
Oregon Health Sciences University
Structural biology of cell growth, development, and regulation

Konstantin V. Severinov, Ph.D.
Rutgers, the State University of New Jersey-Piscataway
RNA polymerase and its subunits: structure and function

Krishna V. Shenoy, Ph.D.
California Institute of Technology
Early reach plans in posterior parietal cortex

Judith A. Shizuru, M.D., Ph.D.
Stanford University School of Medicine
Transplantation of purified allogeneic hematopoietic stem cells: applications for transplantation tolerance induction and the treatment of autoimmune disease

Elaine K. Sia, Ph.D.
University of Rochester
Analysis of yeast mutants with altered simple repeat stability

Upinder Singh, M.D.
Stanford University School of Medicine
Transcriptional control in *Entamoeba histolytica*

Barry P. Sleckman, M.D., Ph.D.
Washington University School of Medicine
Lineage specific regulation of T-cell receptor beta rearrangement

Leonidas Stefanis, M.D.
Columbia University College of Physicians and Surgeons
Proteases in neuronal cell death

Theodore S. Steiner, M.D.
University of Virginia Health Sciences Center
Isolation and characterization of an interleukin 8 releasing protein from enteroaggregative *Escherichia coli*

David J. Sullivan, M.D.
Johns Hopkins University School of Hygiene and Public Health
Iron metabolism in *Plasmodium falciparum*

Surachai Supattapone, M.D., D.Phil.
University of California-San Francisco School of Medicine
The structure and biology of a soluble prion

Michael J. Thirman, M.D.
University of Chicago Medical Center
Functional analyses of the MLL-ELL fusion transcript in acute leukemia

Sarah A. Tishkoff, Ph.D.
Pennsylvania State University
Molecular sequence variation in G6PD and its role in malarial resistance

Katharine S. Ullman, Ph.D.
University of Utah
Biochemical dissection of RNA export using in vitro nuclear reconstitution

Peter C. VanBuren, M.D.
University of Vermont School of Medicine
Role of the thin filament in the altered contractility of heart failure

James A. Waddle, Ph.D.
University of Texas Southwestern Medical Center-Dallas
Generation of markers and methods to study asymmetric cell divisions in living *Caenorhabditis elegans* embryos

Johannes Walter, Ph.D.
Harvard Medical School
Characterization of eukaryotic replication initiation factors using a soluble, cell-free system

William A. Weiss, M.D., Ph.D.
University of California-San Francisco School of Medicine
A transgenic model for neuroblastoma

Carmen J. Williams, M.D., Ph.D.
University of Pennsylvania School of Medicine
Signal transduction mechanisms during mouse egg activation

Ding Xue, Ph.D.
University of Colorado-Boulder
Biochemical analysis of CED-3/ICE cysteine proteases and their roles in apoptosis

Michael Yaffe, M.D., Ph.D.
Harvard Medical School
Scaffolding and chaperone proteins in signal transduction: 14-3-3 regulation of mitosis and programmed cell death

Cassian Yee, M.D.
University of Washington School of Medicine
Adoptive immunotherapy of melanoma using T cells specific for potential tumor rejection antigens

Deborah L. Yelon, Ph.D.
University of California-San Francisco School of Medicine
Patterning during organogenesis: genetic analysis of cardiac chamber formation

John York, Ph.D.
Duke University Medical Center
Nuclear signal transduction: structure and function

BURROUGHS WELLCOME FUND

21 T. W. ALEXANDER DRIVE • POST OFFICE BOX 13901 • RESEARCH TRIANGLE PARK, NC 27709

TELEPHONE (919) 991-5100 • FAX (919) 991-5160

www.bwfund.org