BTI

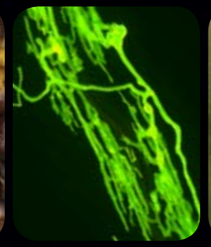
BOYCE THOMPSON INSTITUTE

Annual Report
2005

IN THIS ISSUE








LETTER FROM THE PRESIDENT

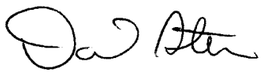
In the 17 years since I joined the Boyce Thompson Institute staff as an assistant scientist and in the 16 months since I had the privilege of assuming the presidency, BTI has seen a great deal of change. The sun has risen and set on research programs, long-time employees have retired and energetic new ones have arrived, and the Internet has poked its tentacles into nearly every corner of our scientific existence. The landscape of our enterprise has been forever altered.

Yet not everything has changed. The bricks and mortar are the same as they were in 1979 when BTI opened its Ithaca doors. Campus life still ebbs in December and June but returns in force in January and August. And young people are drawn to science for the same reason that appealed to da Vinci, McClintock, and Darwin: an intense curiosity about the natural world. This is why BTI's mission is as vital in 2006 as it was when Colonel Thompson presided over its inauguration in 1924.

It is wise and refreshing to examine and react to the apposition of the nearly timeless mission of basic research and the breathtaking pace of technological and political change. While the world moves forward, sometimes haltingly, in confronting poverty, hunger, and the splintering of geopolitical boundaries, we have witnessed a seemingly implacable opposition to real action on climate change and frequently have lost sight of the science behind plant transformation and new visions for agriculture. As we uncover the world of plants and, in so doing, train young minds for future endeavors, we reaffirm our mission, knowing that fertile minds and fertile fields are not unconnected.

In da Vinci's time and even now, private citizens with sufficient resources could chase their curiosity to the furthest corners of the planet and beyond. Steve Fosset's balloon adventures, Burt Rutan's SpaceShip One, and indeed BTI at its inception could operate in this rarified environment. However, today's BTI is different. We depend on public and private funds to pursue plant research. We are tightly integrated with our colleagues at Cornell and other institutions. And we seek to share with the public our excitement, our methods, and our findings.

As you will read in this report, we have taken new steps to educate children, teachers, and citizens about BTI's mission and how BTI research, often unpredictably, can lead to improvements in agriculture, nutrition, and even a vaccine against cancer. You will read stories about our scientists' discoveries and dreams. I hope that in 2006 you will listen to "MicrobeWorld," spend an evening at Science Cabaret, or join us for an event at the institute. We are proud of who we are and of who we are becoming.



David Stern

IN BRIEF

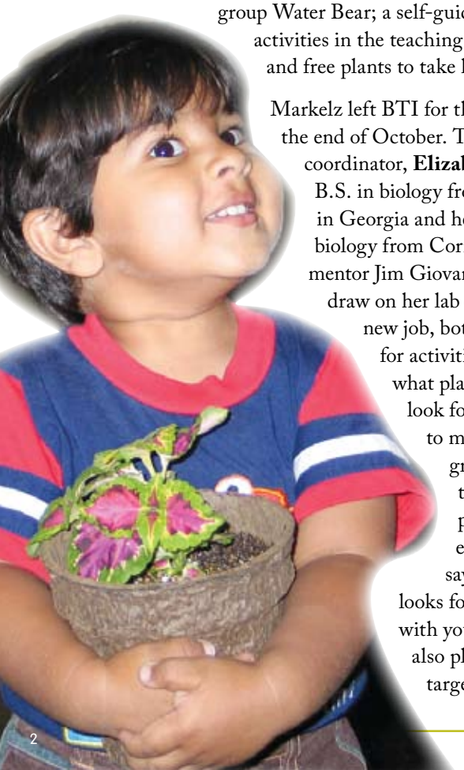
OUTREACH

Boyce Thompson Institute continued its popular after-school program at Ithaca's Northeast Elementary School in spring 2005 with weekly activities such as racing beetles, painting and tie dying with plant pigments, planting seeds, and measuring biodiversity. Outreach Coordinator **Nicole Markelz** also led numerous tailor-made activities for groups of teachers and students who visited the institute.

More than 90 undergraduates applied for 17 Plant Genome Research Program (PGRP) summer internships, double the number of 2004 applicants. The undergraduates worked for 10 weeks in labs at BTI and Cornell. Seven high school students also did research projects at the institute for six weeks. Each student worked with a mentor to complete an independent research project, which they presented at the Summer Student Symposium in early August. Skidmore College student **Leah Elliott**, who spent her summer in Cornell Professor Maureen Hanson's lab, won the Colonel's Cup for best talk. **Jen Brady**, a Cornell student who worked in Gregory Martin's lab, won the PGRP Poster Award.

The institute dedicated the Helen and George Kohut Teaching Laboratory in 2005, celebrating its opening and that of the renovated auditorium at A Closer Look at BTI on September 7. The open-house event included a slideshow of BTI- and plant-related images in the auditorium, accompanied by local musical group Water Bear; a self-guided greenhouse tour; activities in the teaching lab and elsewhere, and free plants to take home.

Markelz left BTI for the Silicon Valley at the end of October. The new outreach coordinator, **Elizabeth Fox**, earned her B.S. in biology from Mercer University in Georgia and her Ph.D. in plant biology from Cornell University, under mentor Jim Giovannoni. Fox plans to draw on her lab experience in her new job, both to give her ideas for activities and to convey what plant research is like. "I look forward to speaking to many different age groups and showing them how important plant science is in their everyday lives," she says. She particularly looks forward to working with younger children and also plans to start a program targeted to local seniors.



HONORS AND AWARDS

Robert Abramovitch was the first-ever recipient of the Barbara McClintock Award, which recognizes "a graduate student with the best potential and greatest background merit" in the plant sciences at Cornell. He has co-authored several high-profile publications, including a recent paper in *Science*.

Thomas Bollenbach and **Rongcheng Lin** won the Lawrence Bogorad Plant Molecular Biology Award in 2005. The award is given annually to BTI postdoctoral fellows who demonstrate excellence in research.

BTI researcher **Thomas Brutnell** was promoted to associate scientist with tenure in December 2005. He joined the institute as an assistant scientist in 1999 and has developed an internationally recognized program in maize genetics. Brutnell studies light signaling and is creating transposon mutant collections to serve scientists throughout the maize community.

Graduate student **Liza Conrad** won the Munger/Murphy Award from Cornell's Department of Plant Breeding and Genetics for her "outstanding research achievements, strong contributions to the teaching program, and active participation in departmental activities."

Gregory Martin was elected a fellow in the American Academy of Microbiology in recognition of his research achievements in the field.

NATIONAL AGRICULTURAL BIOTECHNOLOGY COUNCIL

The National Agricultural Biotechnology Council (NABC), which operates from the Boyce Thompson Institute, used its 2005 annual meeting to focus on the relationship of agricultural biotechnology to health and the environment.

The NABC, founded in 1989 by then-BTI President Ralph Hardy, plans its annual meeting as a forum for scientists, policy makers, and other interested parties to discuss issues related to agricultural biotechnology. NABC also co-hosts the annual World Congress on Industrial Biotechnology and Bioprocessing. The proceedings of both meetings are published and distributed worldwide.

NABC's 34 member institutions include the majority of U.S. land-grant universities, two Canadian institutions with large agricultural research programs, and two branches of the U.S. Department of Agriculture.

The 2006 NABC annual meeting, on job creation and workforce development, will be hosted by Cornell University in June.

BTI AND THE ENVIRONMENT

Through projects such as installing new greenhouse temperature controls, replacing old growth chambers and building air flow and heating controls, and putting lights on timers, the institute has reduced its energy use dramatically since 2002. Electricity use is down 29 percent, chilled water consumption (for cooling) has decreased 41 percent, and the use of steam for heating is 71 percent less. BTI also transitioned from the use of plastic and foam plates, cups, and cutlery to disposables made from materials derived from corn or wheat that are biodegradable.

The recently installed Argus Greenhouse Control System provides tight control over lighting, with sensors that turn lights on during cloudy days and off when the sun comes out. The computerized system also controls temperature and humidity and is expected to lower the cost of running the greenhouses by $20,000 to $30,000 per year.

Summer 2005

Fall 2005

REMODELED AUDITORIUM

The renovation of the auditorium is now complete, with new paint, new carpet, reupholstered seats, an upgraded lighting system, and a modern projection booth. The new podium contains state-of-the-art audio-visual equipment.

IN BRIEF

DEPARTURES

Alice Churchill worked at BTI from 1993 through January 2005, first as a postdoctoral fellow, then as a Molecular Mycology Center scientist. She continues research on plant and insect pathogenic fungi as a visiting scientist in the Cornell Department of Plant Pathology. During her time at the institute, she studied the genetic basis of secondary metabolism in several fungal pathogens. Her research group was the first to develop *Agrobacterium*-mediated gene silencing methods for three fungi infecting American chestnut, bananas, and insects and use these methods to identify fungal genes that cause disease. They also demonstrated that a metabolic pathway believed crucial for the banana pathogen to cause disease was not important, thereby refocusing research efforts on other possible disease mechanisms.

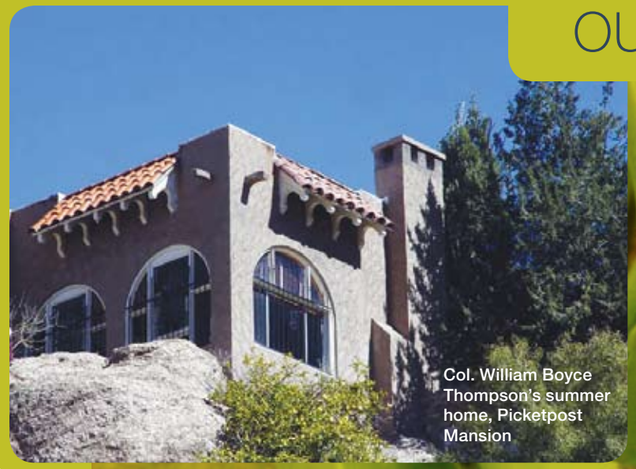
In collaboration with U.S. Department of Agriculture colleagues, Churchill's group used genetic approaches to uncover two families of secondary metabolites not previously reported from the insect pathogen, and they now study the functions of these compounds in the fungus. Churchill is also a member of a consortium sequencing the genome of the black Sigatoka pathogen of bananas.

Scientist **Robert Kohut** retired from BTI at the end of 2004. He had worked at the institute since 1980, researching plant responses to changes in air quality. In addition to air pollution experiments at the BTI field site in Ithaca, he conducted two research projects at remote locations—an assessment of the effects of nitrogen oxides on tundra vegetation at Prudhoe Bay, Alaska, and a project to identify ozone-sensitive species and assess the impacts of ozone on plants at Acadia National Park in Maine. He also served on the institute's management team as director of operations from 1991–96. Kohut's recent research examined the effects of escalating levels of carbon dioxide on the physiology and population genetics of plants. He recently completed an assessment of the risk of ozone injury to plants in 270 national parks and developed a handbook for the National Park Service on assessing foliar ozone injury to plants in the field.

Associate Research Scientist **David Weinstein** left the institute at the end of June after 14 years and is now a senior research associate in the Cornell Department of Natural Resources. Weinstein established and led the Plant Modeling Group, which discerned the connections between basic plant properties, characteristics, and functions and the behavior of plant communities and ecosystems. The group developed simulations and analytical techniques for gleaning pattern from complex data sets. Weinstein created TREGRO, a model of tree physiology and carbon processing, and applied it to a wide variety of tree species to identify how trees' short-term use of carbon determines annual and decadal differences in the survival of stressed trees. His group then linked the model to one developed to simulate ecosystems in order to forecast long-term ecological outcomes. With these tools they predicted how cellular impacts of ozone will affect more than 70 different forests across the United States. The Plant Modeling Group's methods and models were adopted by the U.S. Environmental Protection Agency as the standard for investigating effects on forests.

BOYCE THOMPSON ARBORETUM

"I want visitors to have an **outstanding** time during their visit."



Col. William Boyce Thompson's summer home, Picketpost Mansion

Mark Bierner (above, with his wife, Cassandra James) joined the Boyce Thompson Arboretum (BTA) as director in August 2005. He had been a lecturer at the University of Texas at Austin since 1999 and served as executive director of Marie Selby Botanical Gardens in Sarasota, Florida, before that.

Bierner's immediate goal is straightforward: "I want visitors to have an outstanding time during their visit and to leave the arboretum feeling as if they have had a truly exceptional experience," he says.

William Boyce Thompson founded BTA in Superior, Arizona, in 1929. The 320-acre desert garden and botanical collection is now a state park, with more than two miles of paths and shaded trails for visitors. Weekend tours explore themes such as "Plants of the Bible" and "Edible and Medicinal Plants of the Desert." More information at 520-689-2811 or http://ag.arizona.edu/bta.

Three generations of the Villegas family learn about agave plants, lizards, birds, and hundreds of *Opuntia* varietals in the Cactus Garden.

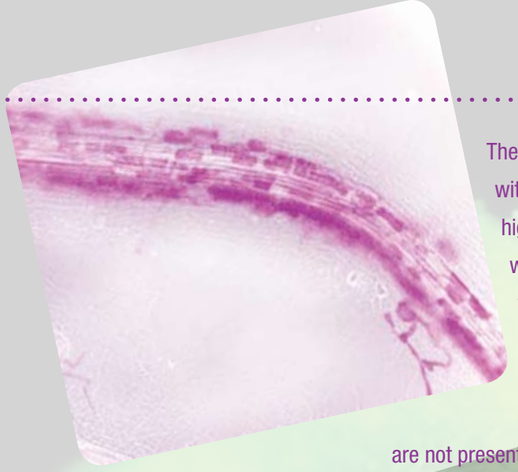
Glowing Results

Fluorescence microscopy reveals internal processes

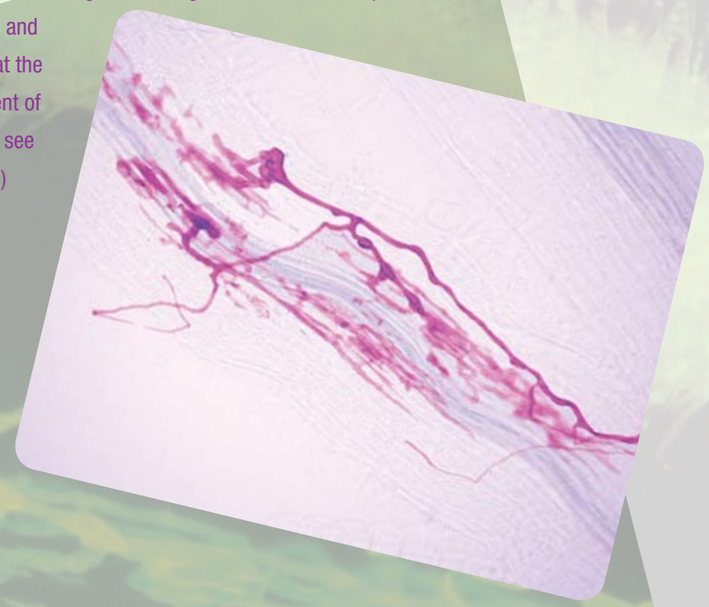
Molecular plant research is rarely a photogenic pursuit. Its scientists track genes and proteins that are far too small to be photographed directly, and the tools of the trade—plastic micropipettors, bullet-sized test tubes, powerful databases—bear little resemblance to the colorful bubbling flasks or amber-encased mosquitoes of science fiction movies.

Enter fluorescence microscopy. Paired with a judicious use of fluorescing compounds, the microscope enables researchers to look inside living things to see how they work. For example, scientists can tag a protein with green fluorescent protein (GFP), a molecule first discovered in jellyfish, to see where in the cell or organism the protein works.

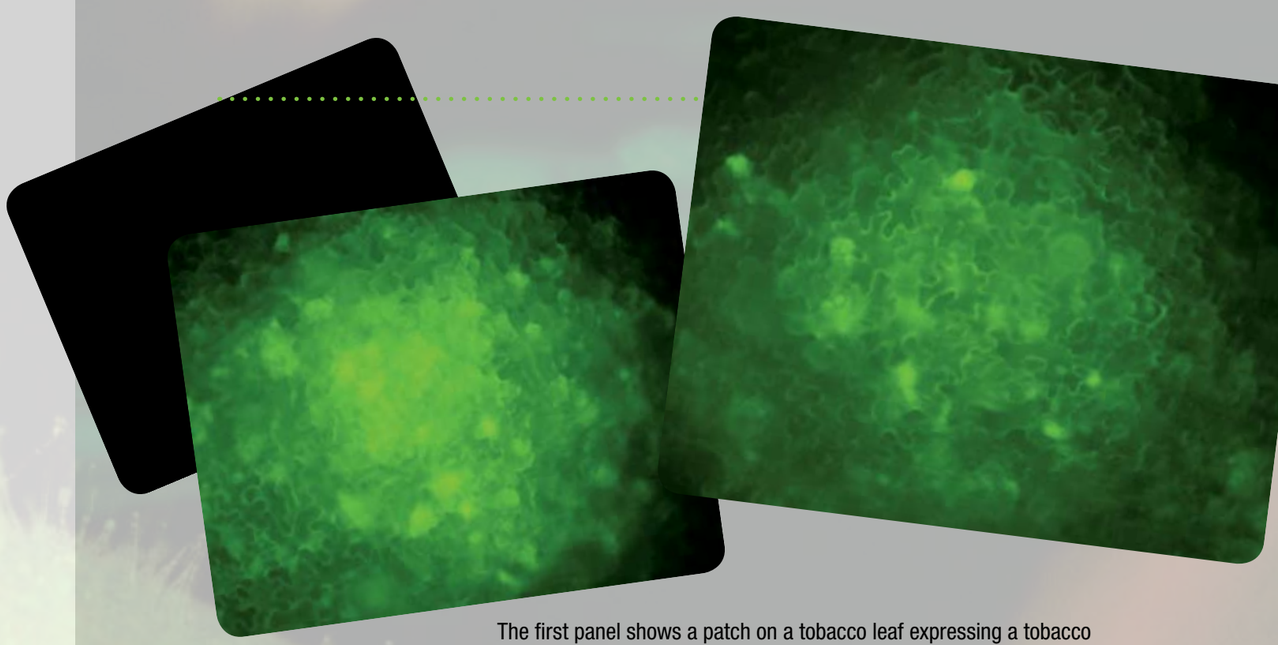
Used with other techniques, fluorescent microscopy gives BTI researchers a window into a host of processes, as these images illustrate.



The left image shows a *Medicago truncatula* root associated with symbiotic soil fungi *Glomus versiforme*. The stain highlights the fungal structures within the plant cells, while the root itself is translucent. In the right image, the *Medicago* root has had the gene for one enzyme "knocked out." The fungi living in this root show a different pattern of growth, and the structures that enable the fungi to exchange nutrients with the plant are not present. This and other experimental evidence indicates that the targeted enzyme is required for development of a normal symbiosis. (For more information see "Subterranean Choreography" on page 16.)



These images show *Arabidopsis* plants in which the protein HFR1 has been tagged with GFP. Both plants were left in the dark for four days, but the plant on the right was then exposed to white light for four hours before the picture was taken. The green spots in the right plant show a buildup of GFP-tagged HFR1 in the cells' nuclei. This quick accumulation of HFR1 turns on genes in the nucleus that are required for light response. (For more information see "Illuminating Decisions" on page 19.)



The first panel shows a patch on a tobacco leaf expressing a tobacco resistance protein N, protein P50 from Tobacco Mosaic Virus, and the Potato Virus X (PVX) genome tagged with GFP. N recognizes P50 and induces a resistance response that eliminates PVX, leaving no detectable GFP. The next two images confirm that the N/P50 interaction is key to this response: the second image shows a leaf with the gene for N but not P50, while the third leaf has P50 but not N. PVX was able to spread through both. (For more information see "Smart Proteins" on page 18.)

This figure shows a spore of an arbuscular mycorrhizal fungus that has been treated with DAPI, a dye that stains the nuclei. This image and a series of other images through the spore enabled members of the Harrison lab to estimate the average number of nuclei in a *Glomus intraradice* spore.

PROTEIN FACTORIES

Work on insect cells yields an unforeseen payoff.

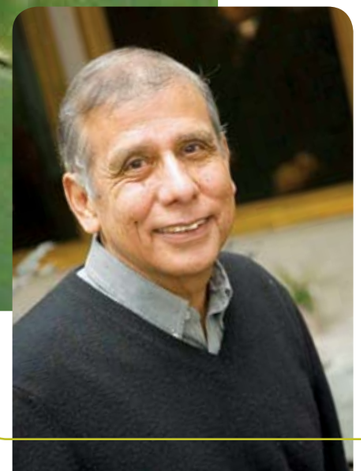
the Granados laboratory's project took an unexpected turn. For years in the mid-1980s the lab tried to develop an insect cell culture system for use in studying a type of virus. Scientist **Bob Granados** later used one of the lines to devise a system for producing large amounts of protein. Now GlaxoSmithKline is using that system to produce a vaccine with the potential to prevent millions of cervical cancer cases worldwide.

Scientists had long been interested in baculoviruses because of their potential as biological pesticides—since they liquefy rapacious moths but don't affect people, a modified baculovirus could be a safe and effective alternative to synthetic chemicals. To study baculoviruses, researchers developed insect cell lines in which to grow them. But the system only worked for one type of baculovirus, while granuloviruses—the kind Granados wanted to study—wouldn't grow in the culture. So his lab set out to develop a line that would work. They extracted cells from cabbage looper embryos and coaxed them into growing in flasks of nutrient-rich solution.

Some of the hundreds of cell lines that the Granados team developed were susceptible to granuloviruses at first but grew resistant after several cell generations. The lab eventually abandoned the project, but first they tested the cell lines with other viruses. Granados froze several of the lines that proliferated quickly, were easy to work with, and were highly susceptible to infection by a commonly studied type of baculovirus.

A few years later, Granados began working with Cornell chemical engineering professor Mike Shuler to devise a system for large-scale production of recombinant proteins. Two recent papers from other laboratories had shown that a baculovirus could be modified to carry DNA encoding a desired protein into insect cells, effectively turning them into factories for that protein.

"I was the only person on the Cornell campus who had the expertise, a collection of insect cell lines, and an active lab developing novel cell lines," Granados explained. "So we decided to test a few of the frozen cell lines and determine whether any were superior to the lines that had already been published."



"I know that one of the discoveries from my lab is going to benefit people. I'll see it in my lifetime. And that's very satisfying."

OTHER BTI DISCOVERIES

In addition to High Five cells, many other patented BTI discoveries soon may be applied to real-world problems.

DOW Agrosciences licensed three BTI patents on methods to produce pharmaceuticals in cultured plant cells. The company plans to market a poultry vaccine for Newcastle disease beginning in mid-2006, making it the first-ever plant-made vaccine to earn government approval.

Nektar Therapeutics is using a drying technique developed by **Carl Leopold** to make an inhaled insulin for diabetics. The FDA approved the powdered insulin in 2006.

BTI recently filed a patent on a discovery from **Greg Martin's** lab that has potential applications in tomatoes, people, and everything in between. The lab found that bacterial protein AvrPtoB inhibits programmed cell death (PCD), a defense response in which an organism kills off its own infected cells to keep disease from spreading. Since runaway PCD is responsible for problems ranging from unsightly spots on fruit to human autoimmune disorders, this finding could have broad implications.

Granados thawed out the lines from his prior experiment and infected them with a modified nucleopolyhedrovirus, a type of baculovirus that grows well in insect cell cultures. It worked. One of the lines produced more of the desired protein than any other cell line they tested and many times more than the cell lines in published papers. Most importantly, the infected cells produced protein more efficiently than other commonly used systems such as bacteria, yeast, and cultured human cells.

Granados and Shuler knew that their results wouldn't be enough to entice most researchers to adopt the insect cell/baculovirus system, however. "It's always very difficult to introduce a new piece of technology into the scientific community, because scientists are used to using certain cells, certain media, and certain pieces of equipment, and it's hard to make them change," Granados says. The barrier would be even greater for drug makers and other companies, which have expensive and long-established infrastructures devoted to harvesting proteins from bacteria, cultured human cells, or even (in the case of flu vaccine) chicken eggs.

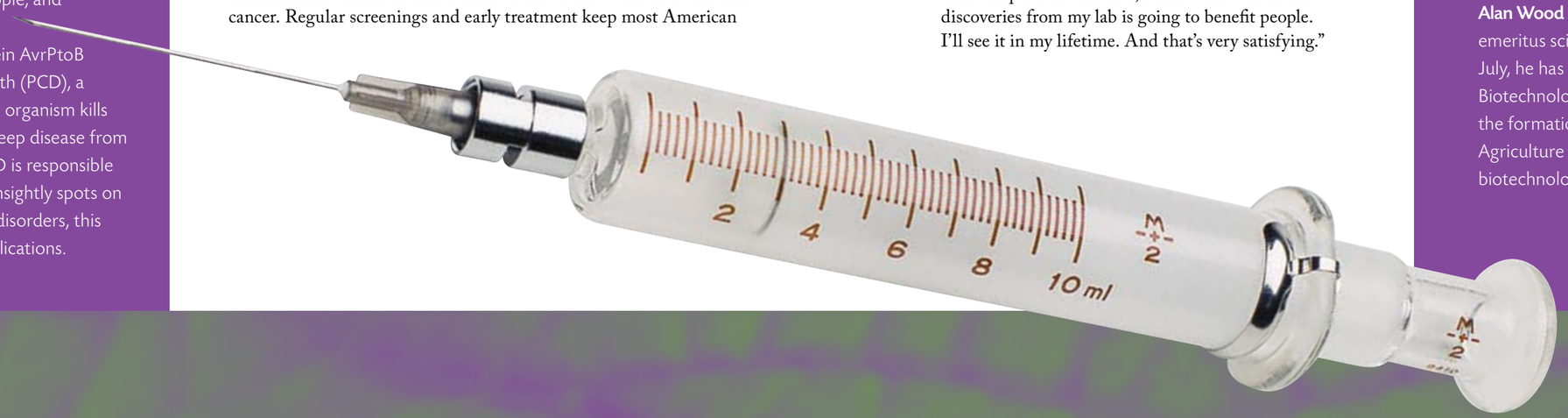
To convince researchers of the merits of their technique, the Shuler and Granados labs set out to design a way to scale up the new production system—that is, to grow the cells at high volumes. Shuler, Granados, and colleagues published many papers demonstrating the cells' potential. In 1994, Granados and BTI licensed the cell line to a biotech company, Invitrogen, which commercialized the cells for academic research use. The company named the line "High Five" and advertised it in scientific journals. Many scientists began to use the High Five system to make proteins that they wanted to study. And the new intellectual property office at BTI fielded calls from companies interested in licensing High Five for commercial research and production.

One of these companies was Medimmune, which was trying to develop a vaccine for genital human papilloma virus (HPV). About 80 percent of women will contract this sexually transmitted virus in their lifetimes. If not detected some strains of HPV can cause cervical cancer. Regular screenings and early treatment keep most American women safe, but more than 10,000 still develop cervical cancer each year, and nearly 4,000 die. HPV takes a higher toll on women with little access to health care: worldwide, 230,000 women die of cervical cancer each year. Medimmune licensed the High Five system to produce HPV proteins for a vaccine that would train immune systems to kill the virus.

The results were promising, and Medimmune later teamed up with GlaxoSmithKline (GSK) to produce and test the vaccine, which they called Cervarix. Phase II clinical trials showed Cervarix to be 100 percent effective in healthy young women, and phase III results have shown similar success thus far. According to Luc Fabry, the director of technical operations for Cervarix, the High Five system has proven to be a good one for making the vaccine. "It provides high-expression yields, the system is quite robust and receptive to scale-up, and it's a cost-effective process," he says.

GSK submitted Cervarix for registration in Europe early in 2006, beginning the approval process there. But the company plans ultimately to make the vaccine available to women worldwide, according to GSK spokesperson Pamela Duncan.

Now an emeritus scientist, Granados has seen his work applied in ways he never anticipated when he set out to study granuloviruses in insect cell lines. "I spent almost 40 years doing research that I hoped was somehow going to benefit society and mankind," he reflects. "The vast majority of scientists feel this way, but most of us never know if anything we do in our career will have a direct impact. In this case, I know that one of the discoveries from my lab is going to benefit people. I'll see it in my lifetime. And that's very satisfying."



ACTIVE EMERITI

Emeritus Scientist **Robert Granados** continues to work at BTI as a licensing consultant. Here's what other emeriti were doing in 2005.

A. Carl Leopold remained active in several environmental organizations and gave talks in New York and Wisconsin. He also gave guest lectures in Cornell courses in horticulture and in neurobiology and behavior. And he displayed ceramic masks in "Out of Africa," an art show at a local gallery.

Alan Renwick reviews and edits papers for the *Journal of Chemical Ecology*. He published two papers in 2005 on plant chemistry and insect food choice based on taste and smell. He also researched the history of the 20-year BTI program in Grass Valley, California, that examined bark-beetle attacks on pine trees. The local land trust is renovating the buildings used for the program.

Richard Staples serves as an adjunct professor in the Cornell Department of Plant Pathology and as the MiniReviews editor for *FEMS Microbiology Letters*. Staples co-authored a research paper in 2005 based on a gene that he cloned in the 1980s. He and **Leonard Weinstein** completed *A Personal History of Boyce Thompson Institute: 1974 to 2000* (available for download on the BTI web site). Weinstein also co-authored a chapter entitled "Effects of Fluorides on Plants, Animals, and Other Organisms" for the first volume of the upcoming book *Advances in Fluorine Chemistry*. He works as a consultant for a new aluminum smelter in Reydarfjordur, Iceland, writes short stories, and creates stained-glass panels.

Alan Wood retired from BTI in 2001 and became an emeritus scientist in 2005. Since returning to Ithaca in July, he has helped organize the National Agricultural Biotechnology Council's 2006 meeting, promoted the formation of a National Institute for Food and Agriculture in Washington, D.C., and consulted for biotechnology companies.

IN THE LABS

THE ENEMY WITHIN

Viruses need to evolve fast to stay one step ahead of their hosts' defenses. One shortcut they use is to steal genes from their hosts, then modify them for their own purposes.

Virologists know this because they have found many analogs of viral genes in the genomes of their hosts. Baculoviruses multiply in the nuclei of insect host cells. While replicating, they sometimes mix their DNA with that of their insect host, giving them a chance to copy or steal a host gene before departing for fresh prey.

One shortcut is to STEAL GENES from their hosts, then modify them for their own purposes.

Many mysteries remain surrounding viral evolution, however, including the genesis of the baculovirus fusion proteins that the laboratory of **Gary Blissard** studies.

Called F and GP64, these proteins are the virus's Swiss Army knives. They allow the virus to fuse with and gain entry to hosts by binding to proteins on the cell surface, and later in the infection cycle they help assemble new viral particles and get them out of the cell.

When the fruit-fly genome was sequenced, it revealed several genes that resembled the gene for the viral F protein. While most of those genes were left behind by viruses called retroviruses, one appeared to be a genuine host gene. If this turned out to be a real, functioning host gene, it would suggest that the *f* gene originally may have moved from an insect to a baculovirus, not the other way around.

To see if this was the case, Blissard's lab investigated whether the fruit fly *f* gene was actually being expressed, and they found that it was. The lab also observed that unlike viral F proteins, which are found on the surfaces of infected cells and later on the virus surface, host F proteins were found in small structures inside the cell.

Blissard speculates that picking up the *f* gene could have enabled baculoviruses to go from causing mild symptoms to aggressively liquefying their hosts within days. Thus it might have been a host's own gene that turned baculoviruses into the malevolent killing machines they are today.

JUMPING GENES

Efforts to sequence species' genomes get a lot of press, but by itself a genome sequence is merely a long string of letters, often with vast stretches containing no genes at all. Making sense of that data requires considerable effort in investigating where genes are and what they do—a job that **Tom Brutnell** is working to make a little easier for maize researchers.

For corn, in contrast to many model organisms, "no simple, inexpensive resource has been developed to knock out genes of interest," Brutnell says. That's why he and two colleagues at Iowa State University have set out to create and catalogue a library of seeds carrying mutations. Scientists will be able to order these seeds and use them to investigate a specific gene's role in the plant.

Brutnell's lab creates these mutations using unusual stretches of DNA called transposons, or "jumping genes." As their name suggests, jumping genes can hop from one place in the genome to another. If they happen to land inside a gene, that gene can't function anymore.

To harness this phenomenon, Brutnell incorporated some of the findings from Barbara McClintock's early work using a transposon called *Ds*. *Ds* can't move unless helped by a protein encoded by another gene, *Ac*. Members of the Brutnell lab cross the lines carrying *Ds* with the lines carrying *Ac*. They then screen the offspring to identify those kernels in which the *Ds* has hopped into a new gene and determine which gene that is. The team's five-year goal is to generate and map 10,000 of these potentially useful insertions—knocking out up to 20 percent of all maize genes—and make them available to any researcher for a small fee.

The Brutnell lab starts with a line of corn that has a *Ds* transposon interrupting the gene for purple color. Purple kernels in its offspring indicate that *Ds* has jumped to a new location.

EXPLORING THE GENOME

Most scientific exploration is much less telegenic than planting a flag on the moon or tracking whales on the high seas. Molecular biology is no exception—its giant leaps usually appear as lines on gels or colored dots on microarrays. But when **Jim Giovannoni**, **Joyce Van Eck**, and their collaborators proposed an international endeavor to sequence the tomato genome, they drummed up support by promising countries a prize befitting the occasion: the chance to put their flag on one of tomato's 12 chromosomes.

This will be the first fruit or vegetable genome sequenced. The sequence will yield information not only about tomato and other members of the *Solanaceae* (nightshade) family—including potato, pepper, eggplant—but also other related species such as coffee and sunflower.

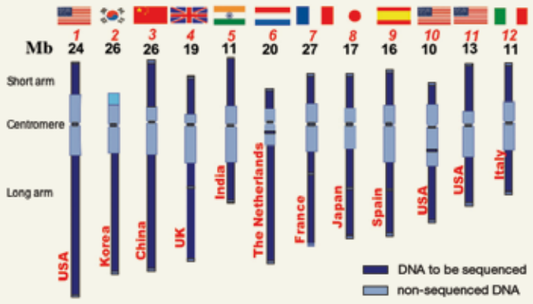
The potential agricultural benefits weren't quite enough to entice some nations to participate, so the U.S. team structured its proposal so that each country would be in charge of sequencing a discrete piece of the genome. "If these countries had each given a little money to the project, they wouldn't have gotten much credit," Giovannoni explained. "With this approach, each of them can point to a chromosome and say, 'we did that.'"

The U.S. team—Giovannoni and Van Eck at BTI, two laboratories at Cornell, and one at Colorado State University—received funding late in 2004 to lay the foundation for the project. Soon afterward, labs and funding agencies in nine other countries each agreed to take on a chromosome. Tackling the project this way ruled out shotgun sequencing, in which the genome is cut into small pieces and sequenced, with the sequence information then pieced together into the complete genetic code. Instead, the U.S. team would have to cut the genome into larger DNA segments and determine which chromosomes those segments belonged to before they were sequenced. Fortunately, previous studies had shown that most tomato genes are clustered near the ends of chromosomes in areas called euchromatin islands. The team made the project manageable by concentrating on these islands, leaving out vast stretches of DNA that offered little useful genetic information.

Giovannoni believes that the information contained in the euchromatin will help his lab learn more about fruit ripening and nutritional quality in tomato. His lab's role in the sequencing is to isolate the DNA and break it into large chunks. They then paste the chunks into vectors for U.S. collaborators and labs overseas to work with.

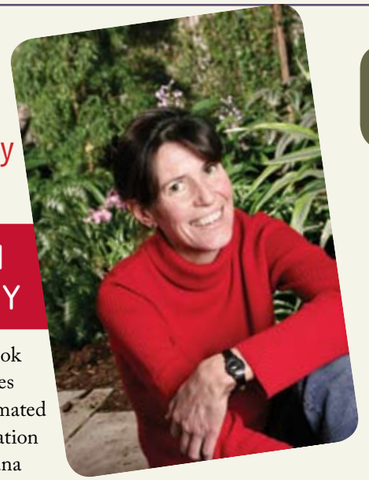
Van Eck manages the project, communicating with researchers around the world, coordinating shipments of DNA, and writing reports. She edits the *SOL Newsletter*, which keeps researchers apprised of sequencing progress and other information of interest to the *Solanaceae* community. Van Eck also runs the outreach component of the sequencing project, including a summer bioinformatics internship that blends biology and computer science. She hopes the completed sequence will yield information helpful to her work on antioxidant accumulation in potatoes.

Giovannoni's and Van Eck's U.S. collaborators determine where and on which chromosome each section of DNA belongs and later process the sequence information from abroad and make it available on a web site. The team recently submitted a proposal to the National Science Foundation to sequence the three remaining tomato chromosomes. If all goes well, the tomato genome should be unlocked sometime in 2008. In addition to helping scientists understand economically important nightshades, the sequence should yield clues to the puzzle of how such genetically similar plants evolved very diverse traits.



Tomato DNA is bundled into 12 chromosomes, each of which is assigned to a country for sequencing.

UNDERGROUND
some plants and fungi forge a cozy relationship



SUBTERRANEAN CHOREOGRAPHY

At first glance, plants and fungi look about as amicable as the Montagues and Capulets. Fungal strains decimated the American chestnut tree population and now threaten the world's banana supply. But underground, some plants and fungi forge a much friendlier relationship.

In fact, arbuscular mycorrhizal (AM) fungi partner with most plants worldwide, helping them extract phosphate, nitrogen, and other minerals from the soil in exchange for carbon. When plant roots sense AM fungi nearby, they create a welcoming home by ramping up production of proteins from some genes and suppressing others. This change in gene expression keeps the fungus from being destroyed by the plant's defenses and encourages it to form arbuscules, the specialized structures through which the two species trade minerals and carbon.

To find out more about this sophisticated partnership, **Maria Harrison** studies the interaction of alfalfa relative *Medicago truncatula* with several AM fungal varieties. Using microarrays—glass slides that can detect RNA from numerous genes—her lab compares the expression levels of *Medicago* genes before and after the fungi move in. This yields a snapshot of which genes might be needed to forge the symbiosis; the Harrison lab and collaborators have identified close to 900 such genes thus far.

The next step is to comb through those genes to find the few that are crucial for symbiosis. To do this, the lab knocks out candidate *Medicago* genes one by one and looks for a disruption in the resulting plants' interaction with AM fungi. The group recently used this process to identify a calcium-dependent enzyme needed for root development and for symbiosis with AM fungi and certain beneficial bacteria. The discovery indicates that calcium may be an important signal for both kinds of symbioses. Through this and other findings, Harrison and her collaborators continue to piece together the delicate subterranean choreography that goes into a good AM working relationship.

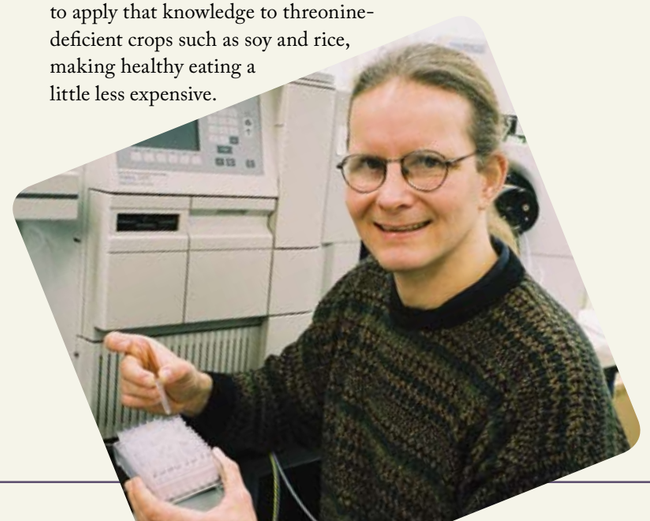
GOOD FOR YOU, BAD FOR *ARABIDOPSIS*

For college students, tight finances often motivate a diet of ramen noodles. For farmers faced with a tough bottom line, skimping on livestock feed isn't an option: while using just one crop for fodder would be cheap and simple, their animals need all of the essential amino acids to stay healthy. Since any one plant cannot provide all of the amino acids in sufficient quantities, farmers fill in the gaps with supplements, at a cost of more than a billion dollars a year in the United States.

Developing crops with elevated levels of needed amino acids could cut down on those costs, but to engineer fortified foods, scientists need to know more about the pathways that plants use to make the compounds. (Like animals, plants need 20 amino acids to make vital proteins, but unlike animals, most plants can make them all). To better understand the pathways and how tinkering with them might affect other processes in plants, **Georg Jander** is working with his lab to try to make an *Arabidopsis* plant that contains more threonine. In *Arabidopsis* seeds, the enzyme threonine aldolase normally converts most threonine into another amino acid, glycine.

The scientists in Jander's lab tried knocking out a gene for threonine aldolase, but found that this killed the plant. They then tried making a plant that had no threonine aldolase but extra threonine deaminase, an enzyme that converts threonine into yet another amino acid. The doubly mutated plant appeared healthy, indicating that excess threonine (not lack of glycine) probably killed the plants with only threonine aldolase knocked out.

If the lab can find a way to make healthy, threonine-rich *Arabidopsis*, other researchers might be able to apply that knowledge to threonine-deficient crops such as soy and rice, making healthy eating a little less expensive.



SENDING OUT AN SOS

When a virus, bacterium, or other pathogen attacks a leaf, the whole plant goes on heightened alert, readying itself to take on all attackers. Plant researchers have long puzzled over the signal that the infected part of the plant uses to warn the rest of impending danger.

The signal would probably travel through the phloem, the specialized system that transports nutrients throughout the plant—but what is the signal? One promising candidate was salicylic acid (SA), a molecule—as **Dan Klessig** and his lab have shown—that is involved in the general defense response provoked when part of a plant is threatened. But experiments seemed to rule out SA as the so-called phloem mobile signal.

Klessig's lab wasn't looking for the phloem mobile signal when they identified several proteins that interact with SA. Instead, they wanted to learn how SA's presence gets detected and ultimately affects the expression of defense-related genes—a process known as a signaling cascade. Proteins that bound to SA were likely to be part of this cascade, they thought.

Klessig and his team found several such proteins, including SABP2, which was present in such low levels that it took the lab years to purify enough to clone its gene and determine its three-dimensional structure. When they crystallized SABP2 and used X-rays to reveal its shape, they discovered something strange. SA itself was bound to SABP2's active site—that is, the part of the protein with the ability to chemically alter a molecule and thus pass along a signal. But this didn't seem to make sense if SABP2 was part of a signaling cascade that started with SA.

Instead, they found, SABP2's job is to lop off a methyl group from methyl salicylate, thereby converting it to SA. Klessig hypothesizes that methyl salicylate could be the long-sought phloem mobile signal—methyl salicylate sent through the phloem later could be converted to SA and thus set off a defense response. His group plans to test this hypothesis in 2006.

PLANT RESEARCHERS have long puzzled over the signal that the infected part of the plant uses to warn the rest of impending danger.

STEALTHY ATTACKERS

Millions of years of evolution have equipped viruses, bacteria, and other attackers with clever ways to slip past a plant's defenses. One strategy recently studied by **Greg Martin** and his lab is molecular mimicry: evolving proteins that mimic plant proteins and can alter the plant's physiology for the benefit of a pathogen.

Martin's lab recently found two instances of this mimicry. In the first case, graduate student Jeff Anderson was looking for ways in which infection by the bacterium *Pseudomonas syringae* affects tomato cells. Specifically, he wanted to find whether the *Pseudomonas* protein AvrPto, which makes the bacteria more virulent, influences phosphorylation of host proteins, since attaching or taking away a phosphate group is a common way to change protein function.

He discovered instead that AvrPto itself gets phosphorylated by a host protein, but not by chance—a phosphate group is added at a specific amino acid. Mutating AvrPto so that it cannot be phosphorylated reduces the ability of AvrPto to promote plant disease. This observation suggests that a host enzyme specifically recognizes AvrPto and adds the phosphate. For that to happen, AvrPto likely mimics a host protein on which the enzyme normally acts.

In a second case of molecular mimicry, graduate student Rob Abramovitch was looking for tomato proteins that interact with AvrPtoB, another *Pseudomonas* virulence protein. He found ubiquitin, a small protein that plant cells commonly use to mark unneeded proteins for degradation.

Why would AvrPtoB bind to ubiquitin? Part of the answer came when Abramovitch found that AvrPtoB acts as a ubiquitin ligase, a host enzyme that attaches ubiquitin to host proteins. A collaborator on the project showed that the three-dimensional crystal structure of AvrPtoB looks like a typical host ubiquitin ligase.

Martin and Abramovitch speculate that AvrPtoB makes plants more susceptible to disease by attaching ubiquitin to plant proteins involved in defense responses, thus tagging them for demolition.

IN THE LABS

SMART PROTEINS

People have it pretty good. With an army of specialized white blood cells to patrol our bodies and kill off germs, it's a rare bug that manages to slip past our defenses and make us sick. For plants, though, it's a different story. With no circulatory system, each cell has to fend off viruses, bacteria, and other organisms that would do it harm.

But how? This is a question scientists are only beginning to answer. In plant cells, one line of defense starts when a resistance protein recognizes part of a disease-causing virus or bacterium, a fungus, or even an insect. **Peter Moffett** and his lab study a resistance protein called Rx. When one end of Rx recognizes a specific molecule from Potato Virus X (PVX), it triggers a change at the other end of the protein, which signals the plant to defend against the invader.

Sometimes the cell's reaction is to commit suicide to cut the spread of the virus, but the plant has other tools in its arsenal. Moffett's lab is searching for other defense responses that resistance proteins activate. Intriguingly, when Rx recognizes PVX, it spurs the plant cell to kill off other viruses that it might not otherwise have targeted. Moffett hypothesizes that every resistance protein may activate multiple resistance pathways, including one that appears to attack viral RNA.

The lab also looks for proteins with which Rx interacts, since these are likely to be part of the chain of signals the resistance protein sets off. In 2005, the scientists found one such protein and are now working to learn more about what it does and how it interacts with Rx.

HOW TO MAKE A RIBOSOME

The ribosome is a strange beast. Often likened to a factory, the ribosome can also be seen as a translator, using the four-letter code of RNA to build proteins with 20 kinds of amino acids. To accomplish this, the ribosome needs its many components—that is, dozens of proteins and a few specialized RNA molecules—to work together like a well-oiled machine.

The situation is even more complicated for mitochondria and chloroplasts, cellular compartments with their own genes and ribosomes. Of the 54 proteins in a chloroplast's ribosome, 33 come from genes in the nucleus of the cell, while the chloroplast's own genes code for the rest. **David Stern** and his lab study how the chloroplasts and nuclei of plant cells accomplish ribosome construction and other feats of cooperation.

In one experiment, research associate Tom Bollenbach investigated the origin of the four RNA molecules incorporated into chloroplast ribosomes. He knew that all four came from genes adjacent to each other on the chloroplast chromosome and were copied into RNA in one long stretch. He wanted to learn how that stretch of RNA gets cut into useful form.

Bollenbach and other lab members started by identifying three nuclear plant genes that look similar to a bacterial gene known to be involved in RNA processing. Using microscopy, they found that only one of the three acted in the chloroplast. Next, the lab knocked out this gene, RNR1, and got white plants that couldn't photosynthesize—their chloroplasts weren't working. Uncut ribosomal RNAs were building up in these mutant chloroplasts, Bollenbach found, and their ribosomes weren't coming together. He deduced that the RNR1 gene must code for an enzyme that processes the ribosomal RNA.

Cells somehow sense the buildup of unprocessed RNAs in the absence of RNR1 and scale back production of the chloroplast proteins needed to make the ribosome. The lab is working to determine how plants manage this regulation.

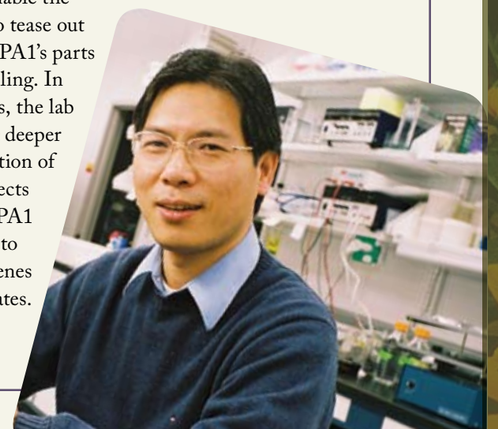
ILLUMINATING DECISIONS

Survival for plants hangs on their ability to constantly read and adjust to their environments, to carry out a complex calculus to determine how best to develop and thrive. **Haiyang Wang** studies how light of different colors and strengths changes the expression of certain plant genes, affecting whether a seed germinates, how tall and in which direction the plant grows, when it flowers, and other responses. He and his lab team also study how the pathways that link light detection to gene expression interact with signals for other environmental cues such as temperature, moisture, and presence of pathogens.

In one 2005 project, the Wang lab investigated HFR1, a protein that prompts light responses by switching certain genes on or off in the presence of far-red or blue light. They found that HFR1 interacts with two proteins that repress light responses, COP1 and SPA1. They also showed that COP1 and SPA1 form a protein complex that tags HFR1 with ubiquitin, which labels proteins for demolition.

Wang hypothesized that the COP1-SPA1 complex might break down HFR1 under dark conditions. To test this, lab members knocked out the gene for COP1 in some *Arabidopsis* plants and SPA1 in others. While unaltered plants had very little HFR1 in the dark, those missing either COP1 or SPA1 showed a buildup of HFR1 in their nuclei. These studies suggest that COP1 and SPA1 break down HFR1 to prevent light responses, and that light represses them, allowing HFR1 to accumulate and activate the plant's light response.

To learn more about SPA1 and its role in this complex, the lab is studying plants with mutations in various sections of this protein. Comparing these to unaltered plants will enable the researchers to tease out the roles of SPA1's parts in light signaling. In future studies, the lab aims to delve deeper into the question of how light affects the COP1-SPA1 complex and to find which genes HFR1 regulates.



GIFTS AND GRANTS 2005

Chairman's Circle ($5,000+)
Helen I. Graham Charitable Foundation
Triad Foundation, Inc.
WMB Thompson Fund

President's Circle ($2,000–$4,999)
Anonymous
Philip Goelet
Elizabeth McNew
Carolyn W. Sampson
Anne and Constantine Sidamon-Eristoff

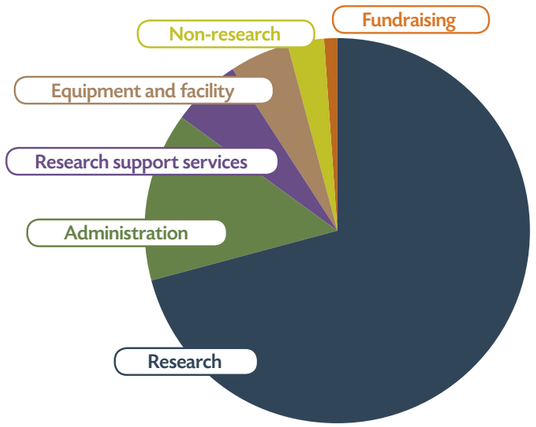
Senior Scientist's Circle ($1,000–$1,999)
John M. Dentes
Francille M. and John D. Firebaugh
George and Helen Kohut
Elizabeth and Roy H. Park, Jr.
David B. and Karen A. Stern
Leonard and Sylvia Weinstein

Scientist's Circle ($500–$999)
Evelyn Berezin
Johanna and Robert Granados
Jacqueline M. and Ralph W.F. Hardy
Gregory and Susan Martin
Larry and Nancy Russell

Associate Scientist's Circle ($100–$499)
Charles and Kathy Arntzen
John B. Babcock
Harry P. Burchfield and Eleanor Storrs-Burchfield
Lisa Christian
Greta M. and Luke J. Colavito
Peter Cousins
James Giovannoni and Julia Vrebalov
Thomas Brutnell and Mary Howard
Elizabeth and Stephen Howell
Mary F. Jaffe
Ernest and Pauline Jaworski
Anthony M. Marzullo, Sr.
Donna Meyer
Robert and Virginia Miller
Al and Lucy Pola
Nancy Ray
Anne D. and J. Alan Renwick
Ruth and Samuel Ristich
Donald and Marcia Slocum
Mildred and Richard Staples
Marguerite and Norman Uphoff
F. Ben Williams
Alan and Judith Wood

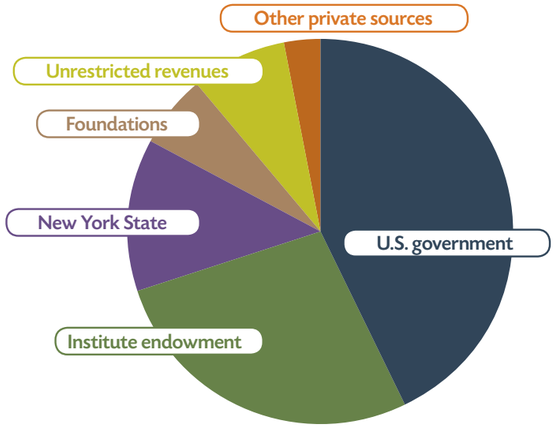
Donor Circle
David G. and Mary Quick Flinn

FINANCIAL REPORT 2005



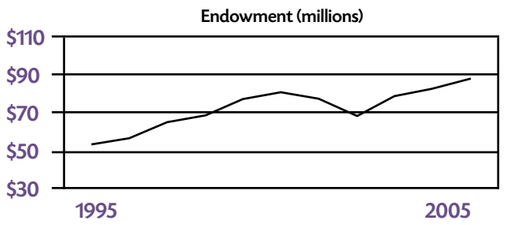
Uses of Funds

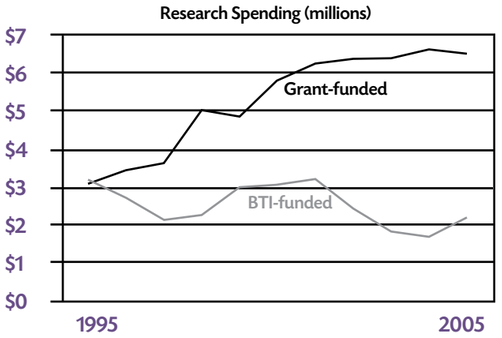
Research	$8,672,000	71 percent
Administration	1, 763,000	14 percent
Research support services	704,000	6 percent
Equipment and facility	674,000	5 percent
Non-research	323,000	3 percent
Fundraising	117,000	1 percent
TOTAL	$12,253,000	



Sources of Funds

U.S. government	$5,396,000	43 percent
Institute endowment	3,379,000	27 percent
New York State	1,749,000	13 percent
Foundations	736,000	6 percent
Unrestricted revenues	962,000	8 percent
Other private sources	358,000	3 percent
TOTAL	$12,580,000	





Writing: Shawna Williams
Photography: Gary Blissard, Steve Davidson, Nicole Markelz, Lucy Pola, Sheryl Sinkow Photography, Susan Strom, University Photography/Kevin Stearns, Shawna Williams, Paul Wolterbeek

Printed on recycled paper.

Produced by the Office of Publications and Marketing at Cornell University.

3/06 2700 1SP 060268

BOARD OF DIRECTORS

Evelyn Berezin
New York, NY

Alan J. Biloski
Visiting Lecturer of Finance, Cornell University

Peter Bruns
Vice President, Grants and Special Programs, Howard Hughes Medical Institute, Chevy Chase, MD

Vicki L. Chandler
Carl E. and Patricia Weiler Endowed Chair for Excellence in Agriculture and Life Science, Department of Plant Sciences, University of Arizona, Tucson, AZ

Ezra Cornell
Vice President for Investments, Salomon Smith Barney, Ithaca, NY

William E. Crepet
Chair, Department of Plant Biology, Cornell University

Gregory Galvin
President and CEO, Kionix, Inc., Ithaca, NY

Philip Goelet
Red Abbey LLC, Baltimore, MD

Maureen R. Hanson
Liberty Hyde Bailey Professor, Department of Molecular Biology and Genetics, Cornell University

Ralph W. F. Hardy
President Emeritus, Boyce Thompson Institute for Plant Research, West Chester, PA

John E. Hopcroft
Professor, Department of Computer Science, Cornell University

Theodore L. Hullar
Director, Higher Education Program, Atlantic Philanthropies, Inc., Ithaca, NY

Karen L. Kindle
Lead, Bioinformatics Genomics Technology, Monsanto Company, St. Louis, MO

Stephen Kresovich
Vice Provost for Life Sciences, Cornell University

Roy H. Park, Jr.
President and CEO, Park Outdoor Advertising of New York, Inc., Ithaca, NY

Ralph S. Quatrano
Spencer T. Olin Professor and Chairman, Department of Biology, Washington University, St. Louis, MO

Carolyn W. Sampson
Ithaca, NY

Constantine Sidamon-Eristoff
Of Counsel, Lacher & Lovell-Taylor, PC, New York, NY

David B. Stern
President, Boyce Thompson Institute for Plant Research

Steven D. Tanksley
Liberty Hyde Bailey Professor, Plant Breeding and Genetics, Cornell University

Crispin Taylor
Executive Director, American Society of Plant Biologists, Rockville, MD

Emeritus Directors

Paul H. Hatfield, St. Louis, MO
Paul F. Hoffman, Chicago, IL
Christian C. Hohenlohe, Washington, DC
Robert M. Pennoyer, New York, NY
Leonard H. Weinstein, Ithaca, NY
Roy A. Young, Corvallis, OR

Officers

Ezra Cornell, chair
Roy H. Park, Jr., vice chair
David B. Stern, president
John M. Dentes, vice president for finance and operations, treasurer
Gary W. Blissard, vice president for research
Donna L. Meyer, secretary
Lucy B. Pola, assistant secretary/treasurer

82ND BTI ANNUAL REPORT

Boyce Thompson Institute for Plant Research
Tower Road
Ithaca, NY 14853-1801
607-254-1234
http://bti.cornell.edu