



Boyce Thompson Institute
for Plant Research

2008 Annual Report



LETTER FROM THE PRESIDENT

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In my 2008 travels, I heard excitement around the world regarding the roles plants are likely to play in our future energy economy. The Obama administration seems likely to invest in this area, and BTI is well-positioned to capitalize and contribute. Indeed, several of the 18 new extramural grant awards to Institute faculty are directly relevant to bioenergy, underscoring BTI's commitment to being part of the solution to this complex global dilemma.

Our Board of Directors supports this commitment, and BTI staff began a strategic planning process in the fall that will guide us in the years ahead. To enrich our research agenda and capabilities, five new faculty were hired in 2008, swelling our ranks to 19, a level last seen in 2000. Inside this report, and on our updated Web site, you can explore their assorted approaches to understanding organismal function, including molecular biology, genetics, chemistry, cell biology and bioinformatics.

As these new scientists staffed their laboratories, BTI experienced a flood of new postdoctoral fellows and technicians, and overall the Institute's employee count rose nearly 36 percent, to 170 at the end of 2008. Remarkably, every one of our new scientists obtained one or more external research grants, and all of our other faculty maintained or increased their funding. Given the very tight federal budget, their success speaks strongly to the relevance of our mission and the quality of our staff. Their productivity and diversity is reflected in 52 research publications covering no fewer than 18 organisms as well as broadly applicable databases, tools and reviews.

As part of BTI's broader mission, our educational activities continued apace in 2008. We welcomed a new Education/Outreach Coordinator whose efforts are summarized in this report. BTI endeavors to increase public understanding of science, and to educate future scientists, through the media (Web and radio), by visiting schools, and by bringing students and teachers into the Institute. These programs would not be possible without the enthusiastic participation of BTI staff, and are augmented by gifts from individual supporters. The Helen Graham Foundation has been an ongoing supporter, and in 2008 it made a significant gift to equip our teaching laboratory.

The world faces serious economic, environmental and societal challenges in 2009 and beyond. BTI is planning and budgeting prudently so that our mission can continue, and so that our outstanding employees can remain part of it. Your support and engagement are deeply appreciated.

David Stern, Ph.D.
President, BTI



Outreach Programs Engage Students and Adults

BTI continued its wide variety of education and outreach programs in 2008. From the daily public radio segment BTI sponsors, called "MicrobeWorld," to student, teacher and adult education, the Institute works to promote national, state and local interest in science.

2008 highlights for adults included the monthly "Science Cabaret," a series of talks sponsored by BTI at an Ithaca café, and a science curriculum development workshop for high school teachers, which was supported by the National Science Foundation and designed and held at BTI.

Elementary school students enjoyed "Science After School," a program that brought the wonders of science to Ithaca-area schools through creative, hands-on lessons. Middle school girls and their parents attended a plant biology workshop at BTI called "Expanding Your Horizons," which aimed to encourage girls to pursue careers in science and math.

Once again, BTI took a leadership role in 2008 and offered the Research Experience for Undergraduates (REU) internship program in partnership with Cornell. This year, 21 undergraduates from universities throughout the U.S. and three Ithaca high school students interned at the Institute during the summer.

Lectures, Seminars Highlight Post-Graduate Society Events

The Post-Graduate Society (PGS) offered its members a number of opportunities in 2008 to develop their careers and interact with leading scientists. The 2008 PGS Professional Development Series included a seminar on how to successfully apply and interview for faculty positions, and a workshop on "The Art of Grant Writing," which attracted the majority of BTI postdoctoral fellows.

PGS-sponsored lecturers during the year included Cornell faculty members Tony Bretscher, Ph.D.; Joss Rose, Ph.D., and Karl Niklas, Ph.D. Social events and other programs rounded out the year's activities.

BTI Alumni Society: Connecting with Old Friends

The BTI Alumni Society was formed in 2007 to increase communication between the Institute and its former faculty and staff. In 2008, the Society conducted a survey of all past employees to determine the kind of activities they prefer. According to the results, alumni are interested in having a Web-based network through which they can connect as well as occasional social events and special lectures. BTI is currently considering ways to fulfill their requests.

International Staff Study "American" English

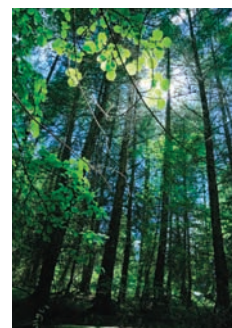
BTI postdoctoral fellows and staff are a highly international group. In fact, Japanese, French, Hindi, Thai, Spanish, Mandarin Chinese and Vietnamese are only some of the native languages represented at the Institute.

Though most of our international staff are fluent in English, many of them want to improve their pronunciation or learn American idioms and slang. For that reason, BTI began offering an English conversation class in 2008, which attracted 28 "students" during the summer and fall sessions. The classes included games and activities to make learning fun. Students also enjoyed sampling typical American food each week. Class evaluations were positive, which indicates our "students" feel their time was well-spent.

BTI Progress on Sustainability Continues

In our effort to save on energy costs and reduce our carbon "footprint," BTI continued exploring the development of a long-term sustainability plan in 2008. We made environmental inroads during the year by installing more than 1,180 yards of reflective shade cloth in BTI's 20 greenhouses. The greenhouses consume a large portion of the Institute's energy budget, so they were obvious targets for cost savings.

By reflecting sun in the summer and retaining heat in the winter, the shade cloth is expected to reduce BTI's greenhouse energy consumption by about 35 percent. Like awnings, the shades can be opened or closed depending on weather conditions — a feature which also adds to the comfort of the staff who work in the facilities.



ARRIVALS AND DEPARTURES

Five Scientists Join BTI Faculty

Klaus Apel received his Ph.D. in biology from the Max Planck Institute of Molecular Cell Biology and Genetics in Dresden, Germany. He joined BTI in May 2008 as Scientist after serving as a professor at the Swiss Federal Institute of Technology in Zurich.

His primary research interest is to better understand, at the genetic level, how plants sense and respond to stress in their environment, such as heat, drought or cold. His work could lead to plants that cope better with stress and produce higher yields. (See research summary on page 5)

Carmen Catala joined the BTI faculty in March 2008 as an Adjunct Scientist. She received her Ph.D. in proteomics from the University of Valencia in Spain and then completed a postdoctoral fellowship at the R.W. Holley Center for Agriculture & Health at Cornell.

Her research is focused on understanding the role of certain plant hormones, called auxins, in the development and ripening of fleshy fruits, such as tomatoes. Dr. Catala's discoveries could lead to fruits with improved flavor, texture or other unique qualities. (see research summary on page 6)

Lukas Mueller completed his Ph.D. at the University of Lausanne in Switzerland, and then accepted a postdoctoral fellowship at Stanford. In 2000, he joined Cornell as a senior research associate in the Department of Plant Breeding and Genetics. He joined BTI as Assistant Scientist in May 2008.

Dr. Mueller's primary area of research interest is bioinformatics. He specializes in the development of software and databases for the organization, storage and retrieval of the vast genetic data about plants that is being discovered by scientists at BTI and elsewhere. (see research summary on page 11)

Eric Richards received his Ph.D. in epigenetics from Harvard University. In 1989, he became a Fellow at Cold Spring Harbor Laboratory where he was appointed a staff member in 1991. He accepted a professorship at Washington University in St. Louis in 1992, and arrived at BTI as an Assistant Scientist in September 2008.

Epigenetics — the study of non-genetic factors that can change the information contained in DNA without changing the DNA itself — is Dr. Richards' area of interest. He is working to discover how many epigenetic changes exist in the natural plant population, whether the environment causes these changes, what observable characteristics result from these changes, and how many of these epigenetic changes are inherited. (see research summary on page 12)

Sorina Popescu received her Ph.D. in molecular biology from Rutgers University, and then was a research associate at Yale. She joined BTI as an Assistant Scientist in September 2008.

Dr. Popescu is interested in isolating, identifying and then determining the function of the various proteins that control plant cell processes. As part of that work, she has refined a technique, called a microarray, which scientists use to study protein function. Her enhanced technique will enable her and others to efficiently study protein function on a scale not possible before. (see research summary on page 11)

BTI Human Resources Director Joins Cornell

After nearly ten years at Boyce Thompson, **Lucy Pola**, Director of Human Resources, accepted a Human Resources position at Cornell University's College of Human Ecology in 2008. Lucy kept a sharp eye on employee benefits, oriented hundreds of new employees, ensured that the staff was informed of the latest employment laws, policies and procedures, and brought employee dental insurance to the Institute.



Klaus Apel



Carmen Catala



Lukas Mueller



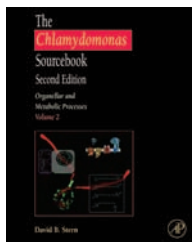
Eric Richards



Sorina Popescu

Chlamydomonas Sourcebook Revised

The fast pace of *Chlamydomonas* research has led a group of three scientists, including **President David Stern**, to revise "The *Chlamydomonas* Sourcebook." The first edition



was published in 1988, and the new 2009 version (published in 2008) has expanded into three volumes. Stern's Volume 2 is the longest and contains 29 chapters, which serve both

as a reference manual and a biological view of the "green side" of *Chlamydomonas*, along with a number of processes that possess biosynthetic and/or regulatory links to photosynthesis. This set of books, published by Elsevier, may well become the bedrock for *Chlamydomonas* research for years to come.

Three BTI Faculty Members Featured in the Media

Eric Richards, an epigenetics expert, was quoted in a New York Times feature article on new discoveries about genes and how they work, entitled "Now: The Rest of the Genome."

"Can You Rescue a Rainforest?" — an article published by Ecological Restoration magazine — featured Emeritus Scientist **Carl Leopold** and his work to restore a tropical rainforest ecosystem in Costa Rica.

The Cornell Chronicle Online focused on **Frank Schroeder's** research with *C. elegans* nematodes (worms) in an article entitled "Come-hither chemicals also slow aging — in worms."

BTA: Colonel's Home Back in the Fold

A 32-acre parcel of land in Arizona which includes Colonel Boyce Thompson's summer home — Picket Post House — is once again part of the Boyce Thompson Arboretum (BTA). Sold in 1946, the house and property were purchased in 2008 by the Arizona State Parks Department. This important acquisition adds the Colonel's presence to BTA once again.

Evelyn Berezin

New York, NY

Alan J. Biloski

Visiting Lecturer of Finance,
Cornell University, Ithaca, NY

Peter Bruns

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

Thomas Burr

Director, New York State
Agricultural Experiment Station at
Geneva, Geneva, NY

Vicki L. Chandler

Director, BIO5 Institute,
University of Arizona, Tucson, AZ

Mary E. Clutter

Washington, DC

Ezra Cornell

President, Cornell Pochily
Investment Advisors, Inc., Ithaca, NY

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Cornell University, Ithaca, NY

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and Genetics, Cornell University,
Ithaca, NY

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President & CEO, Tompkins County
Trust Company, Ithaca, NY

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Technology, Monsanto Company,
St. Louis, MO

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Vice Provost for Life Sciences,
Cornell University, Ithaca, NY

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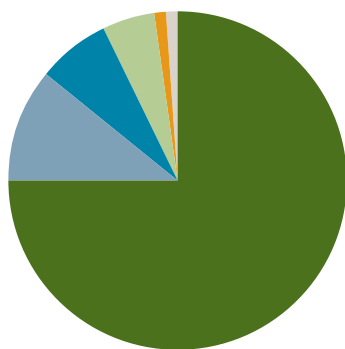
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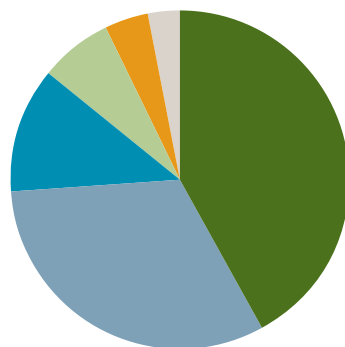
Leonard H. Weinstein,
Ithaca, NY

Roy A. Young, Corvallis, OR



USE OF FUNDS

| | | |
|----------------------|---------------------|-------------|
| Research | \$12,883,000 | 75% |
| Administration | 1,836,000 | 11% |
| Research Support | 1,232,000 | 7% |
| Equipment & Facility | 787,000 | 5% |
| Non-research | 156,000 | 1% |
| Fund-raising | 156,000 | 1% |
| Total | \$17,050,000 | 100% |



SOURCE OF FUNDS

| | | |
|-----------------------|---------------------|-------------|
| U.S. Government | \$6,958,000 | 42% |
| Institute Endowment | 5,243,000 | 32% |
| New York State | 1,954,000 | 12% |
| Unrestricted Revenue | 1,240,000 | 7% |
| Other Private Sources | 732,000 | 4% |
| Foundations | 568,000 | 3% |
| Total | \$16,695,000 | 100% |

GIFTS

Chairman's Circle

(\$5000+)

Helen I. Graham Charitable Foundation
Philip and Anette Goelet
Christian Hohenlohe
Wm. B. Thompson Fund

President's Circle

(\$2000 - \$4999)

Carolyn Sampson
David and Karen Stern

Senior Scientist's Circle

(\$1000 - \$1999)

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John M. and Marie Dentes
Dr. Ralph W.F. and Jacqueline M. Hardy
Ji-Young Lee and Sang-Hun Oh
Roy H. and Elizabeth P. Park, Jr.

Associate Scientist's Circle

(\$100 - \$999)

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Gary Blissard and Elizabeth Mahon
Tom Brutnell and Mary Howard
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Eleanor Storrs Burchfield
James J. and Terry R. Byrnes
Bruce Cahoon

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Charles and Jane Walcott
Frederic A. Williams
Jianjun Yang
Roy A. Young

Donor

(\$1 - \$99)

Carl Leopold
David and Mary Sirois
Mary Westlake

GRANTS

| | |
|---------------------------------|--------------------|
| National Science Foundation: | \$3,782,000 |
| U.S. Department of Energy: | 1,266,000 |
| U.S. Department of Agriculture: | 399,000 |
| BARD: | 261,000 |
| Monsanto Company: | 204,000 |
| Nat'l Institutes of Health: | 200,000 |
| DARPA: | 125,000 |
| BSF: | 51,000 |
| Total: | \$6,288,000 |

How do plants respond to environmental stress?

Klaus Apel, Ph.D.

Scientist, BTI



Plants can endure extreme environmental stress (heat, drought, cold or intense light) through genetically controlled defenses, such as wilting, loss of leaves or stunted growth, but these very defenses can also reduce yields, among other effects. As a result, one effect of global warming could be reduced food production just when the world's population is burgeoning.

Understanding how plants sense and respond to stress at the genetic level is the ultimate objective of **Klaus Apel's** laboratory at BTI. His findings could enable scientists to mitigate the negative results of stress, such as yield loss, or fine tune a plant's ability to survive climate change.

It turns out that chloroplasts — the tiny organs that contain chlorophyll and carry out photosynthesis — play an important role in a plant's ability to sense environmental stress. Conditions such as drought, heat, cold and intense light interfere with the normal photosynthetic process in the chloroplasts, which leads to over-production of sometimes toxic forms of oxygen, called reactive oxygen species (ROS).

High levels of ROS were previously considered detrimental to the cell. However, recent work with an *Arabidopsis* mutant by Apel and his research group indicates that the release of one ROS, called singlet oxygen, in the chloroplast actually triggers a variety of positive stress adaptation responses in the plant. These responses include slowed plant growth, cell death, and the activation of a broad range of defense genes, which normally are turned on only in the presence of pathogens.

In further work, Apel's group proved that certain genetic mutations in *Arabidopsis* eliminate the plant's stress responses without interfering with the release of singlet oxygen. It appears these mutations prevent the plant from sensing the presence of singlet oxygen, which, in turn, prevents symptoms of stress. Apel's group has identified these mutated genes, which is a first and crucial step toward understanding the genetic basis of the stress response in plants.

The results of Apel's work could lead to plants that cope better with the environmental stress of global warming. Ultimately, such a discovery could help increase food supplies or predict a plant's susceptibility to environmental changes.

How do viruses infect insect pests?

Gary Blissard, Ph.D.

Vice President for Research and Scientist, BTI

Adjunct Professor, Dept. of Microbiology and Immunology and Dept. of Entomology, Cornell University



Certain viruses are our allies in the fight against insect pests. Research that leads to a better understanding of these viruses could in turn lead to more environmentally friendly, natural insect control, and even to advances in human health.

Among other research projects, **Gary Blissard** is studying how certain viruses, called baculoviruses, infect insects. He and his colleagues have focused on how a particular baculovirus envelope protein, called GP64, enables the virus to invade an insect cell, multiply and then exit in massive numbers.

Blissard's group has found that GP64 has three major functions in the viral infection cycle. First, they showed that GP64 is an attachment protein — a protein that enables the virus to bind to receptors on the surface of the host insect cell, which is the first step in the process of infection. Blissard's laboratory identified the particular portion of GP64 that is necessary for this binding activity.

After the virus binds to the host cell, it enters the cell where it is surrounded by the cell's membrane. To cause infection, the virus must fuse with that membrane and deliver its DNA into the cell nucleus. Having proved that GP64 is independently able to fuse membranes, Blissard's team is now involved in a detailed investigation of how this process occurs.

The third step in the infection cycle calls for new virus particles to emerge or "bud" from the cell surface. To determine whether GP64 played a role in virus budding, Blissard's lab "knocked out" the gene for the GP64 protein, which severely limited virus budding, and the remaining new virus particles were not infectious. These studies show that GP64 plays a critical role in the assembly of the new virus particles. Current studies aim to understand these three major functions of GP64 in much greater detail.

Knowing how baculoviruses infect insect cells may enable scientists to improve the virus' insect control capabilities, which could reduce the use of chemical pesticides. But this work also has other exciting applications, such as in gene therapy. Because baculoviruses cause disease only in insects and because they are highly effective at entering cells and depositing DNA in the cell nucleus, they may be excellent vehicles for inserting beneficial new genes into mammalian cells — an advance that could improve our ability to safely correct genetic disorders in humans.

How does maize produce beta-carotene?

Tom Brutnell, Ph.D.

Associate Scientist, BTI

*Adjunct Associate Professor,
Dept. of Plant Biology and
Dept. of Plant Breeding and Genetics,
Cornell University*



Two BTI laboratories — Brutnell and Van Eck — are studying the genetic basis of beta-carotene production in certain staple foods (see Van Eck, page 13). Beta-carotene is a carotenoid and is the precursor to Vitamin A, which can prevent the eye disease and health disorders that currently plague hundreds of millions of children in the developing world. In their studies with maize, which is low in beta-carotene, the Brutnell laboratory is working to enhance beta-carotene production in that crop — research that could lead to more nutritious varieties of corn, and healthier diets for some of the world's poorest people.

Brutnell's team previously proved that the enzyme lycopene beta-cyclase is required for the first step in beta-carotene production in maize. However, his laboratory recently showed that co-expression of the lycopene beta-cyclase enzyme with another enzyme, called lycopene epsilon-cyclase, leads to the accumulation of lutein rather than beta-carotene in seed tissues. This is because epsilon-cyclase is often expressed at high levels in seed tissues, where it competes with the beta-cyclase for the lycopene.

Next, Brutnell discovered rare alleles, or alternative forms of the epsilon-cyclase gene, that are associated with high levels of beta-carotene production. Brutnell's laboratory then used a polymerase chain reaction assay to monitor levels of the epsilon-cyclase enzyme transcripts during maize seed development. These studies confirmed that the gene's expression levels are consistently low in maize lines that produce high amounts of beta-carotene, and high in lines that produce high levels of lutein. Therefore, the ability to single out rare maize alleles with low epsilon-cyclase production could lead to the development of lines that produce more beta-carotene.

To that end, the team developed a polymerase chain reaction tool kit — currently being tested in Mozambique — that enables African corn breeders to identify these rare alleles in North American germplasm and then cross these plants to lines that have been adapted for optimal growth in Africa. The assay is relatively inexpensive, and will help African breeders generate high beta-carotene producing lines of corn.

How do plant hormones control fruit development?

Carmen Catala, Ph.D.

Adjunct Scientist, BTI

*Senior Research Associate,
Dept. of Plant Biology,
Cornell University*



When home gardeners or horticulturalists grow plants from stem cuttings, they often dip the cut end of the stem in a white powder that encourages the stem to develop roots. The white powder is a hormone, called an auxin, which plays an important role in plant growth and development. Auxins also influence cell division and differentiation, which is why the powder form used by gardeners helps the stem cutting to start growing root cells.

It's also known that auxins, particularly one called indole-3-acetic acid, are central to the development and ripening of fruit, such as strawberries and tomatoes. But very little is known about the molecular basis of auxin production, transport and signalling in fleshy fruit-producing plants. This is the area of research that **Carmen Catala** is pursuing at BTI.

Until now, the majority of research into auxins has been done in the model plant *Arabidopsis*, which is a flowering plant that only produces a dry fruit. Work on auxins has been conducted with strawberries, however, and has proven that the hormone is produced in the tiny seeds that speckle the outside of the berry, and that this auxin helps the fruit grow. It also has been found that when the berry is ready to ripen, the auxin is inactivated. A molecular explanation of how and why this happens in strawberries remains elusive.

Catala is applying knowledge gained about auxins in *Arabidopsis* and strawberries and using new molecular techniques to understand exactly how the auxin indole-3-acetic acid works in tomatoes. She aims to discover how and where this auxin is produced in the plant, how it is transported to the cells that will become fruit, and how it signals the cells in that tissue to grow, develop and ripen. What Catala learns in tomatoes will be applicable to other fleshy fruits as well, including strawberries.

Knowing at the molecular level how auxins help set fruit on plants and how they influence fruit development and ripening could one day lead to higher quality fruits. And, because auxins directly stimulate or inhibit the expression of specific genes, understanding how to control the production or transport of these hormones could lead to fruits with improved flavor, texture or other unique qualities.

How is massive plant genomics data organized and interpreted?

Zhangjun Fei, Ph.D.

Assistant Research Scientist, BTI



Consider that just one tomato plant contains about 35,000 genes that express thousands of different proteins. Then consider how many different plants are currently under study, and it's easy to understand the enormity of the data generated in all of biological research.

Organizing that data and making it accessible for further research is an area called bioinformatics. The interface between biology, statistics and computer science, bioinformatics develops computational tools and resources that organize massive amounts of data into usable sets so that the knowledge contained in them can be retrieved, analyzed and applied in biology research. **Zhangjun Fei's** laboratory at BTI develops both the databases and the interfaces needed to help scientists understand how genes work together and how they form functioning cells and organisms. He also has developed analytical and data-mining tools that allow scientists to efficiently extract biological information from the database for use in their research.

Fei has collaborated with the Giovannoni laboratory at BTI to develop databases that contain information on the expression of more than 10,000 tomato genes, as well as profiles of tomato fruit nutrition and flavor-related metabolites, during different developmental stages, upon various stresses, and in different genetic backgrounds.

As a part of International Cucurbit Genomics Initiative (ICuGI), the Fei lab has also developed a database, along with its corresponding interface and tools, for the organization of all the genomics information gathered to date on the cucurbit family of plants, which includes melon, watermelon, cucumber and pumpkins, among others.

Work in Fei's laboratory is providing tools and resources that organize genomics information about an organism into a form scientists can easily use to analyze and visualize the data they've gathered.

How do fruits ripen?

Jim Giovannoni, Ph.D.

Adjunct Scientist, BTI

*Plant Molecular Biologist,
USDA-ARS Plant, Soil and
Nutrition Laboratory*

*Adjunct Professor, Dept. of Plant
Pathology and Plant-Microbe Biology,
Cornell University*



Learning the genetic basis of fruit ripening could significantly impact the quality and availability of certain foods. This knowledge would be particularly useful in countries where food spoilage due to over-ripening is a cause of hunger.

Jim Giovannoni's laboratory at BTI is working to understand the process by focusing on the genes and regulatory networks that control fruit ripening in tomatoes — knowledge that will have applications in other plants such as pepper, peach, pineapple, banana, strawberry and melon. Because many fruits ripen in response to the release of the hormone ethylene, understanding the mechanism that controls the plant's sensitivity to ethylene can lead to basic knowledge of the ripening process.

In studying mutant tomato plants that produce only unripe tomatoes, Giovannoni's team discovered an alteration in a gene called *Gr*, or *Greenripe*, that causes overproduction of a certain protein in the fruit that decreases the fruit's sensitivity to ethylene. So, though the plants produce ethylene in normal amounts, the fruit does not respond to it and, therefore, fails to ripen. Being able to control the production of this protein, which would make the fruit under- or over-sensitive to ethylene, could lead to the ability to speed or delay the ripening process.

Giovannoni further found that overproduction of the *Gr* protein throughout the plant has no effect on any part of the plant except the fruit. This discovery is important because it indicates there are constituents specific to the fruit involved in its response to ethylene.

The next step in the research is to develop transgenic tomato plants in which the *Gr* gene has been "knocked out," or disabled, so that the plants are no longer able to produce the *Gr* protein. This work will demonstrate whether or not normal *Gr* expression plays a significant role in ripening. Giovannoni predicts that these plants will be highly sensitive to ethylene and will, therefore, ripen early.

The ability to control the ripening process by controlling the ethylene response could lead to fruit, such as strawberries, papaya, and even tomatoes, which have a longer shelf life. And because ripening is directly related to fruit flavor, texture and nutrient content, these discoveries could lead to higher quality food as well.

How do soil fungi supply plants with mineral nutrients?

Maria Harrison, Ph.D.

Scientist, BTI

Adjunct Professor, Dept. of Plant Pathology and Plant-Microbe Biology, Cornell University



In nature, certain plants and fungi have evolved a complex, symbiotic relationship in which the plants provide the fungi with carbon while the fungi provide the plants with phosphate needed for cell function and growth. Understanding this relationship could result in scientists' ability to develop plants that require fewer applications of phosphate fertilizers.

Working with a soil organism called arbuscular mycorrhizal fungi and a model legume, *Medicago truncatula*, **Maria Harrison's** laboratory is unraveling the mechanisms underlying mineral transfer from fungus to plant. The fungi, which are ubiquitous in soil, live in close proximity to the plant's roots. The fungal spores grow on the root surface and, in response to a signal from the plant, grow into the cells of the root. Once there, the plant forms a membrane, called the arbuscular membrane, through which the mineral exchange occurs.

Harrison theorized that a particular transporter protein in the arbuscular membrane mediates the movement of phosphorus from the fungus into the plant cell. In 2007, her team demonstrated that this theory was correct. When the plant gene that produces the transporter protein in question was "knocked out," or disabled, phosphate in the arbuscule did not cross into the plant cell.

Harrison's research yielded another, somewhat surprising result. She discovered that in the mutant plant, the arbuscules die very quickly. One interpretation is that the plant, on detecting that the phosphate transfer is not occurring, responds by triggering the death of the arbuscule. Understanding how and why this occurs will be a next step in her research.

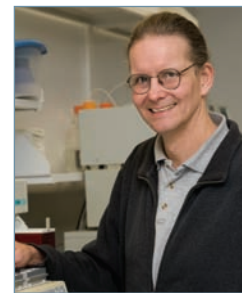
Today, growers use fertilizers derived from rock phosphate to enhance plant nutrition, but rock fertilizer reserves are being depleted and at the current rate of use, they will last only an additional 90 years. Furthermore, excessive application of phosphate fertilizers contributes to the pollution of streams. Harrison's work may lead to plants that can use naturally occurring phosphate in the soil more completely and efficiently through enhanced symbiotic relationships with fungi — an advance that would lead to more environmentally friendly, sustainable agriculture.

How do plants ward off insects and produce amino acids?

Georg Jander, Ph.D.

Associate Scientist, BTI

Adjunct Assistant Professor, Dept. of Plant Biology, Cornell University



As world population continues to increase, demand for more food with greater nutritive value also will increase. Finding new methods to protect crop yields and develop foods that are more nutritionally complete are essential in a world where starvation continues to be a serious issue.

About 15 percent of the world's yield from major food and feed crops is lost each year due to insect damage, so developing more environmentally friendly ways of controlling insects can help increase yields and protect the environment.

Georg Jander's laboratory is working to understand the complex biological interactions between plants and insects in order to find new, sustainable ways to reduce crop losses.

To do that, Jander's lab is studying the biochemistry and molecular biology of plant-insect interactions. Plants are not passive targets for insect herbivores. In fact, plants know when they are under attack and, in response, alter their gene expression to produce toxins that can deter further insect feeding. Understanding the molecular mechanisms that cause the plants to produce these toxins is a major area of Jander's research.

He and his colleagues are studying this phenomenon using phloem-feeding aphids, which cause relatively little overt damage, but can transmit numerous viral diseases to crop plants, which, in turn, reduce yields. Jander's group recently found that when certain components of aphid saliva are infiltrated into the plant's leaves, they induce defense responses that make the plants more aphid resistant. The laboratory has also identified previously unknown molecules that deter aphid feeding on cabbage and related plants. Ongoing research in the lab aims to identify the plant enzymes that catalyze the formation of these aphid-deterrent molecules.

Jander also is interested in the production of amino acids in plants. Unlike animals, plants are able to synthesize all 20 protein amino acids, and half of those are made only in plants and microbes. Because of that, plants are essential in the mammalian diet. Therefore, Jander's group is working to understand the regulation of essential amino acid production in plants, and, so far, they have characterized the roles of certain enzymes involved in the production of two of them: threonine and methionine.

Jander's work in plant-insect interactions and the regulation of amino acid production in plants may have important implications for agriculture. Finding new ways to control insect damage will increase the amount of food available, while his work to understand amino acid production in plants could lead to food with higher nutritional quality.

How do plants acquire immunity to disease?

Dan Klessig, Ph.D.

Scientist, BTI

Adjunct Professor, Dept. of Plant Pathology and Plant-Microbe Biology, Cornell University



Plants have various ways to protect themselves against pathogens, including preformed physical and chemical barriers and induction of multiple defense responses. Perception of an invading pathogen often involves recognition of a pathogen-encoded factor by plant resistance (R) proteins. Over the past 15 years many R proteins have been identified in a variety of plants. They provide protection against diverse pathogens, including viruses, bacteria, fungi, and oomycetes.

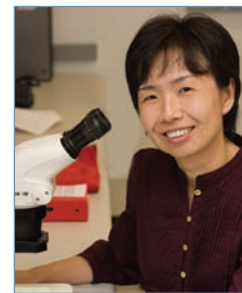
Activation of R proteins by pathogens triggers an array of defense responses including production of the key defense hormone salicylic acid, activation of defense genes and induction of host cell death at the site of infection. R proteins appear to exist in a repressed or inactive form in the absence of pathogens, due to inhibitory folding of the R protein or interaction of it with negative regulator proteins. Many R proteins are in stable complexes, which contain a small number of host proteins that facilitate proper folding of R proteins and are required for their activation.

To help understand how R proteins are activated and then transmit their information to the rest of the cell to induce defense, **Dan Klessig's** group in 2000 identified an R protein called HRT that confers resistance to Turnip Crinkle Virus (TCV) in the model plant *Arabidopsis thaliana*. They recently identified *Arabidopsis* mutants which are unable to effectively resist infection by TCV. One of these mutants, called CRT1 (for comprised recognition of TCV), produces a defective ATPase, an enzyme that uses ATP as an energy source to perform various cellular functions. CRT1 interacts with HRT and a wide variety of other R proteins. In addition to its role in resistance to TCV, it is involved in resistance to bacterial and oomycete pathogens, indicating that CRT1 is an important mediator of defense signaling triggered by distinct classes of R proteins.

How do stem cells divide and differentiate in *Arabidopsis*?

Ji-Young Lee, Ph.D.

Assistant Scientist, BTI



In response to certain genetic cues, stem cells in animals can differentiate into a wide variety of specialized cells. Plants, too, have stem cells that divide and differentiate into specialized cells in response to currently unknown, genetically based developmental cues. Understanding these cues is essential to understanding — and influencing — how plants grow.

Ji-Young Lee is studying the genetic factors that cause plant stem cells in procambium/cambium tissue to divide and generate specialized vascular tissues, called xylem and phloem, using the *Arabidopsis* root as a model. Because procambium/cambium tissue is widely present in all vascular plants, the knowledge she obtains from *Arabidopsis* will be transferable to many plant species.

In *Arabidopsis*, approximately 500 genes in the procambium/cambium tissue are highly expressed and work in regulatory networks that cause stem cells to generate xylem and phloem. Using gene expression data already in hand, Lee is trying to determine the function of each of the genes in the underlying regulatory network. Through microarray technology combined with genetics, her goal is to identify the transcription factors, their developmental functions, and their targets. Lee's laboratory is also studying stem cell activity as it relates to tree growth, using the *Populus* tree as a model system.

The potential applications of Lee's work are far-reaching. Understanding the mechanisms that cause vascular tissue stem cells to divide and differentiate in *Arabidopsis* and *Populus* will shed light on stem cell regulation in other herbaceous plants and woody trees. With that knowledge, scientists may one day be able to influence the growth rate of trees and grasses, which could in turn increase the amount of biomass they produce for use as biofuel, feed, or other purposes. Because the molecular mechanisms underlying stem cell regulation in animals and plants are similar, Lee's research may help advance knowledge about stem cell regulation in humans.

How do bacteria overcome a plant's disease defense system?

Gregory Martin, Ph.D.

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There's an arms race underway in the plant world in which plants and disease-causing bacteria are continually evolving ways to outsmart each other. Plants have developed a defense system that enables them to resist disease, but some pathogens have evolved survival methods that undermine this system. Understanding the details of this race for dominance could lead to crop plants with more effective, natural resistance to disease.

Gregory Martin's laboratory studies a bacterium called *Pseudomonas syringae*, which causes bacterial speck disease of tomatoes. When *P. syringae* invades a tomato plant, it injects a disease-promoting protein called AvrPtoB into the plant cells. However, the plant is ready and waiting with the protein Fen, which was recently discovered by Martin's team. Fen recognizes AvrPtoB and, in doing so, activates the plant's defense system.

Fighting back, *P. syringae* has cleverly engineered AvrPtoB to act as a tomato E3 ligase, a protein that tags other proteins to be destroyed. When AvrPtoB binds the Fen protein, Fen is tagged and the plant's own system takes Fen to the cell's garbage bin before Fen can activate the plant's defenses. This eliminates the plant's ability to resist speck disease and ensures the survival of the bacterium.

In further studies, Martin's laboratory found that the *Fen* gene is present in many wild species of tomatoes, suggesting it is an ancient plant defense strategy. But if the bacterial protein AvrPtoB is so effective at destroying the *Fen* protein, why would the *Fen* gene be so prevalent? Martin answers that there are some strains of *P. syringae* that produce a version of AvrPtoB that cannot destroy Fen, and, therefore, cannot turn off the plant's defense system. Consequently, he reasons that the bacteria have only recently evolved the version of AvrPtoB that can sabotage the plant's defenses.

Martin's work helps to explain how the plant/pathogen arms race works at the molecular level and sheds new light on how disease-resistant plants can suddenly become susceptible again. Understanding the strategies pathogens use to overcome plant defenses against disease may lead to crops that have more effective, longer lasting resistance — an advance that could lead to more productive varieties and less dependence on pesticides.

How do plants protect themselves from disease?

Peter Moffett, Ph.D.

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Though plants do not have an adaptive immune system like animals do, plants have evolved a defense mechanism of their own that protects them from disease. **Peter Moffett** is studying one such system using a disease resistance gene from potato.

All plants have a unique repertoire of several hundred disease resistance genes, each of which produces a resistance protein that protects the plant from specific pathogens. But this system works only if the resistance protein recognizes a protein from the pathogen, called an avirulence protein. Moffett is working to understand how this recognition event occurs and how it elicits a protective response in the plant.

Moffett's research team has discovered that the resistance protein they study actually works in concert with another protein, called RanGAP2, which is present in all plants. He found that RanGAP2 physically interacts with the appropriate resistance protein, and allows it to sense the presence of a particular pathogen. RanGAP2 activates a protein called Rx when a virus attacks a potato cell, but if the cell is attacked by a nematode, RanGAP2 activates a protein called Gpa2. Working together, RanGAP2 and the resistance protein initiate a programmed response that will kill the cell, and with it the pathogen.

Most recently, Moffett's laboratory discovered how the Rx protein interacts with RanGAP2 to signal the cell that a pathogen has invaded. This discovery is important because any plant can defend itself against most pathogens, but only if it can recognize the pathogen. Understanding how RanGAP2 works in concert with resistance genes like Rx and Gpa2 to recognize a specific pathogen, may enable scientists to adjust a plant's defense system to mobilize against pathogens it couldn't previously recognize. This, in turn, may lead to a new way to transfer naturally occurring resistance to a particular pathogen from one plant into another — an advance that could have important agricultural implications.

How will researchers store and retrieve scientific information in the future?

Lukas Mueller, Ph.D.

Assistant Scientist, BTI



Consider that thousands of scientists are working worldwide to identify all the genes in a wide variety of plants. Then, consider that a plant like tomato has about 35,000 genes, some of which function in complex networks. And then consider that new genetic discoveries are being made each and every day about how these genes are regulated and how they interact. How can all this information be efficiently stored, updated and made accessible to scientists in a timely manner so they can use it and build on it?

This is the question that **Lukas Mueller's** laboratory at BTI is answering. Among other projects, Mueller's group coordinates the Solanaceae Genomics Network — a database of all the genetic information known about solanaceous plants, such as tomatoes and peppers. He's also involved in the tomato genome sequencing project, which is the work of scientists in 10 countries.

Mueller is developing software that will make it easier for scientists to access vast amounts of genomic data. He's also working to make it simpler for scientists to annotate, or update, the data as they make new discoveries. It's similar to the idea that gave rise to Wikipedia — an encyclopedia that can be added to or revised by anyone who reads it. The difference is that Mueller's software and the database it runs are specifically designed for complex biological data.

Until the advent of the Internet, scientists could only share the results of their research with others through personal communications and by publishing their work in scientific journals. But neither communication method enabled colleagues to access all the data that backed up the results. With the software Mueller is developing, scientists not only can see the results of others' work, they can also see — and use — the data the results were derived from.

Mueller's goal is to make these databases so easy to use that they will become the primary place for storing, sharing, updating and accessing genomics information. At that point, the practical applications of his work would multiply. For instance, plant breeders could use the database to more quickly develop new varieties of crop plants with innovative genetic characteristics, such as higher yield or enhanced nutritional quality.

Which proteins control which biological processes in plants?

Sorina Popescu, Ph.D.

Assistant Scientist, BTI



Proteins are the workers that participate in every process within cells, such as biochemical reactions (metabolism), structure (building and maintaining the shape of the cell) and defense mechanisms (disease or insect resistance), among others. Identifying all the proteins in a plant cell and determining the function of each is a major area of plant biology research.

Until recently, identifying and isolating these proteins and then using them to discover their specific function has been an arduous task. But as a result of discoveries made by **Sorina Popescu**, the work has become easier and more efficient. Popescu enhanced a technique called the microarray, which was previously used only to study DNA, and adapted it to be used for the study of proteins.

Working with *Arabidopsis* proteins, Popescu discovered that she could "print" as many as 5,000 minute protein samples on one microscope slide (the microarray) and that these tiny amounts of protein could then be used in other research that would reveal the function of each. There are about 30,000 proteins in *Arabidopsis*, which means that the entire proteome (the complete protein library) of this plant could be stored on six standard microscope slides.

Using her technique, Popescu and other scientists will be able to more efficiently study protein function on a very large scale. Prior to the development of protein microarray technology, scientists were able to select only a few proteins at a time for study, which restricted their ability to identify which proteins are involved in which processes. Now, Popescu and others can easily study thousands of proteins — even those no one thought were involved in a particular process. As a result, her work has significantly expanded the universe of study involving proteins and their functions.

Popescu plans to use her protein microarray technology at BTI to identify proteins involved in protecting plants from various diseases. Her work could lead to the identification of proteins that were never before thought to be involved in disease resistance, which, in turn, could lead to a new understanding of how plant defense mechanisms work. It could also lead to new ways to protect plants from disease and enhance their yields.

How are genes turned off?

Eric J. Richards, Ph.D.

Scientist, BTI



Everyone knows by now that excessive exposure to the sun's radiation can cause skin cells to become cancerous. Cancer occurs because radiation causes changes (mutations) in the cell's DNA sequence. But cancer and other diseases can also occur when certain genes that might have protected the cell are "silenced" or turned off. In this case, the protective genes become unreadable by the cell and disease can result. Understanding how these genes become "unreadable" is the goal of a relatively new area of genetics research called epigenetics.

Eric Richards' laboratory studies epigenetics in the model plant *Arabidopsis*. "Epi-" means "on top of" or "in addition to," so epigenetic traits exist on top of or in addition to the cell's DNA sequence. Epigenetics research seeks to understand the molecular mechanisms that change the information contained in the DNA (making it unreadable) without changing the underlying DNA sequence. Richards focuses on one of these mechanisms, called "DNA methylation." In this process, a small chemical group is added to one of the DNA bases (cytosine) which can make the gene unreadable by the cell. The methylated gene can die out with the cell or it can be passed on to new generations of cells and, in some cases, organisms. Consequently, DNA methylation may play an important role in evolution, just as mutations do.

Richards is working to understand how, where and when DNA methylation occurs, its consequences on the organism, and to what extent variation in methylation is passed on to future generations. He is studying this process in plants because they can survive major epigenetic alternations that other organisms, like mice, cannot.

Understanding the basic biology of DNA methylation in plants could have applications to human health, such as the detection and prevention of disease. But it also has important applications in agriculture. Epigenetics controls important traits in crop plants, such as disease resistance and flowering time, so advances in this field of genetics could lead to higher quality food or increased yields.

How do secondary metabolites affect human health?

Frank Schroeder, Ph.D.

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The relevance of nucleic acids, proteins and carbohydrates for all aspects of biology is well established, but the varied and often unexpected roles of so-called "secondary" metabolites are just now emerging. Secondary metabolites regulate development and immune response in plants and animals (such as hormones) and also play an important role in the interactions of different organisms with each other.

Identifying secondary metabolites and determining their function is an important area of biomedical research that can help scientists better understand diseases such as bacterial infections, diabetes and cancer, as well as the phenomenon of aging.

Secondary metabolites are very different from proteins and nucleic acids. They constitute a chemically extremely diverse class of compounds, which have so far resisted systematic analysis.

Frank Schroeder's laboratory is developing new analytical methodology based on a technique called NMR spectroscopy, which promises to greatly simplify scientists' ability to identify the chemical structure of these compounds and find their biological functions. Using this approach, Schroeder's team is investigating the role of secondary metabolites in specific aspects of plant and animal biology.

In one area of research, Schroeder is investigating secondary metabolites produced by the nematode *Caenorhabditis elegans*. Nematodes are roundworms that are about 1 mm in length and ubiquitous in the soil. Scientists believe that many of the physiological pathways in *C. elegans* are analogous to corresponding pathways in higher animals, and as a result, nematodes have become an important model system for human disease and aging. Although the entire *C. elegans* genome — about 20,000 genes — was sequenced more than 10 years ago, little is known about its secondary "metabolome."

Recently, Schroeder's group identified several new compounds that influence mating behavior, as well as pathways involved in nematode development and life span regulation. The ultimate goal of this research is to identify the entire *C. elegans* secondary metabolome for chemical structure and biological function.

In other research, Schroeder and colleagues recently used his NMR-spectroscopic approach to identify a previously unknown human hormone that controls the excretion of sodium via the kidneys. This discovery may one day lead to a new approach for treating high blood pressure.

How do plants co-regulate genes?

David Stern, Ph.D.

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About two billion years ago, a photosynthetic bacterium was engulfed by another single-celled organism, forming the first photosynthetic eukaryote. Eukaryotes differ from bacteria because they have multiple genetic compartments within the cell. In the case of plants there are three genetically-active compartments: the nucleus, the mitochondria, and the chloroplasts. While the nucleus contains most of the plant cell's genes, the organellar (mitochondrial and chloroplast) genomes are essential, encoding proteins involved in energy metabolism. **David Stern's** laboratory studies how plant organellar genes are regulated, thus coordinating their expression with nuclear genes.

The Stern laboratory uses a single-celled alga, *Chlamydomonas reinhardtii*, as one experimental model. The *Chlamydomonas* chloroplast contains about 100 genes, which are regulated by the nuclear factors. One group of nuclear genes under study are the MCD loci, which are responsible for stabilizing and/or processing chloroplast messenger RNAs, and can be required for photosynthesis. The laboratory also studies the enzyme polynucleotide phosphorylase (PNPase), which can both polymerize and degrade RNA. Surprisingly, PNPase is also required for *Chlamydomonas* cells to survive phosphorus deprivation. This suggests that PNPase activity is not only important for RNA metabolism, but also to help balance phosphate chemistry in the cell.

The laboratory also uses *Arabidopsis* as a model to study chloroplast ribonucleases, including PNPase and RNR1, the latter being required for ribosomal RNA maturation, and thus for protein synthesis. Mutants lacking RNR1 are pale green and stunted, illustrating the importance of correct chloroplast RNA processing for plant growth. Another pair of ribonucleases, CSP41a and CSP41b, regulate chloroplast transcription, perhaps by interacting both with the RNA polymerase and messenger RNAs.

Maize is being used to study the assembly of Rubisco, the major CO₂-fixing enzyme. Rubisco is composed of eight each of large and small subunits: LS being encoded in the chloroplast and SS in the nucleus. The Stern laboratory is investigating the mechanism that controls LS synthesis, based on a model where limiting SS triggers a translational shutdown of LS, mediated by the excess unassembled LS present in the chloroplast. Such mechanisms prevent wasteful synthesis of photosynthesis proteins that would ultimately be degraded, an energetically costly scenario.

How do potatoes produce and accumulate beta-carotene?

Joyce Van Eck, Ph.D.

Senior Research Associate, BTI



According to the World Health Organization, 100 to 140 million children in the developing world suffer from vitamin A deficiency, which can cause blindness and death. Finding ways to produce food with higher beta-carotene content — the precursor to vitamin A — could significantly alleviate this serious health issue.

Working together, **Joyce Van Eck** and Li Li, a scientist with the U.S. Department of Agriculture, developed two lines of modified potatoes that accumulate more beta-carotene than conventional varieties. Van Eck knocked out, or "silenced," a gene in her line that converts beta-carotene into zeaxanthin, another carotenoid that is not converted into vitamin A. She theorized and proved that silencing the gene would cause the potatoes to accumulate more beta-carotene. Li isolated a gene called *Or* from a naturally occurring mutant orange cauliflower. When Van Eck inserted the *Or* gene into potato, they found that the modified potatoes accumulated more beta-carotene than other potatoes.

In 2007, Van Eck's lab worked to better understand the molecular pathway involved in the production of carotenoids in potatoes. In analyzing the *Or* lines, the scientists discovered that early in the pathway (about four steps before beta-carotene is produced), certain genes caused the accumulation of some carotenoids, but limited the accumulation of others. As a result of this discovery, Van Eck's lab inserted certain genes earlier in the pathway to counteract the limiting effect she had found and increase carotenoid production.

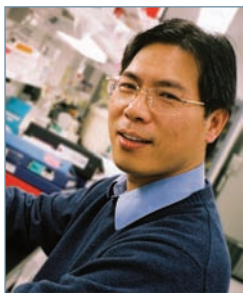
Then, these genes were inserted into both the silenced lines and *Or* lines. Potatoes from these newly modified lines have been harvested, early analyses have been completed, and initial results are encouraging, but Van Eck is waiting for data from the second and third analyses to confirm success. If the plants do produce significantly higher amounts of beta-carotene, the next step will be field trials to ensure that yield, plant health and other factors are not affected in the modified potatoes.

In the meantime, Van Eck is working with the International Potato Center in Peru. Together, they are preparing for the eventual introduction of the more nutritious potatoes into developing countries where vitamin A deficiency is a significant problem.

How do plants perceive and respond to far-red light?

Haiyang Wang, Ph.D.

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Plant growth and development are largely dependent on a plant's response to light. The color or wavelength of light, its intensity, direction and duration influence when, how fast, how tall and in what direction plants will grow, and when they will flower. But understanding fully how plants "see" and respond to light is an enduring mystery.

Haiyang Wang is studying how plants sense and respond to far-red light (light in the visible spectrum that we see at sunrise and sunset), using *Arabidopsis* plants as a model. When far-red light photons hit specialized protein photoreceptors in the cell called phytochrome A or PHYA, the receptors are activated. Activated PHYA molecules then move from the cytoplasm of the cell into the nucleus where they orchestrate the plant's physiological response to light by regulating gene expression. What causes the photoreceptor to move and how the process is regulated is not well understood.

Wang's laboratory is studying the biochemical function of two proteins, FHY3 and FAR1, that appear to be essential for the light response chain of events. They found through a series of biochemical, genetic and cell biological studies that these proteins can bind as transcription factors directly to the regulatory regions of two direct target genes, *FHY1* and *FHL*, and cause the genes to express products required for PHYA to accumulate in the nucleus.

This discovery, recently reported in *Science*, has important evolutionary significance because the two proteins appear to be related to certain enzyme products of transposable elements, which are residents of the genome that can "jump" from one place to another. Wang's research indicates these "jumping genes" may actually have evolved into important cellular genes, which, in this case, allow the plant to respond to its light environment.

The ability to fine tune a plant's response to light could significantly benefit agriculture. Such adjustments could lead to plants that yield more, produce more biomass or could be grown in higher density.

2008 PUBLICATIONS

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