The MicroScope

Department of Microbial Sciences Alumni Newsletter

2007-2008

New Culturing Method Speeds Up Research

Two University of Tennessee scientists discover a way to make quick work of culturing a troublesome cyanobacteria.

Jeff Morris and Erik Zinser

Prochlorococcus (*Pro*) is the smallest marine cyanobacterium on record, measuring only 0.6 micrometers in diameter. Yet what they lack in size, they make up for in numbers: They are the most abundant photosynthetic organisms on Earth. A single drop of sea water can hold up to 10 thousand cells, and they are ubiquitous at the ocean surface between 40 degrees North and South latitude. *Pro* accounts for approximately 20 percent of the oxygen we breathe, and plays a major role in the global carbon cycle. *Pro* thus has a strong influence on the Earth's climate, which makes it an attractive system for research.

Unfortunately, *Pro* isn't very cooperative in laboratory cultures. It grows very slowly, doubling about once every day, and growing pure plate cultures of *Pro* has proved to be more frustrating than successful. If the *Pro* in the culture number fewer than 10 thousand cells per milliliter they die off. That is, unless there are heterotrophic bacteria around.

Assistant Professor Erik Zinser first noticed that Pro would grow on seawater-based agar Petri plates only where there was a contamination of heterotrophic bacteria, suggesting that the heterotrophs were helping *Pro* in some way. Heterotrophic microbial organisms are by definition consumers of organic carbon that originates from primary producers, which fix carbon dioxide into organic

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Message from the Department Head

Dr. Jeffery M. Becker

This represents the first edition of the department's newsletter cleverly called "The MicroScope." Thanks go to Steven Wilhelm, Professor of Microbiology who got the project started and Joel Smithson, an intern from UT's School of Journalism and Electronic Media, who did the majority of writing and all of the formatting. We trust future editions will come out on a regular basis.

Microbiology has a long-standing, strong identity as a distinct scientific discipline supported by extremely robust national and international organizations. Microbiologists have historically contributed to major advances in the biological sciences contributing to humanity's health and welfare. Current scientific developments, such as the rise of multidrug-resistance microbes, the growing awareness of the contributions of microbes to the environment, and the use of microbes in the biofuel industry, indicate that Microbiology is still a leading force and will continue as a major scientific discipline throughout the 21st century.

The Microbiology Department at UT is one of a handful of departments in the U.S. that

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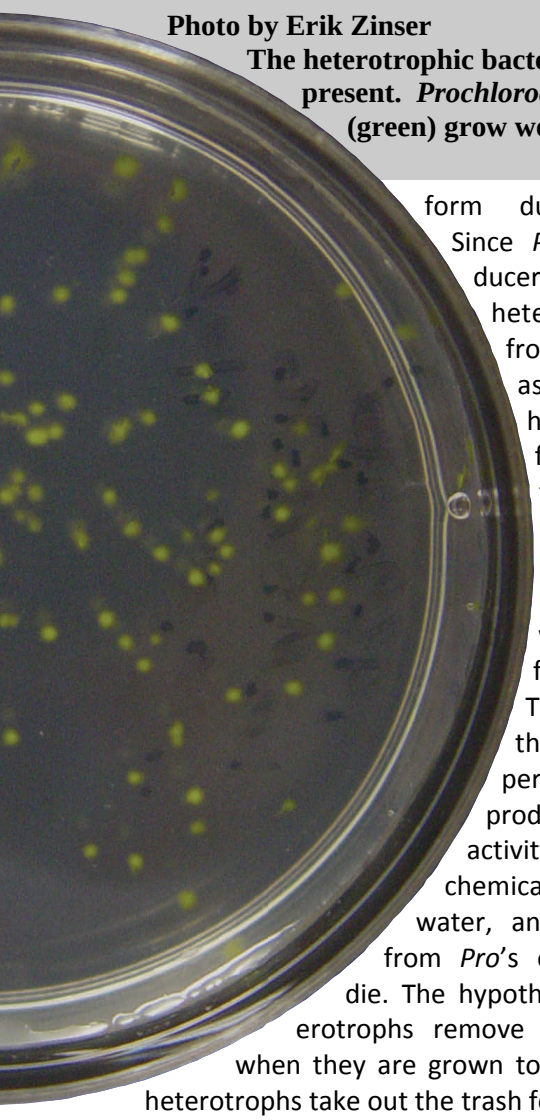

Photo by Erik Zinser
The heterotrophic bacteria are invisible, but present. *Prochlorococcus* colonies (green) grow well in the presence of

form during photosynthesis. Since *Pro* is a primary producer it was likely that the heterotrophs benefited from the presence of *Pro* as a source of food, but how does *Pro* benefit from the presence of the heterotrophic "helper"?

Dr. Zinser theorized that the heterotrophs were protecting *Pro* from oxidative stress. This oxidative stress, in the form of hydrogen peroxide, is a natural by-product of photosynthetic activity and light-driven chemical reactions in the seawater, and if it isn't removed from *Pro*'s environment they will die. The hypothesis was that the heterotrophs remove the peroxide for *Pro* when they are grown together. Essentially, the heterotrophs take out the trash for *Pro.*

This hypothesis makes sense from an ecological and evolutionary point of view. Since the evolutionary origin of *Pro* it has lived in the presence of abundant heterotrophic bacteria. They evolved to live in low-nutrient environments by ridding themselves of as many unnecessary genes as they could in order to conserve resources. Since the heterotrophs were providing ample protection against oxidative stress, *Pro* took advantage and evolved a less adequate ability to protect itself. Each *Pro* cell maintained some ability to remove peroxide, but this ability is effective only when cells are in artificially created high concentrations.

Working with this theory Dr. Zinser and graduate student Jeff Morris discovered a way to generate pure *Pro* cultures by using heterotrophs to sustain them in the early growth stages. This procedure allows scientist to readily obtain pure cultures of this important bacterium, a problem that has impeded studies until now.

In the first stage, heterotrophs are used to grow *Pro* into high concentrations. In the second stage antibiotics are added that specifically target the heterotrophs. Since *Pro* is able to handle oxidative stress on its own in high cell concentrations, all that is left is a culture of healthy, pure *Pro.*

Morris and Dr. Zinser have published their findings in the July issue of the peer-reviewed journal *Applied and Environmental Microbiology*. After such a successful discovery one would imagine a break in the action, but the scientists are determined to take their knowledge onto the seas to take a close look at the real thing.

"We've done it here in the lab and that's what the paper is about," Dr. Zinser said. "Now we're trying to get conducing evidence that heterotrophs are helping *Pro* in the natural environment."

Having readily available plate cultures of such an important family of cyanobacteria will greatly facilitate laboratory studies, allowing scientists to perform more studies quicker on a pure culture. This organism is responsible for 20 percent of the oxygen we breathe, so the sooner we know everything about it the better.

-Joel Smithson-

Head's Message Continued

has strong faculty representing both microbial pathogenesis and microbial ecology. The unique strength of our Microbiology Department has resulted in many strong, funded projects some of which are highlighted in this Newsletter.

It has been an exciting past five years for the Department as we have recruited eight new faculty members with training from institutions such as Harvard, Yale, MIT, UC-San Diego, Washington U., Emory, and St. Jude. These new professors are taking their research and teaching very seriously. We have a primary responsibility for teaching pre-professional students going into medical and environmental fields and many new courses have been developed. We have come a long way in the past five years, and we have a strong vision of our future.

It is a great honor to serve the faculty, students, and alumni as Head of UT's Department of Microbiology. I look forward to working with all of our constituents to make our teaching and research distinguished world-wide.

Mark Sangster and The New York Influenza Center of Excellence

When we are born we receive a cocktail of vaccinations that serve us for life. Yet every year we line up at hospitals and immunization stations to prevent the flu because, year after year, the flu keeps coming back. What quality does the influenza virus have that makes it so successful against vaccination?

Antigenic drift is the process by which the influenza virus is constantly changing and making our immunities obsolete. This series of mutations gradually leads to the evolution of the virus by changing the antigens that the immune system uses to recognize the virus, allowing it to slip past unnoticed.

The desire to create broader vaccinations coupled with the advent of avian influenza, or bird flu, has lead the United States government along with the National Institutes of Health to create six centers of excellence for influenza research. These centers allow the best minds from the fields of immunology, virology, biochemistry, and medicine to collaborate efforts to create a vaccine that can provide protection against the influenza virus regardless of its mutations.

From The University of Tennessee, Immunologist Dr. Mark Sangster is providing the New York Influenza Center of Excellence (NYICE) with expertise on B cell responses to infection.

B cells are white blood cells that fight infections by producing antibodies that bind to and eliminate infectious agents. Some antibodies are produced at mucosal surfaces, such as the lungs, where they provide an important barrier to infection. During infection, B cells are activated when they encounter a viral antigen. They start producing plasma B cells, which secrete antibodies to fight the infection, and memory B cells, which live for a long time. When activated by re-exposure to the specific viral antigen that induced their formation, memory B cells develop into antibody-producing plasma B cells more rapidly than do regular B cells and provide strong resistance to infection.

If the body is infected with the same strain of influenza that stimulated the formation of plasma B cells and memory B cells, then the immune response will be successful in eliminating the virus. However, antigenic drift changes the virus' surface proteins that the memory B cells use for recognition. As a result, the immune system is less effective in fighting the infection. Vaccination with the changed virus is required to provide strong protection.

Dr. Sangster's lab is dedicated to understanding the nature of the B cell response to infection. This information is important in creating a vaccine that would protect against a larger range of influenza viruses rather than just one specific strain. One B cell antibody in particular has Dr. Sangster very interested. Immunoglobulin A (IgA) is the most important antibody that provides protection at mucosal linings. It has a special mechanism that carries it across the mucosal surface and releases it into the open spaces of the lung or nasal passage, where it can combat the influenza virus and other infectious agents.

Recent studies show that IgA provides broader protection against a range of influenza viruses than do other types of antibodies. Its ability to cross mucosal linings and actually enter open airways and its broad protection could be what NYICE is looking for to improve the effectiveness of vaccines. Dr. Sangster feels that a vaccine that produces a large IgA response could provide superior protection against influenza.

Dr. Sangster regularly contributes his findings and advice to NYICE and several times a year makes a trip to the University of Rochester to meet face-to-face with other experts contributing to the cause. He has been researching the influenza virus for over 15 years and remains committed to understanding the immunology of influenza infection and vaccination. After finishing graduate school at the University of Western Australia he held a five year post-doctoral position at St. Jude Children's Research Hospital. Afterwards he stayed at St. Jude and joined the lab of distinguished Immunologist Dr. Peter Doherty, who won the Nobel Prize in Medicine in 1996 for Identifying the mechanism by which immune system T cells recognize other cells as infected and eliminate them. Dr. Sangster said the six years in Dr. Doherty's lab were priceless.

"I really became an immunologist and became interested in B cell responses at St. Jude," he said. "Dr. Doherty is a T cell expert and it was great collaborating with him."

Dr. Sangster joined the University of Tennessee in 2003 and has continued his research on B cell responses to infection in his lab in Walter's Life-Science building. As he continues his work with NYICE his research may one day contribute to a vaccination that makes the flu season a thing of the past.

-Joel Smithson-

Spanning the Globe

Photo by Ryan Kimbirauskas

Subin River, Ghana

Curious children watch as Heather Williamson collects a sediment sample. Williamson and the rest of Dr. Pamela Small's lab focus much of their effort on studying *Mycrobacterium ulcerans*, the causative agent of an emerging disease in West Africa, the Buruli Ulcer.

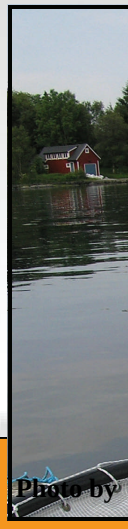
Photo by

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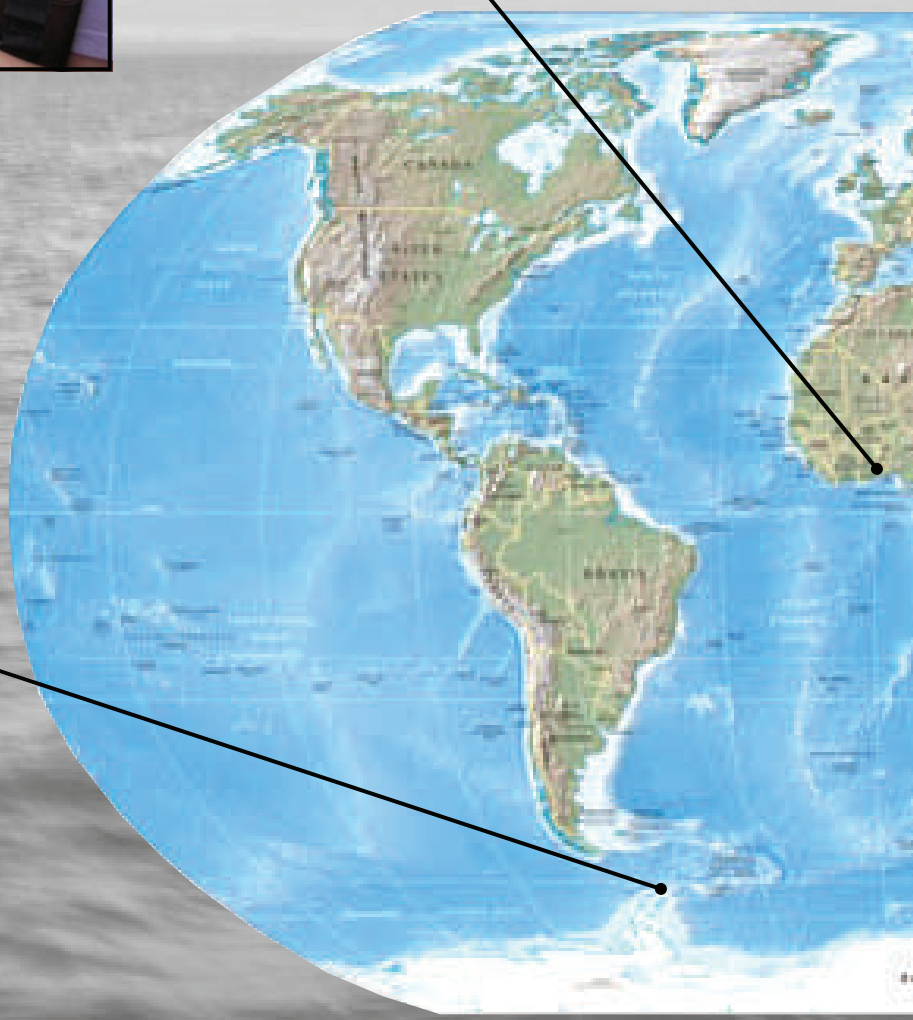
Charles Budinoff tists from ments with con- project was an algae, but Roseobacters, sal relationships

Antartica

Before his post doctoral work at UT, Dr. Gary LeCleir spent six weeks aboard the Antarctic Research and Supply Vessel the Laurence M. Gould. LeCleir was studying the ability of bacteria in Antarctic waters to oxidize ammonia. Ammonia oxidation is the first step in nitrification, which is an important part of the nitrogen cycle in aquatic systems.



Charles Budinoff

way

from Dr. Allison Buchan's lab joined scien- around the globe to run a series of experi- trolled algal blooms in these "bags." The analysis of the impact of viruses on the Budinoff was focusing his attention on heterotrophic bacteria that form commen- with algae. He studied the viruses that

Dacca, Bangladesh

Graduate student Matt Scholz (center, standing) joined researchers from the University of North Carolina and Columbia University to study the geological and microbiological aspects of sub surface aquifers. Along with researchers from the University of Dhaka, the group focused on the effects of arsenic contamination and the transport of pathogens through the aquifers.

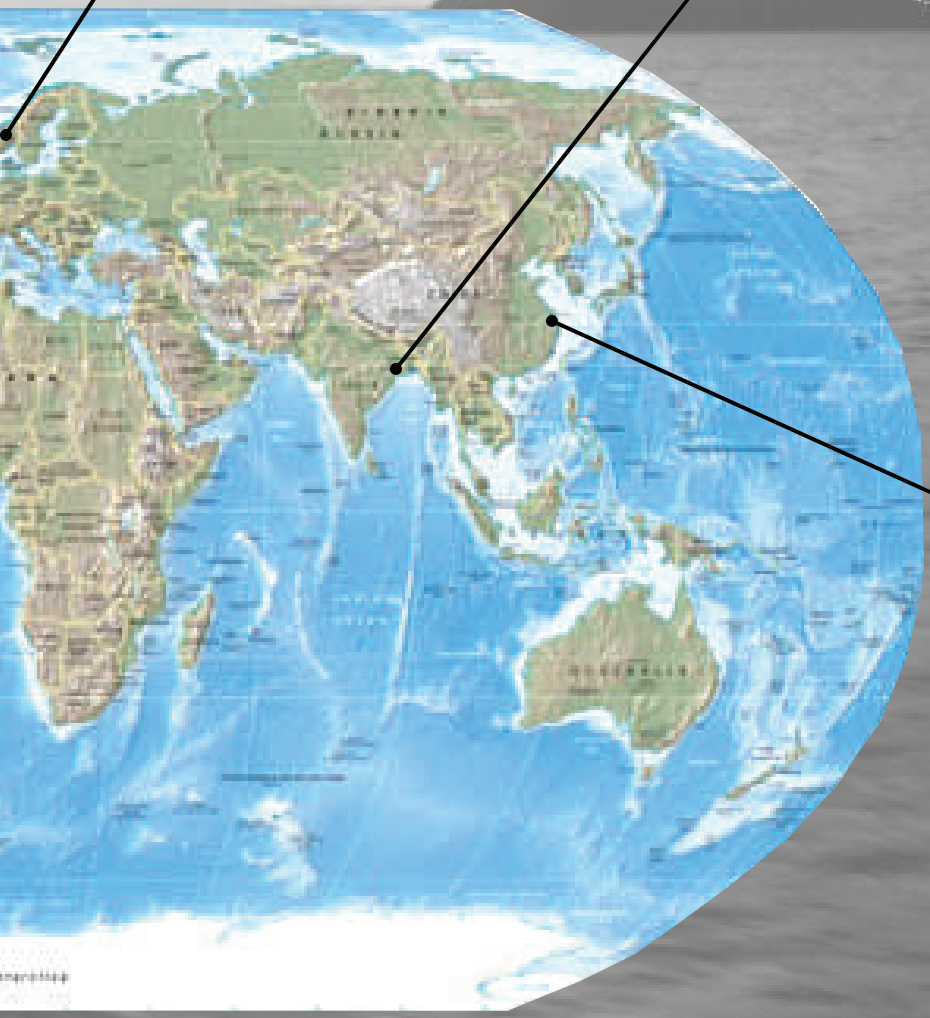


Photo by Dr Qin Boiqiang

Lake Taihu, China

40 million people relied on Lake Taihu for water, fishing or other purposes, but decades of pollution made the water undrinkable, and now there is a destructive algal bloom making things worse. Scientists from UT's Aquatic Microbial Ecology Research Group will head to China to work on the development of controls for toxic *Microcystis* plaguing the lake.

Janet Rowe

From Undergraduate Student to Dr. at UT

In 1999 Janet Rowe walked onto The University of Tennessee's Knoxville campus with a keen interest in microbiology. Just nine years later, Janet is months away from leaving UT with a Ph.D.

During her undergraduate studies Janet's interest grew with every microbiology course she took. She started graduate school in Dr. Steven Wilhelm's lab in 2004 and quickly became well versed in the study of marine viruses and their effect on microbial communities. Over the years she has published several peer reviewed articles and looks forward to defending her thesis this fall. Despite the heavy work load that came with the study of marine viruses, Janet couldn't see herself taking any other path in life. Her fascination with microbial life that began in a high school cell biology class hasn't waned ever since.

"Working in the lab, I realized that this was what I wanted to do," she said. "It's so incredible to me that everything that happens for life to occur can happen in this tiny little space you can't even see with your naked eye."

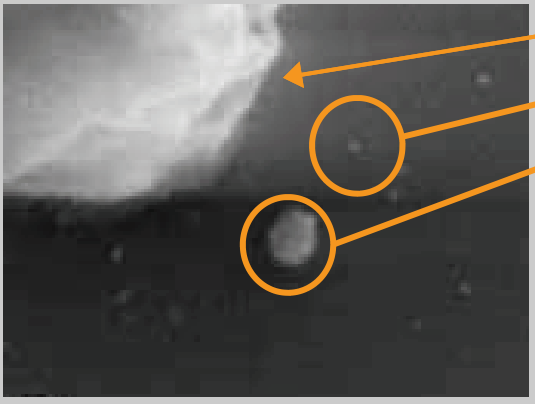
Janet's first big discovery focused her interests on microbial viruses. It started as a lab rotation that is now part of her dissertation and thesis. The alga *Aureococcus anophagefferens* is known for causing 'brown tide' blooms along the Eastern United States coast. These massive blooms wreak havoc on algal grazer populations, such as clams and mussels. In fact, the first 'brown tides' in 1985 damaged the microbial population so severely that a multi-million dollar scallop industry in Long Island collapsed because the scallops' food source was eradicated.

"It's so incredible to me that everything that happens for life to occur can happen in this tiny little space you can't even see with your naked eye."

A. anophagefferens was believed to be commonly infected by phage-like virus resembling those that infect bacteria. These phage were identified as being small in size, approximately 50 nanometers in diameter, and having a protruding tail. Janet was confused by this unusual infection because *A. anophagefferens* is a eukaryotic organism, not bacterium. Similar algae are infected by Phycodnaviridae. Phycodnaviridae is a family of viruses that infect marine and freshwater eukaryotic algae. They are much larger than phage viruses, about 100-200 nanometers, and always without a tail.

Janet found this discrepancy unacceptable because *A. anophagefferens* has such a profound impact on its ecosystem that there is no room for ambiguity. Since these viruses have such an influential effect on *A. anophagefferens*, it is important to understand as much about them as possible. She decided to run a series of tests.

After collecting field samples of

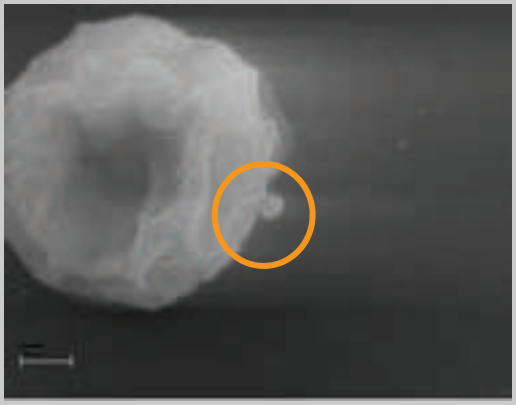


***A. anophagefferens* cell**

smaller phage-like particles

larger Phycodnaviridae-like virus particles

***right:* a larger virus particle attached to an *A. anophagefferens* cell.**



photos taken by Janet Rowe with a scanning electron microscope

A. anophagefferens Janet's colleagues noted that the phage-like viruses were not the only viruses present. There were larger particles that were similar to Phycodnaviridae. Janet observed cultures of *A. anophagefferens* after 30 minutes of exposure to the isolated viruses using a scanning electron microscope (SEM). The SEM works by scanning the culture surface with a beam of electrons that interact with the atoms on the culture, then return information about the physical properties it detected. Janet discovered that 63 percent of the larger viruses appeared attached to *A. anophagefferens* cells while none of the phage-like particles were, contrary to previous reports of phage-like infections.

Janet continued to work with the marine viruses, and in 2005 and 2008 she took her knowledge out of the lab and into the oceans to study virus-host parameters in the Sargasso Sea, the North Atlantic and the Western Pacific. The two expeditions carried her across almost 10,000 miles of water to collect samples of microbial life. The results of her experiments revealed that marine viruses interact with their hosts differently across a multitude of regimes, supporting the argument that the open ocean is full of many microbial ecosystems with unique characteristics. Janet is now working to document these studies, with information from the Sargasso Sea and the North Atlantic as foci for a recently accepted paper.

Due to her professionalism and exceptional research, Janet was award the 2008 Alexander Hollaender Graduate Fellowship in 2007. Dr. Hollaender established the Biology Division of Oak Ridge National Laboratories in 1944. He loved East Tennessee and had a strong desire to support the scholarly achievements of up-and-coming scientists from this region. Janet's research on the open ocean and in the labs expanded the picture of virus effects on photosynthetic organisms and made her a perfect candidate.

Janet has two plans when she earns her Ph.D. The first thing she wants to do is "sleep for about a month." After that, she hopes to earn a faculty position after a post-doctoral research position. The fast-paced academic field has served her well thus far in her career and she wants to return the favor by helping future microbiologists.

Nine years ago Janet Rowe became a student at the University of Tennessee. Soon, she will be leaving as Dr. Janet Rowe. The keen interest in microbiology that she brought here has expanded into a formidable expertise that will serve her, the scientific community, and UT for the rest of her career.

-Joel Smithson-

Awards

From **Dr. Pamela Small's** lab, **Lydia Mosi** has been awarded the 2008 Association for Women in Science Educational Foundation predoctoral award in the amount of $1,000 for her projects: Investigations into the role of the proposed aquatic reservoirs; Naucoridae and Belostomatidae, in the transmission of the Buruli ulcer (Mycobacteriium ulcerans disease). Mosi also received a $250 Wind River travel grant.

From **Dr. Steven Wilhelm's** Lab, **Janet Rowe** has been Awarded the 2008 Alexander Hoellaender Graduate Fellowship Award in the amount of $6,000 for her work on the influences of marine viruses on their microbial hosts and communities

From **Dr. Pamela Small's** lab, **Samantha Wirth**, and from **Dr. Thandi Onami's** lab, **Jenish Patel**, have been awarded the Emerging Infectious Diseases Laboratory Fellowship sponsored by the Association of Public Health Laboratories and Centers for Disease Control.

From **Dr. Todd Reynolds** lab, **Ying-Lien Chen**, and from **Dr. Pamela Small's** Lab, **Heather Williamson**, have been awarded the Science Alliance Award for Outstanding Scholarly Achievement by a Graduate Student.

From **Dr. Chunlei Su**'s lab, **Debashree Majumdar** has been awarded the Graduate Teaching Award for excellence in undergraduate teaching.

Emily Bethea, Jeremy Chandler, Joseph Chen, Li-Yin Huang, Heejung Kim, Jeff Morris, Lydia Mosi, Jenish Patel, Matt Scholz, George Umanah, and **Samantha Wirth** all received the David White Travel Award in the amount of $1,000 for their research.

From **Dr. Thandi Onami's** lab, **Junwei Zeng** received a $1000 travel award from the Society for Glycobiology to travel to the annual meeting in Boston, Mass.

From **Dr. Todd Reynold's** lab, **Joseph Chen** was chosen to present his abstract as a platform talk at the 9th ASM Conference on *Candida candidiasis*. He received an received an award for outstanding presentation in a platform talk which included a free subscription to Eukaryotic Cell and a $500 travel award.

From **Dr. Todd Reynold's** lab, **Emily Bethea** received an award for outstanding oral presentation at the 15th annual Southeast Regional Yeast Meeting in Gatlinburg, Tenn.

From **Dr. Jeffery Becker's** lab, **George Umanah** received a $1,000 travel award from the Keystone Foundation to attend a meeting in Ireland in May on GPCRs.

From **Dr. Mark Sangster's** lab, **Aarthi Sundararajan** and **Lifang Huan** received scholarships to attend the "Immunobiology and Pathogenesis of Influenza Infection" meeting in Atlanta, Ga. in June 2008.

From **Dr. Pamela Small's** lab, **Heather Williamson** received a Burroughs Welcome Travel Award to attend the NIH Ecology of Infectious Disease Meeting in Albuquerque, N.M. in December 2007.

From **Dr. Erik Zinser's** lab, **Jeff Morris** received the award fpr best presentation at the annual Southeastern Phycological Colloquy in October 2007 at Dauphin Island Sea Labs near Mobile, Ala.

Department of Microbiology Publications

2007-2008

The ultimate judge of an academic department's research production is both the quality of the students that are produced and the academic products (scientific publications) from that group. The Department of Microbiology has a rich history of the highest level of academic productivity. Since the beginning of 2007 the faculty and their students have maintained this productivity as is illustrated by their publications in peer-reviewed literature.

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THE UNIVERSITY of TENNESSEE

Department of Microbiology
M409 Walters Life Sciences
Knoxville, Tennessee
37996-0845