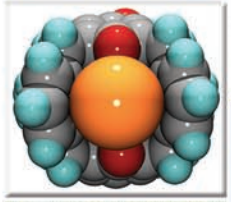
Biological and Environmental Research

BIOLOGICAL AND ENVIRONMENTAL RESEARCH • OAK RIDGE NATIONAL LABORATORY

Annual Report FY 2005



December 2005

BIOLOGICAL AND ENVIRONMENTAL RESEARCH • OAK RIDGE NATIONAL LABORATORY

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Oak Ridge National Laboratory (ORNL) conducts basic and applied research and development to create scientific knowledge and technological solutions that strengthen the nation's leadership in key areas of science; increase the availability of clean, abundant energy; restore and protect the environment; and contribute to national security.

Foreword

Scientists and engineers in the Biological and Environmental Sciences Directorate at the Oak Ridge National Laboratory perform research that is supported primarily by the Department of Energy's Office of Science through the Office of Biological and Environmental Research. The Directorate consists of the Life Sciences Division and the Environmental Sciences Division. We manage a number of research facilities and data archives, including the Mouse Genetics User Facility, the Center for Structural Molecular Biology, the Environmental Remediation Sciences Field Research Center, one of the Free Air Carbon Dioxide Enrichment facilities, the Atmospheric Radiation Measurement Program Data Archive, the Carbon Dioxide Information Analysis Center, and the Oak Ridge National Environmental Research Park.

Our research leads to new knowledge and technologies in areas of critical importance to the nation such as energy security, environmental management, and homeland and national security. We perform research and development that leads to understanding biological systems and their relationship to the environment and to human health, understanding microbial communities and microbe-plant interactions in biogeochemical cycling, measuring and forecasting ecosystem change, and understanding global climate change and to the development of highly effective diagnostic and therapeutic techniques based on fusing nanotechnology with biology from the molecular to the systems level. In addition to work supported by the Department of Energy, we perform research and development supported by other federal and state institutions as well as by the private sector.

This report features selected research highlights by our scientists and engineers during fiscal year 2005. It also summarizes the recognition and awards received by these researchers and provides descriptions of our latest scientific initiatives. You can learn more about the Biological and Environmental Sciences Directorate by visiting our web site at (http://www.ornl.gov/sci/besd/).

Reinhold C. Mann
ORNL Associate Laboratory Director
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Proteomics paper published in Science

A paper describing a study of gene expression in a natural microbial biofilm community was published recently in *Science*. The researchers used mass spectrometry-based proteomics methods to determine the functional activity of microbes in natural acid mining drainage. Nathan VerBerkmoes, Manesh Shah, and Robert Hettich of ORNL are among the co-authors.

Sample of a natural acid mine drainage microbial biofilm.

Microbial communities play key roles in the Earth's biogeochemical cycles. Knowledge of the structure and activities in these communities is limited because analyses of microbial physiology and genetics have been largely confined to studies of organisms from the few lineages for which cultivation conditions have been determined. An additional limitation of purely culture-based studies is that potentially critical community and environmental interactions are not sampled. Recent acquisition of genomic data directly from natural samples has begun to reveal the gene content of communities and environments.

Using genomic and mass spectrometry-based proteomic methods, the authors evaluated gene expression, identified key activities, and examined partitioning of function in a natural acid mine drainage microbial biofilm community. They detected 2,036 proteins from the five most abundant species in the biofilm, including 48% of the predicted proteins from the most abundant biofilm organism, *Leptosprillum* group II. Proteins involved in protein refolding and response to oxidative stress appear to be highly expressed, suggesting that damage to biomolecules is a key challenge for survival. The authors validated and estimated the relative abundance and cellular localization of 216 conserved and 407 unique hypothetical proteins and showed that one abundant novel protein is a cytochrome central to iron oxidation and formation of acid mining drainage. In total, this approach enabled validation of predicted genes and estimation of the relative abundance and cellular localization of expressed proteins, provided clues to protein function, and yielded information about the physiological challenges faced by a self-sustaining, chemolithoautotrophic microbial community (i.e., a microbial community sustained by chemistry other than photosynthesis) in the environment.

R. J. Ram, N. C. VerBerkmoes, M. P. Thelen, G. W. Tyson, B. J. Baker, R. C. Blake II, M. Shah, R. L. Hettich, and J. F. Banfield. "Community proteomics of a natural microbial biofilm," **Science** 308 (5730): 1915–20 (24 June 2005).

Report published on the future of biomass as an industrial feedstock

The U.S. Department of Energy and the U.S. Department of Agriculture are both strongly committed to expanding the use of biomass as an industrial feedstock. In particular, they support the use of biomass in the production of fuels, chemicals, and other products as a way to reduce the need for oil and gas imports; to support the growth of agriculture, forestry, and rural economies; and to foster major new domestic "biorefineries" to make a variety of fuels and other products. As part of this effort, the Biomass R&D Technical Advisory Committee, a panel established by Congress to guide the future direction of federally funded biomass R&D, envisioned a 30% replacement of current U.S. petroleum consumption with biofuels by 2030.

The report, *Biomass as Feedstock for a Bioenergy and Bioproducts Industry*, documents a study that was undertaken to determine whether the land resources of the United States are capable of producing a sustainable supply of biomass sufficient to meet the goal set by the advisory committee. Researchers found potential biomass resources exceeding 1.3 billion dry tons annually, enough to exceed the goal. The full potential of the resource could be realized around the mid-twenty-first century, when large-scale bioenergy and biorefinery industries are likely to exist. This projection is based on a more than sevenfold increase in production from the amount of biomass currently consumed for energy and biomass-based products.

The report has gained prominence in the current climate of rising fuel costs. President George W. Bush has highlighted it in recent speeches on the nation's energy needs, and Secretary of Energy Samuel W. Bodman has discussed the impact of the study on occasion, including his recent visit to ORNL. Others in the biomass field have included the results of the study in their analyses of the status of the bioenergy market and industry, so it is clear that the study is a critical, positive contribution to the field.

R. Perlack, L. Wright, A. Turhollow, R. Graham, B. Stokes, and D. Erbach. **Biomass as Feedstock for a Bioenergy and Bioproducts Industry: The Technical Feasibility of a Billion-Ton Annual Supply**, ORNL/TM-2005/66, DOE/GO-102995-2135 (2005).

Clustered climate regimes to analyze general circulation models

Changes in climate in response to greenhouse gas build-up impact the health of terrestrial ecosystems and the hydrologic cycle. The environmental conditions that influence plant and animal life are often mapped as ecoregions—land areas with similar combinations of environmental characteristics. ORNL scientists have used a modeling technique called multivariate spatio-temporal clustering (MSTC) to establish ecoregions that evolve over time. (Multivariate clustering classifies objects into groups based on the similarity of their properties.)

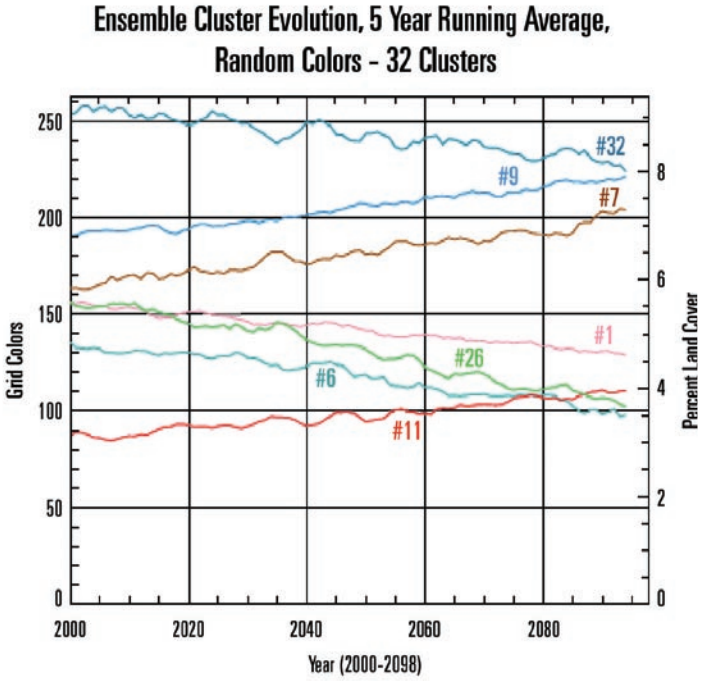
MSTC was applied to the monthly time series output from five 99-year "business-as-usual" transient simulations of the Parallel Climate Model (PCM) from 2000 to 2098.

MSTC establishes an exhaustive set of recurring climate regimes that form a "skeleton" based on the model output from throughout the occupied portion of a climate phase space formed by the characteristics being considered. The analysis included temperature, precipitation, and soil moisture.

Strong predicted year-to-year trends were revealed, including an increase in global desertification; a decrease in the cold, dry high-latitude conditions typical of North American and Asian winters; and significant warming in Antarctica and western Greenland.

MSTC is a powerful tool for helping model developers and environmental decision-makers understand long, complex time series predictions of models. It facilitates direct comparison of ensemble members, ensemble and temporal averages, and observational data because the derived climate regimes provide a basis for comparison.

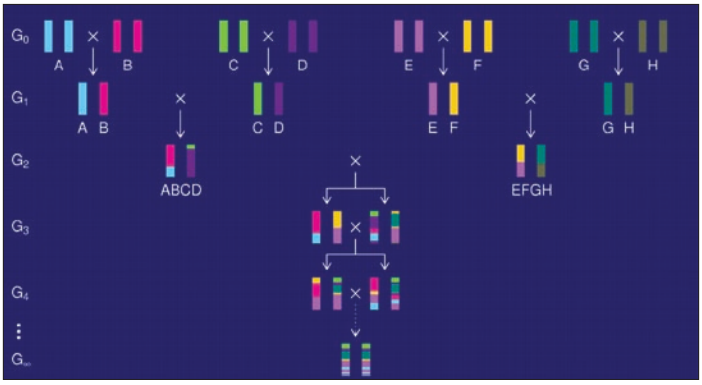
F. M. Hoffman, W. W. Hargrove, D. J. Erickson, and R. J. Oglesby. "Using clustered climate regimes to analyze and compare predictions from fully coupled general circulation models," **Earth Interactions** 9(10): 1–27 (August 2005). Graphics and animations at http://climate.ornl.gov/pcm/.



Ensemble average of five "business as usual" global climate regime evolution curves. The number and its respective color identify each curve as being one of the 32 common climate regimes defined by applying Multivariate Spatio-Temporal Clustering.

The Collaborative Cross: A new approach to human health research

The most common and pervasive human health problems are caused by diseases with complex etiologies. Humans differ greatly in their genetic vulnerability to these common diseases, and the mechanisms that underlie susceptibility and progression are influenced by numerous genetic, developmental, and environmental factors.



The Collaborative Cross strategy will produce recombinant inbred mice specifically for the study of the relationship between complex genetic traits and disease.

ORNL's Collaborative Cross project will provide the international research community with a powerful resource for discovering and analyzing the contributions of those factors to the etiology of common, complex diseases. Eight carefully selected, genetically diverse mouse strains from around the world are being interbred at ORNL's Laboratory for Comparative Functional Genomics to produce a large population of recombinant inbred (RI) mouse strains designed specifically for complex trait analysis. Researchers expect about 1000 strains to be viable as interbreeding proceeds.

The large set of RI strains can support studies incorporating multiple variables into comprehensive statistical models of disease susceptibility, pathophysiology, and progression. The Collaborative Cross mouse population will be unique in providing a comprehensive body of genetic and physiological data derived from a common, reproducible, stable population, in contrast to previous mouse research based on isolated, transient crosses.

The effort will improve modeling of human populations and diseases because scientists will have 1000 lines of mice that carry the kinds of genetic diversity representative of people. Research with this large collection of isogenic strains under controlled environmental conditions will provide data that may ultimately be useful in diagnosing and treating an assortment of chronic human conditions, such as cancer, pulmonary and cardiovascular diseases, diabetes, and behavioral disorders. Software tools developed to predict phenotypes of RI mice will become the prototypes for applications to human health.

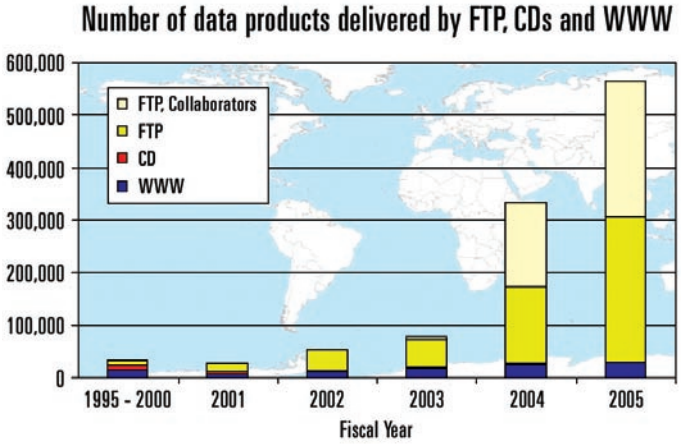
Several universities and institutions, including The Jackson Laboratory, the University of Tennessee, and the University of North Carolina, are participating in the effort.

The Complex Trait Consortium. "The Collaborative Cross, a community resource for the genetic analysis of complex traits," **Nature Genetics** 36(11):1–5 (November 2004).

ORNL DAAC for Biogeochemical Dynamics

Scientists throughout the world are using data from NASA's Distributed Active Archive Center (DAAC) at ORNL to study and assess the impacts of global change. The DAAC archives and distributes terrestrial biogeochemical dynamics data collected as part of the NASA's Earth Observing System (EOS) Program. The DAAC offers more than 760 data sets for four core activities: (1) field campaigns, (2) validation of remote sensing products, (3) regional and global studies, and (4) terrestrial ecosystem models.

Field campaigns combine ground-, aircraft-, and satellite-based measurements of biogeochemical features in specific ecosystems over a few years. They focus on a particular issue or set of issues and are crucial to providing an integrated understanding of biogeochemical dynamics that can be extended across spatial and temporal scales. Understanding environmental processes at multiple scales requires combining and comparing field-based measurements with remote sensing products.



Scientists throughout the world are increasingly using DAAC data to study and assess the impacts of global change.

The ORNL DAAC supports the validation of remotely sensed measurements by providing data, such as net primary productivity (NPP) and leaf area index (LAI), from global test sites for comparison with remote sensing products. The DAAC also offers subsets of selected Moderate-Resolution Imaging Spectroradiometer (MODIS) Land Products for the community to use in conjunction with flux measurements and other field data at the DAAC for studying local-, regional-, and global-scale processes. These subsets show pixel values of MODIS land products in ASCII format for a 7 × 7 km area centered on flux towers or field sites from around the world.

Models that simulate ecosystem properties and processes for improving our understanding of the structure and function of these ecosystems depend on data from multiple spatial and temporal scales. Biogeochemical dynamics data, such as those available from the ORNL DAAC, can be used to parameterize and validate terrestrial ecosystem models at local, regional, and global scales.

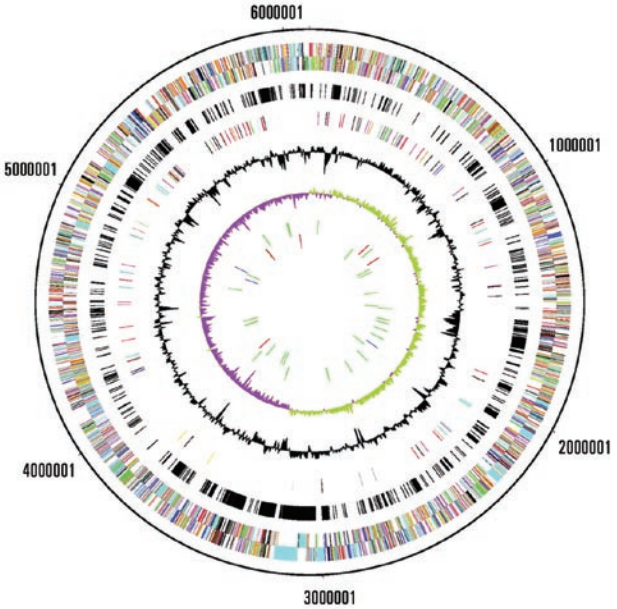
Archived models provide the methodological detail of numerical modeling studies to recreate published modeling results, enabling the synthesis of results across modeling studies and the investigation of new hypotheses. In addition, archived models allow determination of uncertainties for comparison with results from other models in assessment/policy studies. Model source code also allows others to see how models treat individual processes.

Data at the ORNL DAAC are available to the global change research community, policy makers, educators, and the general public at no charge. More than 770 data sets are available from the DAAC; in 2005, more than 560,000 data files were provided to more than 11,000 users.

Economically important plant pathogens compared

A scientific study has compared the complete genomic sequences of two strains of *Pseudomonas syringae*. This economically important species of plant pathogenic bacteria consists of about 50 strains, which differ from each other in pathogenicity and host range. The study of the genomic differences between the two examples has resulted in a better understanding of the mechanisms leading to their pathogenicity. ORNL's Miriam Land and Frank Larimer were among the authors of the paper in which the study is described.

The two strains used in the study were *Pseudomonas syringae pv. syringae* B728a (*Pss* B728a) and *P. syringae pv. tomato* DC3000 (Pst DC3000). *Pss* B728a maintains a large population on the surface of healthy plants, where it is exposed to stressful conditions, such as dryness and sunlight, that are normally hostile to bacterial growth. When a plant's defenses are weakened, Pss can then invade host plants and initiate disease. *Pst* DC3000, however, colonizes poorly on the exterior of plants and multiplies mostly within them.



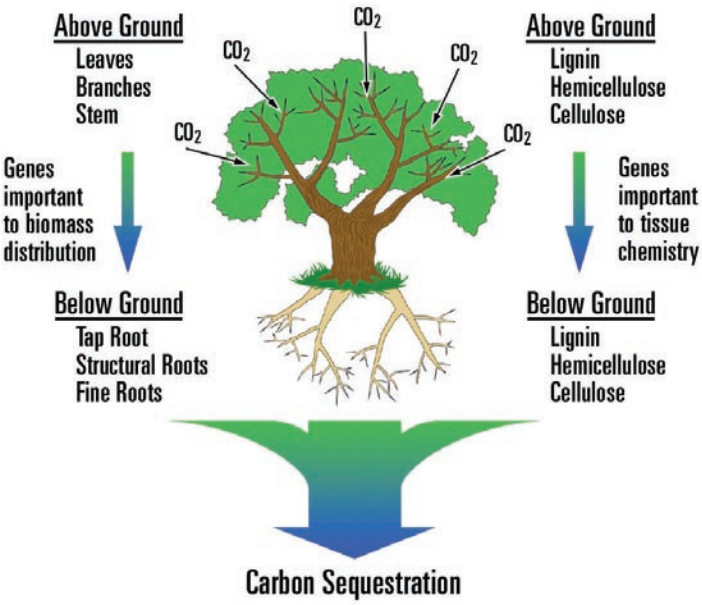
Chromosome of Pss B728a with the outermost circle depicting open reading frames (ORFs) on the positive strand and the second circle depicting ORFs on the negative strand. The ORFs are color-coded based on the major grouping of role categories.

Because environmental conditions are more diverse for a plant's outer surface than they are for its interior, it had been hypothesized that *Pss* B728a has more genes that confer tolerance to environmental stress than *Pst* DC3000 has. The paper shows that among the nearly 1000 genes unique to *Pss* B728a are genes for resistance to ultraviolet light, copper, and streptomycin; tolerance to reactive oxygen species; promotion of ice crystal formation; and production of unique virulence proteins and the mechanisms to introduce them into the plant (type III secretion system effectors). This work serves as the beginning point for associating these genes with specific traits of the bacterial pathogens.

H. Feil, W. S. Feil, P. Chain, F. Larimer, G. DiBartolo, A. Copeland, A. Lykidis, S. Trong, M. Nolan, E. Goltsman, J. Thiel, S. Malfatti, J. E. Loper, A. Lapidus, J. C. Detter, M. Land, P. M. Richardson, N. C. Kyrpides, N. Ivanova, and S. E. Lindow. "Comparison of the complete genome sequences of *Pseudomonas syringae pv. syringae* B728a and *pv. tomato* DC3000," **Proceedings of the National Academy of Sciences of the United States of America** 102 (31): 11064–69 (2005).

Genetic analysis could enhance soil carbon sequestration potential

In a paper published in the August 2005 issue of the *Canadian Journal of Forest Research*, scientists from ORNL provide insights into the genetic mechanisms that control rates and magnitudes of carbon transfer and turnover in plants and soil. The authors are among the first to suggest that regions of the genome could be harnessed to enhance the natural capacity of plants and soils to sequester carbon, thus mitigating rising CO_2 concentrations in the Earth's atmosphere. Incorporating information derived from this study into a model shows that increasing biomass distribution to roots and altering tissue chemistry to favor long-term pools of soil organic matter could enhance the rate of global carbon sequestration by an estimated 0.35 gigatons of carbon per year (about 4% of global carbon emissions).



Conceptual diagram showing how genes for biomass distribution and tissue chemistry can influence carbon sequestration in soils.

Scientists reached this conclusion by assembling an extensive above- and below-ground carbon inventory for more than 1000 hybrid poplar (*Populus*) trees. Field data collected over 3 years were combined with a newly developed genetic map to identify regions of the genome responsible for the distribution of dry mass to stems, branches, leaves, and roots. Results indicate that traits associated with distribution of dry mass were controlled by a small number of genes and that different genes controlled the above- and below-ground production of biomass. Such findings provide a glimpse into how fundamental knowledge gained through the basic biological sciences can address questions related to carbon management in terrestrial ecosystems.

S. D. Wullschleger, T. M. Yin, S. P. DiFazio, T. J. Tschaplinski, L. E. Gunter, M. F. Davis, and G. A. Tuskan. "Phenotypic variation in growth and biomass distribution for two advanced-generation pedigrees of hybrid poplar," **Canadian Journal of Forest Research** 35(8): 1779–89 (2005).

CSMB highlights: a new lab and two notable publications

The Center for Structural Molecular Biology (CSMB) is leading research and development in neutron structure analysis of proteins and protein complexes that will support the user access and biology research programs at the High Flux Isotope Reactor (HFIR) and the Spallation Neutron Source (SNS) at ORNL. The CSMB has established capabilities and critical expertise in areas extending from the design, development, and operation of advanced neutron instrumentation for biology to development of isotopic labeling systems for biological macromolecules. Advanced methods are being developed for the characterization and analysis of the structure and function of complex biological macromolecules that extend from the atomic level of detail to their integrated function in the molecular mechanisms of the cell.

Neutron scattering provides a unique, nondestructive probe of delicate biological materials and higher-order assemblies, and the design and production of hydrogen/deuterium (H/D)-labeled material permit selected parts of macromolecular structures to be highlighted and analyzed in situ. CSMB and the Life Sciences Division have established a Bio-Deuteration Laboratory for in vivo production of H/D-labeled bio-macromolecules to support the user research programs at HFIR and SNS.

In an article featured on the cover of *Protein Science*, William Heller and colleagues described a solution-scattering analysis of nonactivated and Ca^{2+}-activated phosphorylase kinase that produced conformer models consistent with

Ca^{2+}-induced conformational changes in both the lobes and the interlobal bridges of the 1.3-MDa $(\alpha\beta\gamma\delta)_4$ hexadecameric complex. The modeling techniques developed and used in this work will enable functional models of multi-meric complexes to be derived from neutron scattering experiments at HFIR and at SNS.

In an article published in the *Proceedings of the National Academy of Sciences* and listed as a Research Highlight in *Nature*, Dean Myles and colleagues described the first example of a protein structure determined at cryogenic temperature (15 K) by neutron crystallography. Their work on the plant lectin Concanavalin A showed how the reduced dynamic disorder in the neutron structure at low temperature enabled twice as many bound water molecules to be identified at 15 K as at 293 K. This methodology opens up new categories of neutron protein crystallography, including freeze-trapped reaction and intermediate structures, that will be of interest to the development of the Macromolecular Diffractometer at SNS.

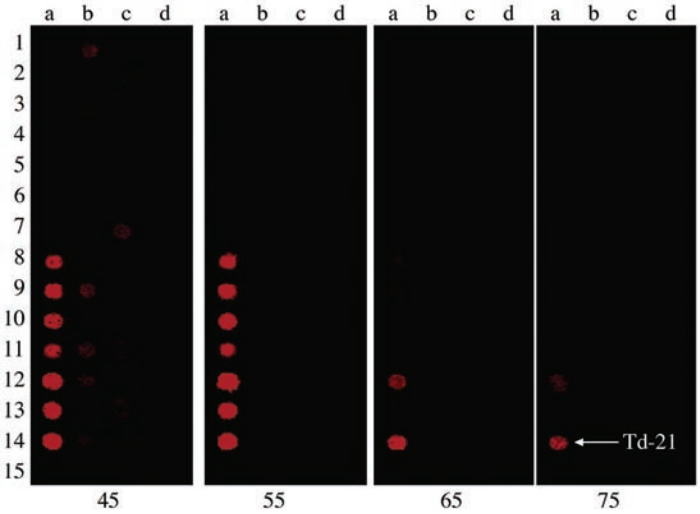
T. S. Priddy, B. A. Macdonald, W. T. Heller, O. W. Nadeau, J. Trewhella, and G. M. Carlson. "Ca^{2+}-induced structural changes in phosphorylase kinase detected by small-angle X-ray scattering," **Protein Science** 14 (4): 1039–48 (April 2005).

M. P. Blakeley, A. J. Kalb, J. R. Helliwell, and D. A. A. Myles. "The 15-K neutron structure of saccharide-free Concanavalin A," **Proceedings of the National Academy of Sciences of the United States of America** 101 (47): 16405–10 (November 23, 2004).

Community genome array identifies species in natural microbial communities

When communities of microorganisms in soil, sediment, or water are exposed to toxic contaminants or temperature changes induced by industrial practices or climate change, which species of bacteria survive these changes and which ones die out? To determine changes in the structure, composition, and adaptive responses of microbial communities, biologists need a way to identify the microorganisms dominating various communities in environmental samples. ORNL researchers have fabricated a microarray-based tool that shows promise as a highly specific, sensitive, and quantitative tool for distinguishing between different species and strains of microorganisms in environmental samples. Called a community genome array (CGA), this glass slide is dotted with labeled whole-genome "probes" that pair up, or hybridize, with "target" microorganisms of genetically identical species in natural microbial communities. CGAs may also be practical for detecting unknown microorganisms that are related genetically to bacteria known to be useful for cleaning up contaminated ecosystems. A part of this study was recently published in *Environmental Science and Technology*.

L. Wu, D. K. Thompson, X. Liu, M. W. Fields, C. E. Bagwell, J. M. Tiedje, and J.-Z. Zhou. "Development and evaluation of microarray-based whole-genome hybridization for detection of microorganisms within the context of environmental applications, **Environmental Science and Technology** 38: 6775–82, 2004.



Fluorescence images showing the improvement in array hybridization specificity with increasing temperature. Genomic DNA from an A. tolulyticus isolate Td-21 was randomly labeled with Cy5 and hybridized with CGA in the presence of 50% (v/v) formamide at 45, 55, 65, and 75°C. The hybridization signal for Td-21 genomic DNA is indicated (white arrow).

New process treats legacy radioactive wastes at lower cost

DOE selected the caustic-side solvent extraction (CSSX) process developed at ORNL to remove radioactive cesium from alkaline nuclear waste stored at its Savannah River Site in South Carolina. The radionuclide ^{137}Cs is present in the waste at low concentrations, yet it is responsible for a large portion of the radioactivity. It is necessary to remove the bulk of the radioactive ^{137}Cs for safe remediation of these legacy wastes.

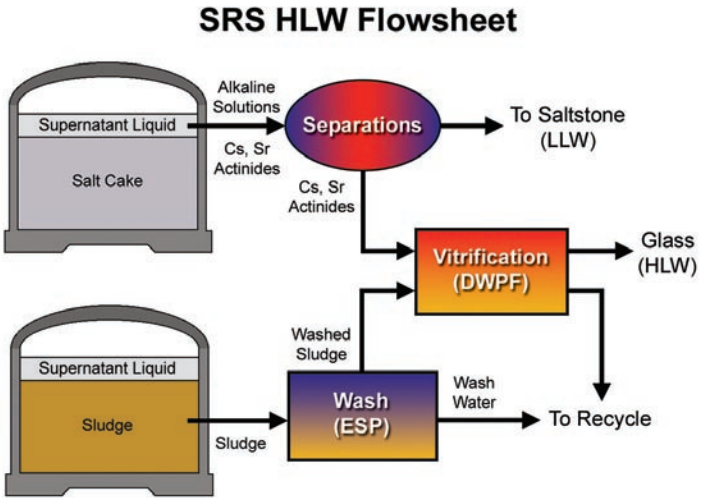
CSSX uses a powerful extractant that is capable of selectively removing cesium from the waste, which can include more than 10,000-fold higher concentrations of competing metal ions. The extractant is dissolved in a kerosene-based solvent; when the solvent contacts the waste, the cesium transfers from the waste to the solvent. The range of appli-

cability of the solvent is influenced by the solubility of the extractant, which is low for the solvent now in use. A new, more soluble version of the cesium extractant prepared at ORNL has been shown to be at least eight times more soluble in the kerosene solvent, thus expanding the potential applications of the technology.

ORNL has recently developed a new process concept involving adding crown ether and carboxylic acid to the CSSX solvent that, if it proves feasible, would extend CSSX capabilities to removing both cesium and strontium in one step. This could lead to additional cost savings and waste volume reduction. Ultimately, researchers hope to develop a process to extract uranium, neptunium, and plutonium as well.

CSSX will be implemented in two ways at the Savannah River Site. The biggest application is the Salt Waste Treatment Facility, a $1 billion plant being designed by private industry to remove 99.9975% of the radioactive cesium from up to 34 million gallons of cold-war high-level-waste stored in underground tanks. CSSX is the centerpiece technology in that plant, slated to begin operation in 2010. A smaller mobile CSSX unit is being designed for implementation within the next 2 years.

Batch tests on Hanford wastes show potential for use of CSSX at Hanford, and an acid-side variant of the process may be applicable to Idaho Nuclear Engineering and Environmental Laboratory waste. The product stream from CSSX is ideal for vitrification or other end processing. Very little secondary waste is generated, and the solvent is reusable for at least a year.



The CSSX process.

Development of a domain map for nodes of the National Ecological Observatory Network (NEON)

The National Ecological Observatory Network (NEON), funded by the National Science Foundation, will be the first ecological measurement system designed both to answer regional- to national-scale scientific questions and to have the interdisciplinary participation necessary to achieve credible ecological forecasting and prediction. Capabilities provided by this infrastructural investment will transform the science of ecology by enabling the integration of research and education from natural and human systems.

A National Network Design Committee (NNDC) of 15 individuals has been tasked with providing a baseline design for NEON, including the continental-scale deployment of NEON network resources. A system of identical nodes, each representing environments within a mother geographic "domain" was envisioned. Each node would itself consist of sub-node components, and all nodes would be focused in unison on a few transformational ecological questions of national relevance.

The NNDC adopted a strategy of prestratification to help determine an optimum number of nodes and to maximize node representativeness. To better sample a phenomenon as diverse as the ecological environments of the United States, those environments were first divided into a set of more homogeneous "strata." Samples could then be arrayed within each stratum, ensuring that NEON nodes are representative of the entire range of environments within the United States.

Dr. William W. Hargrove, who serves on the NNDC, and Forrest Hoffman, both at ORNL, performed a set of statistical ecoregion analyses to assist NEON in determining the geographic domains on which the network would be based. Ecoregions have classically been used by ecologists for such national stratification. Ecoregions have historically been drawn using human expertise in a qualitative, weight-of-evidence approach. To construct NEON domains, a more transparent and repeatable process was needed.

Hargrove and Hoffman used multivariate clustering based on national maps of 9 ecologically relevant climatic "state" variables to repeatably define 25 national climatic zones. The NNDC combined these 25 climate zones with dynamic air mass seasonality data to create 20 NEON domains, each having similar climate.

Such domains are defensible in that the method used to generate them is empirical and data-driven. This analysis

was also used, along with budgetary constraints, to determine the number of nodes that would be necessary to adequately sample the climatic environments within the lower 48 United States.

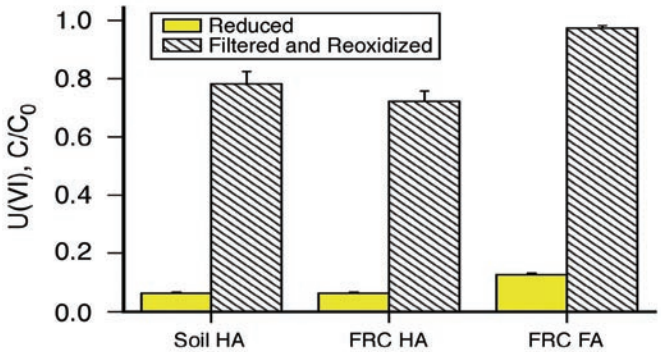
A preliminary version of the NEON domains map was unveiled at the August 2005 Ecological Society of America meeting in Montreal. Such domains provide the NEON design with a statistically based scientific underpinning and will make NEON the first national ecological network that has been statistically designed prior to deployment.

NEON web site: www.neoninc.org

New study shows that humic substances have a surprising influence on uranium stability and mobility

Humic substances, naturally forming organic materials in soil and groundwater, have been shown in past studies to promote the biological reduction and therefore immobilization of reduced uranium(IV) solid owing to its relatively low solubility. Researchers at ORNL recently discovered that, under certain conditions, the same humics can also enhance the reverse reaction; that is, they can make the precipitated solids more soluble in groundwater. This new study suggests that humics can form soluble complexes with precipitated uranium(IV) solids and that the complexes are readily oxidized upon contacting oxygen. Therefore, humics could present a challenge in maintaining the stability of uranium solids. The findings are significant because a proposed remedial strategy for contaminated soil and groundwater is predicated on the lower solubility of reduced uranium in subsurface environments.

B. Gu, H. Yan, P. Zhou, D. Watson, M. Park, and J. D. Istok. "Natural humics impact uranium bioreduction and oxidation," **Environmental Science & Technology** 39(14): 5268–75 (2005).



After the bioreduction, more than 70% of reduced U(IV) in the presence of humics readily passed through 0.2-micrometer syringe filters and was oxidized to U(VI) in open air.

Chapter on remediation to appear in a book about perchlorate

Baohua Gu and Gilbert M. Brown at ORNL have written a chapter for a book about environmental perchlorate and its treatment. Dr. Gu is also one of the book's editors. The book is currently in press and will be available early 2006.

The field-demonstrated ORNL perchlorate treatment system using highly selective ion exchange and perchlorate destruction technologies.

Perchlorate, a chemical mostly used in ordnance and rocket engines, poses a significant threat to human health because it disrupts thyroid function by inhibiting iodide uptake. It has recently been found to be a widespread contaminant in U.S. groundwater and surface water. This water-soluble, environmentally persistent compound now contaminates the drinking water of tens of millions of people and has been found in milk, lettuce, and many other food products in a number of supermarkets. The U.S. Environmental Protection Agency, which has reported perchlorate contamination in groundwater or surface water in more than 29 states, proposed a maximum contaminant level (MCL) of 1 ppb (μg/L) for drinking water in 2002 and recently raised the MCL to ~24 ppb on the basis of National Research

Council recommendations. Cleanup costs are estimated to be in the tens of billions of dollars.

The chapter offers background on treatment strategies and describes a treatment system developed at ORNL. Prior to the work at ORNL, two treatment systems had been devised, one in which the treatment resin is used only once and another in which the resin is regenerated. Both move the perchlorate from one medium to another and thus perpetuate the waste-disposal problem. In the ORNL system, the perchlorate is trapped in a treatment bed of highly specific resin during one cycle; the resin is then regenerated and perchlorate is destroyed during a second cycle. The reaction that destroys perchlorate produces a chemical that is used to regenerate the resin, resulting in a cost reduction of up to 80% and practically zero secondary waste production.

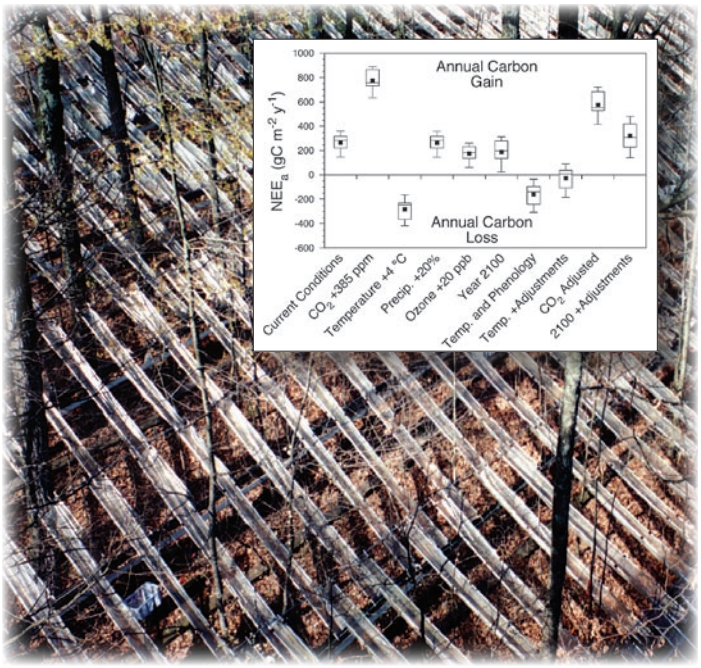
B. Gu and G. M. Brown. "Chapter 11. Field demonstration using highly selective, regenerable ion exchange and perchlorate destruction technologies for water treatment," in **Perchlorate: Environmental Occurrence, Interactions, and Treatment**, ed. B. Gu and J. D. Coates, Springer (in press).

Incorporating experimental results in forest response simulations

Accurate predictions of how eastern deciduous hardwood forests will respond to multiple environmental changes associated with climatic change are a key output for the U.S. Climate Change Science Program. Researchers at ORNL have incorporated their experimental findings from a range of field experiments—including elevated CO_2, warming, precipitation change, and increased tropospheric ozone (O_3)—into stand-level models to predict future responses of eastern forests in the year 2100.

Single-factor model predictions demonstrated a maximum sensitivity of eastern forests to CO_2 and temperature change with minor effects from precipitation change or ozone exposure. Multi-factor simulations that ignored experimentally observed vegetation adjustments predicted a small long-term reduction in forest carbon gain by 2100. However, the opposite pattern (a 20% increase) was obtained when "lessons learned" from experimental studies were included in the analyses. Our analyses identified critical areas of uncertainty for multivariate predictions of future ecosystem response and underscored the importance of long-term field experiments for the characterization of tissue and plant acclimation and growth responses under complex environmental scenarios.

P. J. Hanson, S. D. Wullschleger, R. J. Norby, T. J. Tschaplinski, and C. A. Gunderson. "Importance of changing CO_2, temperature, precipitation, and ozone on carbon and water cycles of an upland-oak forest: Incorporating experimental results into model simulations," **Global Change Biology** 11: 1402–23 (2005).



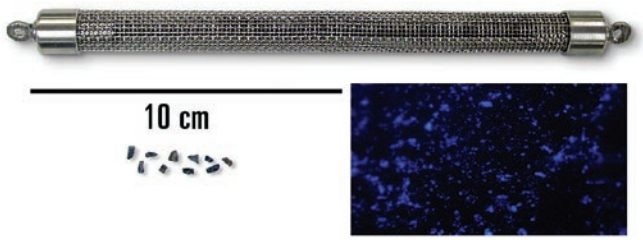
Experimental data inform model predictions of future climatic change.

Subsurface contaminant research and imaging at FRC

Multidisciplinary teams of researchers from across the United States and overseas working at the Environmental Remediation Science Field Research Center (FRC) in Oak Ridge have shown that microorganisms found in subsurface environments can be used to reduce health risks at DOE waste sites by transforming radionuclides, such as uranium and technetium, and other contaminants into chemical forms that are less mobile or less toxic in groundwater. FRC researchers found that introducing naturally occurring humic substances (organic matter found in soil) can accelerate the chemical reduction and immobilization of these contaminants. At the same time, the researchers demonstrated that co-contaminants in the subsurface, such as nitrate, and elevated concentrations of other chemicals, like calcium, can inhibit the chemical reduction process and can reoxidize uranium, making it more mobile.

Extensive work has been conducted to identify the microorganisms present in the harsh FRC subsurface environment (an environment that is acidic and that contains high

concentrations of nitrate and metals that tend to be toxic to most microorganisms). Work conducted to date has begun to determine which specific microorganisms can be used to promote the chemical reduction of radionuclides directly or indirectly. This research relies on genomic sequencing; cutting-edge techniques such as the use of functional gene arrays; and such novel devices as "bug traps," coupons that trap microbes below the ground's surface.



Coupon or "bug trap" deployed in the subsurface. Inset: microorganisms growing on media surfaces.

In addition to investigating naturally occurring microbial communities in the FRC's subsurface, researchers have developed novel geophysical, hydraulic, and tracer techniques for characterizing and monitoring subsurface processes and groundwater flow. For example, they have developed inexpensive surface geophysical techniques in which seismic waves and electrical currents are used to create three-dimensional images of the subsurface geology and of contaminated groundwater plumes. Taken together, FRC research findings have contributed to the DOE Office of Biological and Environmental Research goal of understanding the processes that influence the transport and fate of subsurface contaminants, the effectiveness and long-term consequences of extant remediation options, and the development of improved remediation strategies—especially for currently intractable contaminants or conditions. The FRC has also provided valuable opportunities to researchers engaged in genomics research. As examples, researchers associated with the Joint Genome Institute and the DOE Genomics: GTL program use FRC samples in their investigations.

D. B. Watson, W. E. Doll, T. J. Gamey, J. R. Sheehan, and P. M. Jardine. "Plume and lithologic profiling with surface resistivity and seismic tomography," **Ground Water** 43(2): 169–77 (2005).

J. D. Istok, J. M. Senko, L. R. Krumholz, D. Watson, M. A. Bogle, A. Peacock, Y. –J. Chang, and D. C. White. "In situ bioreduction of technetium and uranium in a nitrate-contaminated aquifer," **Environmental Science & Technology** 38(2): 468–75 (2004).

M. W. Fields, T. F. Yan, S. K. Rhee, S. L. Carroll, P. M. Jardine, D. B. Watson, C. S. Criddle, and J. Z. Zhou. "Impacts on microbial communities and cultivable isolates from groundwater contaminated with high levels of nitric acid-uranium waste," **FEMS Microbiology Ecology** 53(3): 417–28 (2005).

Subsurface Remedial Sciences

Soil, the thin veneer of matter covering the earth's surface and supporting a web of living diversity, is often abused through anthropogenic inputs of toxic waste. The disposal of radioactive and metal waste generated at DOE facilities within the weapons complex has historically involved shallow land burial in unsaturated soils and sediments. Disposal methods from the 1940s to the 1980s ranged from unconfined pits and trenches to single- and double-shell buried steel tanks. The scope of DOE's disposal problem is massive, with landfills estimated to contain more than 3 million cubic meters of radioactive and hazardous buried waste, a significant proportion of which has migrated into surrounding soils and groundwater. Contaminated sites are closing rapidly as a result of the demand for accelerated cleanup, and many remediation strategies have chosen to leave contaminants in place (e.g., by capping). Contaminants continue to interact with subsurface and surface media. These interactions are controlled by a complex coupling of hydrological, geochemical, and microbial processes.

Recent capping of trenches at ORNL. The contaminants were left in place.

A comprehensive, mission-driven science program within the Environmental Sciences Division (ESD) has been under way for the past 15 years. The program integrates ecosystem-based contaminant fate and transport processes across all scales. Our goal is to be able to understand and predict the consequences of our action or inaction with regard to contaminated environments (the consequences if buried waste is left in place or if waste leaks into the subsurface

during retrieval). For example, in 2005 waste trenches were aggressively capped at ORNL. We observed a decrease in water level and the site hydraulic gradient, as well as shifts in geochemistry relative to pre-cap conditions, which suggested a local-scale influence from the cap. However, hydraulic head responses to storm events were of similar magnitude for post-cap conditions relative to pre-cap conditions, suggesting the regional-scale influence of groundwater flow and the continued discharge of contaminants from the burial grounds.

In general, experiment and numerical subsurface science research at ESD have provided knowledge and information in previously unexplored areas of fate and transport in the vadose zone (between the soil surface and the water table) to support site performance/risk assessment and decision-making processes for site restoration. Further, the research has combined DOE's commitment to environmental restoration with its commitment to major user facilities (the Stanford Synchrotron Radiation Laboratory, the Advanced Photon Source, and the Environmental Molecular Sciences Laboratory) and academic education (various universities and the Oak Ridge Institute for Science and Education).

M. A. Mayes, P. M. Jardine, T. L. Mehlhorn, B. N. Bjornstad, J. L. Ladd, and J. M. Zachara. "Hydrologic processes controlling the transport of contaminants in humid region structured soils and semi-arid laminated sediments," **J. Hydrol**. 275:141–61 (2003).

Swaja elected AIMBE fellow

Richard Swaja was elected to the College of Fellows of the American Institute of Medical and Biological Engineering (AIMBE). He was officially inducted as a fellow at the Annual AIMBE Meeting in February 2005 at the National Academy of Sciences. The election was based on his "many distinguished contributions to the field as well as his demonstrated interest, concern, and involvement with critical issues affecting medical and biological engineering."

Since October 1, 2005, Dr Swaja has been the Director of the Life Sciences Division at ORNL. Between September 1999 and October 2005, he was on assignment to the National Institutes of Health as the Senior Science Advisor for the National Institute of Biomedical Imaging and Bioengineering (NIBIB). In addition to supporting NIBIB science programs, he coordinated interagency, trans-NIH, and multi-organizational activities involving biomedical research and training.

Watson coordinates review of NRC airborne particulates research

ORNL scientist Annetta Watson was appointed by the National Research Council (NRC) to oversee the technical review of its fourth and final report on airborne particulate matter. As review coordinator, her job was to ensure that an independent examination was conducted according to NRC guidelines and that all reviewers' comments were carefully considered by the authoring committee. Watson was supported in this activity by program development funds made available through the directorate.

At the request of Congress and the U.S. Environmental Protection Agency, the NRC established the Committee on Airborne Particulate Matter in 1998 as a standing committee for the purpose of improving scientific understanding of particulate matter and its human health effects. This final report is the culmination of the committee's 6 years of work. The document was published in November 2004.

Committee on Research Priorities for Airborne Particulate Matter. **Research Priorities for Airborne Particulate Matter. IV. Continuing Research Progress**, Board on Environmental Studies and Toxicology, Division on Earth and Life Sciences, National Research Council. The National Academies Press, Washington, D.C. (www.nap.edu), 2004.

Thundat honored by APS and Scientific American

Thomas Thundat of ORNL received the Jesse W. Beams award for research excellence at the banquet for the Seventy-First Annual Meeting of the Southeastern Section of the American Physical Society (SESAPS). The Beams Award, first presented in 1973, was established by SESAPS to recognize especially significant or meritorious research in physics, the major portion of which was carried out in the ten-state SESAPS region in the Southeast. The award is named for Jesse Wakefield Beams, whose contributions include construction of the first electron linear accelerator; development of the magnetic ultracentrifuge, with many practical applications in both the physical and biological sciences; and improvement of the Cavendish technique for the determination of the gravitational constant.

Thundat was also chosen as one of the "Scientific American 50 Award" winners this year. The magazine notes "those who during 2003–2004 exhibited outstanding technology leadership in the realms of research, business and policy making." Thundat's award, which he shares with Jesse Adams, of the University of Nevada at Reno, is based on his work with microcantilevers for TNT detection.

FLC recognizes ORNL microcantilever-based research

The Photo-Molecular Comb™ Biomolecular Separator won a Federal Laboratory Consortium (FLC) Award for Excellence in Technology Transfer in the 2004 Southeast Regional competition. It went on to win at the national level and was honored again in May 2005. The Photo-Molecular Comb was developed in the Life Sciences Division in a col-

laborative effort with other ORNL divisions and with Protein Discovery, Inc., a privately held life sciences company based in Knoxville, Tennessee.

ORNL developers are Thomas Thundat and Thomas Ferrell (Life Sciences), Gilbert Brown (Chemical Sciences), Robert Warmack (Engineering Science and Technology), and Russ Miller (Technology Transfer and Economic Development). Developers from Protein Discovery are Charles Witkowski, Jay Harkins, Dean Hafeman, and Kilin Dill.

The Photo-Molecular Comb separates proteins and other biomolecules for identification and analysis. The entire device is miniature; separations take place in a thin sieving layer on the surface of a semiconductor chip. The sieving layer-semiconductor interface is electrified when illuminated with a directed low-power light source, creating localized charges that attract, concentrate, transport, and separate biomolecules.

Unlike microfluidic devices, the Photo-Molecular Comb does not require prefabricated channels in the semiconductor.

Thomas Thundat, Life Sciences Division (left), and Jay Harkins, Protein Discovery, Inc.

The separation concept, developed under an ORNL Seed Money effort, has resulted in two pending patent applications and three new invention disclosures. Protein Discovery has licensed the technology from ORNL and has developed a product, the Serum Profiler™. With the Serum Profiler, Protein Discovery will provide customers in the clinical research and drug discovery and development industries with an ultrahigh-throughput system for preparing complex biological samples, such as human serum, for analysis via mass spectrometry. A cooperative research and development agreement is active with ORNL for continued research and development.

Borges participates in National Academies Toxicity Review

H. T. Borges of ORNL was selected by the National Research Council to serve on the Board on Environmental Studies and Toxicology's Subcommittee on Iodotrifluoromethane. The report, *Iodotrifluoromethane: Toxicity Review*, was released September 2004.

The U.S. Army Center for Health Promotion and Preventive Medicine requested that the National Research Council conduct an independent assessment of the toxicity of iodotrifluoromethane (CF_3I). Iodotrifluoromethane is under consideration by the U.S. Army (and others) as a replacement for halons as fire suppressants in aircraft and ground vehicles; consequently, there is a high potential for exposure. The review identified significant concerns about the validity of the canine cardiac sensitization studies and physiologically based pharmacokinetic modeling. This review has identified for the U.S. Army, and others considering the use of CF_3I, critical information gaps that must be addressed to provide a better understanding of toxicity to ensure the safe application of this chemical.

Committee on Toxicology, Subcommittee on Iodotrifluromethane. **Iodotrifluoromethane: Toxicity Review**, Board on Environmental Studies and Toxicology, Division of Earth and Life Studies, National Research Council, The National Academies Press, Washington, D.C. (www.nap.edu), 2004.

Pinnaduwage honored by American Physical Society

Lal A. Pinnaduwage has been elected a fellow of the American Physical Society (APS). Pinnaduwage is a researcher in the Life Sciences Division's Advanced Biomedical Science and Technology Group and was a member of the team that won an R&D 100 award in 2004 for SniffEx, a system for detecting explosives. He was selected "for his pioneering work in developing micromechanical cantilever-based sensors for detection of explosive vapors and elucidation of fundamental physical

principles underlying the thermally induced nanodeflagartions for chemical selectivity."

Election to fellowship in the APS is limited to no more than one half of one percent of the membership and is in recognition of outstanding contributions to physics. The APS Fellowship Program was created to recognize advances made through research, publication, and contributions in the application of physics to science and technology. It may also recognize significant contributions to teaching, service to APS, and participation in APS activities.

Jizhong Zhou elected ASM fellow

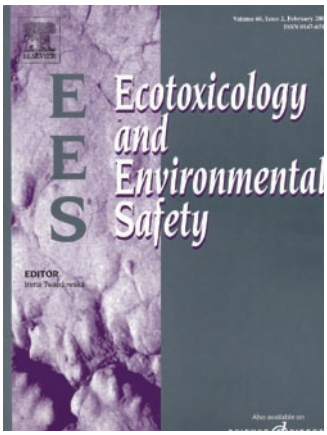
Jizhong Zhou, Distinguished R&D Staff Scientist in Microbial Genomics and Ecology at ORNL, was elected a fellow of the American Society for Microbiology (ASM). Induction was in June 2005 in Atlanta during the 105th General Meeting of ASM.

Zhou has authored more than 120 publications on microbial genomics, molecular biology, molecular evolution, microbial ecology, bioinformatics, bioremediation, and theoretical ecology, and the first book on microbial functional genomics in this emerging field. His election signifies recognition of his pioneering advances and leadership in developing and applying integrated genomics technologies to address research challenges in functional genomics of stress responses and energy metabolism in environmental microorganisms, and in understanding the diversity, structure, mechanisms, and dynamics of microbial communities and "extremophilic" (thriving in harsh environments) metal-reducing bacteria important to bioremediation, global changes, and carbon sequestration.

The American Academy of Microbiology is the honorific leadership group within ASM, the world's oldest life science organization. AAM is the only group of its kind devoted entirely to microbiologists and the science of microbiology. AAM fellows are elected through a highly selective, annual, peer-reviewed process, based on their records of scientific achievement and original contributions that have advanced microbiology (http://www.asm.org/Academy).

Vo-Dinh associate editor of Ecotoxicology and Environmental Safety

Tuan Vo-Dinh, an ORNL Corporate Fellow, has been named associate editor of *Ecotoxicology and Environmental Safety*. The journal focuses on integrated mechanistic research on short- and long-term pathways and interactions of substances and chemical mixtures in environmental compartments; on bioavailability, circulation, and assimilation in target organisms; on the biological response of organisms and damage mechanisms (endocrine disruption, genotoxicity); and on a further fate in the food chain, including humans. It also addresses novel technologies, techniques, and methods, such as biomedical photonic technologies; biomarkers, biosensors, and bioanalytical systems; quantitative structure property and activity relationships; and advanced high-performance computational methods, models, and storage systems and their applications in obtaining and processing interdisciplinary ecotoxicological information.

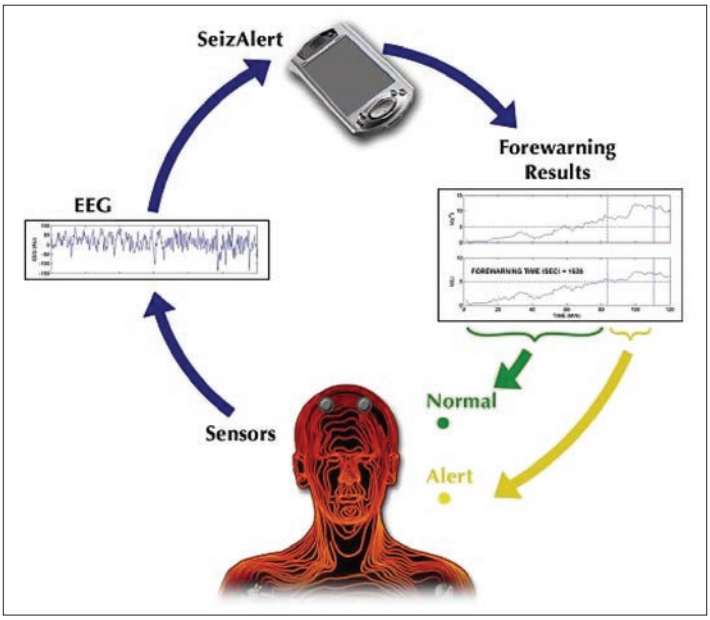


ORNL Research to appear in the Journal of Clinical Neurophysiology

An ORNL team, including Lee Hively and Vladimir Protopopescu, Computational Sciences and Engineering Division, and Nancy Munro, Life Sciences Division, will have an article describing their work on seizure forewarning published in the *Journal of Clinical Neurophysiology* in December 2005. The research was supported by DOE Basic Energy Sciences and by the ORNL Office of Technology Transfer.

Forewarning of epileptic seizures is the goal of several research groups in the United States and elsewhere. Most researchers use brain wave data from highly invasive intracranial electrodes. ORNL is one of the leaders in successful

forewarning from *noninvasive, scalp* EEG data. The article reports new developments in the ORNL approach via two-channel rather than previous single-channel analysis and an extended forewarning window of several hours. While still using customized parameters, the bipolar approach gave a total true rate above 90%, with a very low average rate of false positives.



The SeizAlert epilepsy forewarning device won an R&D100 Award in 2005.

The ORNL forewarning technology described in this paper has since been implemented on a personal digital assistant (PDA) device (SeizAlert) and was selected as a winner of a 2005 R&D 100 Award. SeizAlert is a prototype for a low-cost, compact, wearable device designed to alert the wearer and medical personnel of an impending epileptic seizure. In a real-life implementation, the alerting device would obtain EEG data from wireless sensors on the wearer's scalp and would analyze the data for interpretation and display in real time.

L. M. Hively, V. A. Protopopescu, and N. B. Munro. "Enhancements in epilepsy forewarning via phase-space dissimilarity," **J. Clin. Neurophysiol**. (accepted for publication, December 2005).

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