



INSIDE

- 3 Modeling the Spatial Dynamics of Plasmid Transfer
- 4 INBRE Magnet P.I. Research Highlights
- 8 Predictability of Viral Evolutions
- 9 NIC celebrates opening of Health and Sciences Building
- 10 Publications
- 11 INBRE Releases New Immunology Course
- 11 COBRE Renewal
- 12 Don't forget to cite COBRE/BRIN/INBRE



The people in the photo are: Erin O'Leary-Jepsen, Nino Chelidze, Jon Smith, Patty Retka and Sarah Hobdey (left to right) are viewing data from an Agilent Technologies Bioanalyzer, a microfluidics-based platform for the analysis of DNA, RNA, proteins and cells. The Agilent 2100 Bioanalyzer is the first commercial, analytical instrument based on lab-on-a-chip technology.

INBRE Support Proving to be Boon for Idaho State University

By Andrew Taylor

Pocatello – IDEa Network for Biomedical Research Excellence (INBRE) support has enabled Idaho State University (ISU) research activities to expand dramatically.

INBRE facilities at ISU, which include the Molecular Research Core Facility (MRCF) and the associated Bioinformatics Laboratory, have made it possible for ISU researchers to expand their studies of microbes in Antarctica; determine the evolutionary history, distribution and classification of the world's bamboos; and increased the study of infectious diseases including the presence of pathogenic *E. coli* in Idaho – to name just a few of the activities taking place.

The MRCF has experienced enormous growth in recent years due to increased usage of molecular tools by existing faculty and the arrival of six new tenure-track and one research faculty in the Department of Biological Sciences in fall 2004. They will be joined by two tenure-track, two research, and one visiting faculty next year. Nearly all of these positions come with significant molecular research expectations depending upon the MRCF; the expansion is designed to meet this growing demand.

The MRCF gives ISU researchers the ability to generate DNA gene sequencing reports and the Bioinformatics Laboratory, which features a cluster composed of 24 Apple X-serve dual processor G5 nodes, helps scientists analyze and interpret those reports. The Bioinformatics Laboratory is home to the ISU Evolutionary, Ecological and Environmental Genomics Group.

Besides funding research, INBRE support has been a boon to the training of both ISU undergraduate and graduate students and scientists and technicians throughout the region, and has fostered collaboration between ISU and the other state universities.

"We're investigating infectious diseases, the mutations of nucleic acids and developing computational methods for generating evolutionary trees of relationships among organisms," said ISU assistant biological sciences professor Dr. Scot Kelchner. "INBRE resources are allowing us to tackle a wide variety of projects."

This summer, Kelchner was awarded a four-year National Science Foundation



IDAHO RESEARCH



IDeAs Institutional Development Awards

Volume 2, Issue 1

A newsletter of the Idaho grantees of NIH NCRR Institutional Development Award (IDeA) Program.

The National Center for Research Resources (NCRR), under the guidance of the National Institutes of Health (NIH), has developed the Institutional Development Award (IDeA) Program to assist historically low funded states develop infrastructure that will enhance the state's ability to attain competitive research funding.

Idaho has received three IDeA awards:

P20 RR 015587 – “Molecular and Cellular Basis of Host-Pathogen Interactions,” Dr. Gregory Bohach, Principal Investigator (COBRE)

P20 RR 016454 – “IDeA Networks of Biomedical Research Excellence,” Dr. Carolyn Horde Bohach, Principal Investigator (INBRE)

P20 RR 16448 – “Center for Research on Evolutionary Progress,” Dr. Larry Forney, Principal Investigator (COBREII)

This is a biannual newsletter of the Idaho INBRE, COBRE and COBREII awards. The purpose of this newsletter is to broadcast the programs and activities of the grant participants, and to notify faculty, staff and students of upcoming biomedical activities across the state of Idaho. The goal is to increase awareness of the broad spectrum of biomedical research being performed across the state, and to promote cooperation and collaboration of Idaho researchers.

Comments or contributions may be directed to our online form at:
inbre@uidaho.edu

Or to:

Idaho INBRE Program
University of Idaho
Mail Stop 444207
Moscow, ID 83844-4207

208/885-5373 • e-mail: inbre@uidaho.edu
www.uidaho.edu/inbre



*Michael Thomas, Ph.D. and Loubin Yang M.S.
Bioinformatics Laboratory, ISU*

collaborative grant to provide data coordination, phylogenetic analysis and bioinformatics resources for an international team that will resolve the evolutionary relationships and classification of the world's bamboos. The Apple X-serve cluster of ISU's Evolutionary, Ecological and Environmental Genomics Group (EGG) in the Bioinformatics Laboratory is an integral part of this research, and provides database capabilities and analytical power to the 20 researchers from 10 different countries involved in the Bamboo Phylogeny Group project.

Kelchner also is using the MRCF's computational power to investigate new techniques for determining the evolutionary development of organisms using computer simulation experiments. He and fellow ISU MRCF researchers also are studying the molecular evolution in strains of *E. coli* bacteria that potentially can cause illness in humans.

Peter Sheridan, assistant professor of biological sciences, is another beneficiary of INBRE support. Among other projects, he has been using the MRCF to study the molecular microbiology and biochemistry of microbes living in extreme environments, particularly in Antarctica.

“We're looking at the microbial diversity in extreme environments, trying to determine how these organisms are able to not only grow, but to thrive, in a climate with extremely low temperatures,” Sheridan said. “If an organism can grow extremely quickly at a low temperature it means there is an enzyme that has to work quickly at a low temperatures and there could be a practical application of that.”

He said, for example, most laundry detergents have enzymes that allow them to work at low water temperatures. Finding enzymes that work efficiently at a low temperature without producing a large amount

of waste products would be a boon to the detergent industry.

The research activities of both Kelchner and Sheridan have been greatly helped by the efforts of ISU assistant professor Mike Thomas, who oversaw the University's efforts to set up the Bioinformatics Laboratory. This year, Thomas, along with Dr. Majorie Matocq, MRCF director, were recipients of a \$140,000 National Science Foundation Grant for “major research instrumentation” that enabled ISU to purchase a new DNA sequencer for the MRCF and additional robotics to allow a faster “throughput” of the DNA sequencing. The grant also allowed the purchase of additional computers to add to

the Bioinformatics Laboratory's cluster of computers to process data and increase the overall computational power of the entire system.

“There is a heavy use of the MRCF for sequencing genes and the core facility is generating huge amounts of data,” noted Sheridan. “It has the capacity to produce whole genomes of organisms, and the Bioinformatics Laboratory, with its computing power, helps interpret those results.”

The MRCF has developed classes to teach students, as well as scientists and technicians from other universities and institutions, how to use and interpret the MRCF's equipment.

“A lot of our biological sciences classes have bioinformatics and molecular biology components and with the MRCF, we're able to train our students on how to do research in the 21st century,” Sheridan said. “It's a great opportunity for students and it is the training that is now almost expected from graduates who want to work in a variety of biological fields.”

The MRCF also is one component of INBRE's statewide program, which involves different missions for the state's universities.

“The INBRE program fosters a lot of collaboration and cross-talk between Boise State University, the University of Idaho, ISU and Idaho's other four-year colleges,” said Sheridan. “It has really developed a scientific cohesiveness between the state's major universities. Idaho has been one of the more successful INBRE programs in the country, and we're also developing training opportunities for scientists outside of the university system.”

(Released by University Relations, 14 November 2005, Contact: Dr. Chris Daniels, (208) 282-2682 or 282-4008)



Stephen Krone, Ph.D. Mathematics Department, UI



Laboratory cultures of bacteria containing antibiotic resistance plasmids (pink) and without plasmids (white).



Eva Top, Ph.D. Biological Sciences Department, UI

Modeling the Spatial Dynamics of Plasmid Transfer

By Eva Top and Steve Krone

Steve Krone (Mathematics) and Eva Top (Biological Sciences) were recently awarded a five-year, \$1.3 million NIH grant to study one of the key mechanisms leading to the rapid spread of antibiotic resistance in bacteria. The study focuses on plasmids—relatively small circular strings of DNA that reside in many bacterial cells but are not part of the cell’s chromosome. An amazing feature of plasmids is that, in addition to being passed from mother to daughter cell upon cell division, along with the cell’s chromosomal DNA, these extra-chromosomal packages of genetic material can be exchanged between different cells—even between very different species of bacteria. The genes encoded on plasmids can impart to their bacterial host the ability to adapt quickly to harsh environmental conditions such as the presence of toxic chemicals or antibiotics.

Given that most treatments with antimicrobial agents are now severely compromised due to the rapid increase in the antibiotic resistance levels of pathogens, antibiotic resistance plasmids can have serious human health consequences; thus the interest from the National Institutes of Health.

Krone and Top, who also are active participants in UI’s Initiative for Bioinformatics

and Evolutionary Studies (IBEST), will collaborate on a joint theoretical and experimental investigation into the role of spatial structure in the spread and persistence of self-transmissible antibiotic resistance plasmids. This research deals with the spread of plasmids in a population of bacteria and, unlike most previous studies, takes into account the fact that the predominant mode of growth in



Patterns of plasmid loss and competition between plasmid-containing (pink) and plasmid-free (white) cells of *E. coli* on agar surface. The multiple antibiotic resistance plasmid pB10 was marked with a gene encoding a red fluorescent protein.

microbial communities is one involving spatial structure. Indeed, unlike the free-swimming bacteria found in test tubes, most bacteria in natural and clinical settings are attached to surfaces—e.g., on medical implants and the walls of intestines—and form complex communities such as biofilms. It is by now fairly well established, both experimentally and through mathematical models, that spatial structure can have profound effects on

the ecological and evolutionary dynamics of biological populations.

The long-term goal of this study is thus to understand the population biology of self-transmissible antibiotic resistance plasmids in spatially structured microbial communities. What are the mechanisms that drive the horizontal transfer and persistence of these mobile genetic elements in bacterial communities? Why do they persist over long periods of time when, in the absence of antibiotics, they impose a burden on the cells that carry them? How does the spatial structure of natural microbial communities influence the ecological and evolutionary dynamics of plasmid-bacteria interactions?

The specific aims of this proposal are to construct 2-dimensional and 3-dimensional stochastic cellular automata (CA) models that can be used to accurately predict the spread and persistence of natural antibiotic resistance plasmids in bacterial colonies growing on agar surfaces and in biofilms in the lab. These mathematical models describe the microbial community down to the individual cell level.

The project supports three graduate students (Renie Lu and Grant Guan, mathematics Ph.D. students who are participating in the Bioinformatics and Computational Biology degree program, and Randal Fox, an M.S. student in Microbiology, Molecular Biology and Biochemistry and working in Dr. Top’s lab), a postdoctoral researcher who is soon to arrive on campus, and the two faculty investigators, Krone and Top.

INBRE Magnet P.I. RESEARCH HIGHLIGHTS

The Role of INBRE Supported Magnet Principal Investigators

The INBRE Program supports faculty at primarily undergraduate colleges to conduct biomedical research and train undergraduate students in research methodology. A select subset of these faculty are provided additional resources in the form of release time from teaching and in support of the lab. The role of these “Magnet Principal Investigators” is, as the name implies, to serve as magnets for other faculty and students to do research. The magnet PI’s are to serve as research role models at their college, and collaborate with other faculty and provide opportunities for students to work in their lab. Magnet PI’s are on track to become among the first independent funded NIH investigators at their college.



Arthur Ayers, Ph.D.

Professor of Biology
Biology Department
Albertson College of Idaho

Research Focus: *Biochemistry
and Molecular Biology of the
Extracellular Matrix*

INBRE Project Title:
*Molecular Interactions of the
Pericellular Matrix*

Dr. Arthur Ayers grew up in San Diego and attended the University of California at San Diego. As a graduate student at the University of Colorado-Boulder (MCDB), he determined the structure of the B-1,3-1,6 glucan family of fungal elicitors of plant phytoalexins. He held postdoctoral research positions at the Swedish Forest Products Research Laboratories (discovery, purification and characterization of cellobiose dehydrogenase, an enzyme with both heme and flavo electron transport prosthetic groups), at the University of Missouri, Columbia (immunological characterization of the fireblight toxin), and at Kansas State University (regeneration of plants from single potato leaf cells). Ayers was an assistant professor at Harvard University (CDB), where he studied the molecular basis of plant disease resistance using monoclonal antibodies and he was director of the Genetic Engineering Program at Cedar Crest College in Allentown, Penn. Ayers joined the faculty at Albertson College of Idaho in 1991 and has directed and supported student

research in subjects spanning host-pathogen interactions, molecular taxonomy, phage display, bioinformatics and protein modeling. Ayers became a Magnet P.I. with INBRE in 2004, and currently is working with expression (cDNA-PCR) and deletion (RNA interference) of the heparan sulfate proteoglycans of cartilage.

As part of his INBRE project, Ayers uses cartilage secreting cell lines, chondrocytes, in combination with fluorescence microscopy to identify and locate specific proteins in the secreted cartilage. Specifically, he examines interactions of the proteins and polysaccharides that make up the extracellular matrix (ECM) in the form of cartilage, bone, connective tissue and the other external cellular materials that determine the structure of organisms and how cells interact. The ECM is a major determinant in host-pathogen interactions, degenerative diseases, aging, cancer metastasis and cellular regenerative therapy. His lab uses analytical techniques, such as 2D-gel electrophoresis/western blots, ELISA, fluorescence immunomicroscopy, chondrocyte cell culture and molecular techniques (such as RNA interference) supported by bioinformatics and protein modeling of molecular interactions to explore the dynamic nature of the ECM.

Ayers is particularly interested in how heparan sulfate proteoglycans (HSPGs) are involved in secretion, function of growth factors and protein uptake. Heparan sulfates are the most acidic polysaccharides produced by cells, and the majority of proteins have binding sites for heparan sulfate polysaccharides or

oligosaccharide fragments, heparin sulfate (HS). Secretion of the ECM involves the tight packing of proteins and polysaccharides in an inactive, non-polymerizing form with charge neutralization. HSPGs and bound, positively charged polyamines, are essential for secretion of the ECM, but the process is poorly understood. Subsequent to secretion, most of the HSPGs and polyamines are rapidly recycled into the cell. The more permanent components of the ECM swell, polymerize into aggregates or fibers and hydrate into a rigid gel. In the case of chondrocytes, the dense gel is cartilage with reinforcing collagen fibers.

Recently, Ayers has been studying HSPGs by following the expression of a dozen genes that code for the proteins and the enzymes synthesizing the polysaccharide components of HSPGs. The patterns of gene expression reveal multiple stages in chondrocyte development in cell culture that parallel the alterations in the morphology of cells seen by microscopy. These studies, performed by cDNA-PCR are now being extended and complemented with RNA interference.

Another research goal is to pursue proteins that bind to HSPGs as the HSPGs are drawn back into the cell by a system parallel to endocytosis. There is evidence that a class of HS-binding proteins of the ECM returns to the cell cytoplasm and may have an impact on transcription. Ayers is attempting to follow this process using phage display to generate peptides that mimic HS-binding domains.



Peter Craig, Ph.D.
Assistant Professor of
Chemistry
Chemistry Department
Albertson College of Idaho

*Research Focus: Bioinorganic
Chemistry*

*INBRE Project Title: Cadmium
Chelation Therapy: Development
of New Agents to Prevent/Treat
Heavy Metal Poisoning*

Dr. Peter Craig completed undergraduate degrees in science and business before earning his Ph.D. in bioinorganic chemistry at the University of Auckland in New Zealand. He was a postdoctoral researcher at the University of Colorado at Boulder before joining the faculty at Albertson College of Idaho in 2001.

Craig realized the power of using research as a teaching tool and uses it in his organic and inorganic chemistry laboratory courses at Albertson College of Idaho. This “teaching through research” philosophy is a prominent feature of his research group that includes nine undergraduate students. His research has environmental and biological significance, and appeals to students majoring in chemistry, biology, health sciences or business.

Craig’s research group designs, synthesizes

and studies selective cadmium-chelating agents. Cadmium persists in the environment from smoking, mining and electronic waste. A recent report released by the Center for Disease Control indicates approximately 5 percent of Americans 20 years and older have urinary cadmium consistent with kidney dysfunction and low bone density.

The objectives of Craig’s research include assessment of reagents to (1) remove cadmium from the environment, and (2) treat human cadmium exposure. Both require selective removal of cadmium from environments that may contain numerous other and often essential ionic and molecular species. There are four parts to the project:

Part 1: Agent Development: Craig is developing sulfur containing chelating agents to selectively bind Cd²⁺ (the predominant soluble form of cadmium). The agents also comprise components designed to balance hydrophilic and lipophilic factors essential to the effective chelation of Cd²⁺.

Part 2: Biological Screening: The cytotoxicity of Cd²⁺ chelating agents is being evaluated by measuring loss of cell adherence, trypan blue exclusion, and MTT with a human osteosarcoma cell line, Saos-2 (through collaboration with the research group of Dr. Sara Hegglund). This methodology involves examining the cytotoxicity of (1) the chelating

agent, (2) the Cd²⁺ complex of the chelating agent, and (3) the chelating agent and Cd²⁺ in competition. Testing the chelating agent with and without Cd²⁺ will provide cell-signaling information linked to Cd²⁺ toxicity and lead to the development of better therapies for Cd²⁺ poisoning.

Part 3: Analytical Screening: Assays to assess the dissolution and dissociation equilibria exhibited by Cd²⁺ complexes of chelating agents are being developed using atomic absorption spectroscopy (AAS) and electrochemistry (ISE/conductivity). Craig plans to measure both the efficiency of the uptake of Cd²⁺ from aqueous solution, and the selectivity that agents exhibit for Cd²⁺ in the presence of competing cations.

Part 4: Proteomics: Craig will evaluate the effect of the presence of Cd²⁺ and chelating agents on model cell line (such as Saos-2) proteomes. This involves two-dimensional gel electrophoresis analyses of cells in the presence and absence of Cd²⁺ and/or the chelating agents. He also will identify modified, up-and-down regulated proteins using mass spectrometry. This information will provide insight into the mechanism of Cd²⁺ induced cell death and differential Cd²⁺ chelation mechanisms within the model cell lines.



**Dr. Sarah Hegglund,
Ph.D.**
Associate Professor of Biology
Biological Sciences
Department
Albertson College of Idaho

*Research Focus: Cell Biology
and Toxicology*

*INBRE Project Title:
Investigation Differential Cell
Sensitivity to Cadmium and
Cadmium-Sequestering Molecules*

Dr. Sarah Hegglund earned her Ph.D. in reproductive physiology and endocrinology from Kent State University. Since then, she has devoted her career to teaching and research at private liberal arts colleges.

Her decision to pursue a career in undergraduate liberal arts education was heavily influenced by her experience doing undergraduate research at the University of Minnesota. She views undergraduate research as an extension of the classroom and finds it very rewarding to mentor students through the entire scientific process.

Hegglund believes doing research builds self-confidence in students, as well

as encourages analytical thinking, increases competency in scientific writing, and creates an excitement about learning biology. She is very proud of the students who graduate from her lab. Many of these students go on to graduate school, professional schools, and one former student even returned to her lab as a research technician.

When she is not teaching, mentoring students and coming up with inspiring scientific research, or out discovering new things about cadmium toxicology, she spends time backpacking with her husband, Erik, and their Rhodesian ridgebacks.

Hegglund’s INBRE research explores the cellular mechanisms involved in heavy metal toxicity and focuses on the heavy metal cadmium. There are a variety of sources of cadmium, however, increasing discard into landfills of electronic products (e-waste) that contain heavy metals makes cadmium exposure a growing public health concern. Cadmium is an environmental pollutant that is toxic to many tissues. Human exposure to cadmium is linked to many diseases including kidney, skeletal and liver disease, and several types of cancer. A key to understanding cadmium’s toxic action is to decipher the mechanisms within cells that cause and protect against cadmium toxicity.

Currently, Hegglund research encompasses three ongoing projects. First, a project that explores the molecular signaling cascades involved in cadmium-induced cell death in bone and the relationship to bone diseases, such as osteoporosis. She is specifically interested in the role of cadmium-induced apoptosis in osteoblasts. One exciting new avenue of this project includes examining cadmium-induced epigenetic changes that may alter osteoblast development. The overarching goal of this project is to uncover how cadmium disrupts the molecular regulators of bone formation that ultimately lead to bone disease.

A second project involves developing an in vitro model using cells lines (human and fish) that exhibit differential sensitivity to cadmium to study cellular mechanisms involved in cadmium resistance and toxicity. This approach seeks to identify alternative models in toxicology that might provide insight into cadmium’s role in human health.

The third project is a collaboration with Dr. Peter Craig in the Albertson College Chemistry Department. Using the cell line model, they are screening the cytotoxicity of cadmium-sequestering molecules synthesized in his laboratory that may have promise for human or environmental applications.

INBRE Magnet P.I. RESEARCH HIGHLIGHTS



Cheryl L. Jorcyk, Ph.D.
Associate Professor of Biology
Biology Department
Boise State University

Research Focus: *Cancer Research*

INBRE Research Title:
Oncostatin M Induces VEGF in Human Breast Carcinoma Cells: Stimulation of Angiogenesis in vitro and in vivo

“OSM, an IL-6 family cytokine, is produced by breast cancer cells and tumor-associated cells of the immune system, including activated T-cells, macrophages and neutrophils. Recent studies and interesting data from our lab suggest that OSM could actually contribute to tumor progression, angiogenesis, and the development of a metastatic state.”

— *Dr. Cheryl Jorcyk*

Dr. Cheryl Jorcyk earned her B.S. in Biology from Pennsylvania State College and her Ph.D. in Biology from Johns Hopkins University. After completing a five year postdoctoral fellowship at the National Cancer Institute, she joined the Boise State University Biology Department and now is a tenured associate professor.

Jorcyk’s INBRE research involves Oncostatin M (OSM), a pleiotropic cytokine in the interleukin (IL)-6 superfamily released during the inflammatory process. Her data demonstrates that OSM will stimulate breast cancer cells to produce vascular endothelial growth factor (VEGF), a potent pro-angiogenic factor. She hypothesizes that VEGF produced by OSM-treated breast cancer cells will stimulate angiogenesis in vitro and in vivo and therefore promote tumor progression.

In order to test her hypothesis, she proposes the following specific aims: demonstrate that neutrophil-derived OSM will induce VEGF from breast cancer cells in a paracrine fashion; identify and characterize the signal that stimulates neutrophils to release OSM; and investigate the ability of OSM-induced VEGF to stimulate angiogenesis and promote breast carcinoma progression in vivo. Findings that support this hypothesis may provide a rationale for the development of therapies that inhibit OSM expression, function or signaling.

During her five year postdoctoral research at the National Cancer Institute, Jorcyk “...established cell lines from a transgenic mouse where the males would develop prostate cancer and the females would develop mammary cancer.” She brought these cell lines and her interest in cancer progression with her to Boise State in 1997, where she began her own research on prostate and breast cancer.

During this time, fellow researchers at the Boise Veteran’s Administration were investigating the effects of the cytokine OSM in breast cancer. One theory suggested that OSM would inhibit the proliferation of breast cancer cells; and this initially focused much attention on OSM as a potential breast cancer therapy. However, a contradictory theory by

collaborator Dr. Randy Ryan hypothesized just the opposite. It was this theory that intrigued Jorcyk.

“OSM, an IL-6 family cytokine, is produced by breast cancer cells and tumor-associated cells of the immune system, including activated T-cells, macrophages and neutrophils. Recent studies and interesting data from our lab suggest that OSM could actually contribute to tumor progression, angiogenesis, and the development of a metastatic state.”

Tumor-associated and tumor-infiltrating neutrophils and macrophages can account for as much as 50 percent of the total tumor mass in some types of breast cancer. It is thought that tumors secrete factors that elicit a wound-repair response from the infiltrating macrophages and neutrophils and that this response inadvertently stimulates tumor progression.

Jorcyk explains, “Our lab has shown that human neutrophils isolated from whole blood or breast cancer cells alone express little OSM, but neutrophils express and release high levels of OSM when they are co-cultured with breast cancer cells. We believe that breast cancer cells secrete granulocyte-macrophage colony-stimulating factor (GM-CSF) to signal neutrophils to produce and release OSM. This OSM then binds to the OSMR present on the tumor cells to signal downstream effects.”

Jorcyk’s work also has shown that neutrophil-derived OSM induces the pro-angiogenic factor VEGF from breast cancer cells in co-culture. Importantly, in addition to VEGF induction, neutrophil-derived OSM increases breast cancer cell detachment and invasive capacity, suggesting that neutrophils and OSM may promote tumor progression in vivo.

Research conducted by Jorcyk and her lab recently appeared in the Oct. 1, 2005 issue of *Cancer Research*. The study could someday lead to the development of new drugs to treat the deadly disease.

**Troy Rohn, Ph.D.**

Associate Professor of Biology
Biology Department
Boise State University

Research Focus: *Alzheimer's disease, Parkinson's and Dementia with Lewy Bodies*

INBRE Project Title:

Involvement of Astrocyte Caspase Activation and CD40/CD40L Signaling Interactions in Alzheimer's Disease

Dr. Troy Rohn earned his B.S. in Physiology from the University of California-Davis, and his Ph.D. in Pharmacology from Washington State University. After receiving his Ph.D., Rohn completed three postdoctoral fellowships.

Rohn's first postdoctoral fellowship took him to Paris, France, where he worked for two years at INSERM. In 1997, he began his second fellowship at Montana State University where he worked on free radical biology, and in 1998 he returned to California to complete his third and final postdoctoral fellowship at UC Irvine with Dr. Carl Cotman who inspired his interest in neuroscience and Alzheimer's research.

In 2000, Troy and wife, Lisa, made the move to Boise to accept a tenure track faculty position at Boise State. Today, as an associate professor and one of Boise State's INBRE

Magnet P.I.'s, Rohn continues his research.

The primary focus of Rohn's INBRE research involves neurodegenerative diseases including to a large extent, Alzheimer's disease (AD). His lab, as well as many others, is convinced that the primary cause of cell death (neuronal or glial) associated with AD is caused by neurons undergoing apoptosis. Biochemical markers are being developed that are designed to follow specific cleavage products produced from caspases. In this way, they can use these markers as footprints to the contribution of apoptosis in certain neurodegenerative diseases including AD. Both cell lines and primary cultures of neurons and glial cells will be used to help characterize these markers.

A central tenet underlying the pathology observed in AD is the strong contributory role of inflammatory mediators released from cells such as astrocytes. Astrocytes are glial cells present in the CNS that participate in the formation of the blood-brain barrier and regulation of the internal environment. In AD, the close association of activated astrocytes with neuronal degeneration suggests that astrocytes may have a significant role in progression of the pathology observed in AD. The goal of Rohn's research is to examine if caspase activation occurs in reactive astrocytes of the AD brain and whether such activation is associated with specific markers of inflammation.

Rohn's lab has successfully designed a caspase-cleavage site-directed antibody to

GFAP. Following examination of the amino acid sequence of GFAP, they identified one potential caspase-cleavage consensus site, DLTD266. A peptide was generated to the downstream sequence (AAARNAE) that would be revealed following cleavage of GFAP by caspases, injected into rabbits, and the antibody of interest was affinity-purified. Following characterization of the antibody in vitro, the antibody was tested in vivo using tissue sections from AD or aged-matched controls.

"Using the GFAP-caspase cleavage product antibody, we found widespread astrocytic labeling within plaque regions and along blood vessels. Little immunoreactivity was observed in aged-matched controls. Overall, application of this antibody labeled numerous astrocytes that exhibit all of the hallmark characteristics of apoptosis including condensed nuclei, fragmented processes as well as processes that were swollen and beaded. Taken together, these results suggest a role for caspase activation and cleavage of GFAP within astrocytes of the AD brain and may provide for a hypothesis to explain the compromised blood-brain-barrier that has been associated with this disease," says Rohn.

A paper describing these findings is currently in press in *American Journal of Pathology*.

**Henry Charlier Jr., Ph.D.**

Assistant Professor
Department of Chemistry
Boise State University

Research Focus: *Enzymology and Biochemistry*

INBRE Project Title:

Structure/Function Analysis of Anthracycline Reduction by Carbonyl Reductase

Dr. Henry Charlier Jr. completed his Ph.D. in Biochemistry at the Medical College of Wisconsin, Milwaukee. He continued his research with a postdoctoral fellowship at the University of Iowa, and in 2000, began his career at Boise State University. Henry currently holds the title of assistant Professor.

Since joining the Boise State University Chemistry department in 2000, Charlier has continued to combine his two passions — research and teaching. He describes himself as a researcher and a teacher, or a teacher and a researcher. "I really can't separate the two,"

says Charlier.

He knew he wanted to be a scientist since the third grade and as an undergraduate in college, he decided the best path was a double major in biology and chemistry. The influence of "some outstanding professors" made Charlier realize that for him, research and teaching would always be intertwined. All through graduate school and postdoctoral work, he never lost sight of the concept that doing research would be the best teaching tool at his command.

Charlier's INBRE research is centered on the enzyme carbonyl reductase (CR). CR catalyzes the NADPH dependent reduction of a variety of carbonyls. "Given its wide substrate specificity, its complete physiological role is not known," says Charlier.

CR is known to participate in the metabolism of prostaglandins and is postulated to play a role in quinone detoxification. Studies have connected its biological activity to cancer progression, anthracycline efficacy and associated cardiotoxicity. There also are some potential links to several neurological diseases

as well. Given the breadth of CR participation in a large number of important processes, it is very important to define both its catalytic and kinetic mechanisms, including fully mapping its substrate and inhibitor specificities.

In order to understand both the catalytic and kinetic mechanisms of CR, Charlier's lab uses a variety of techniques to assess the steady state kinetics, transient kinetics of substrate binding and the binding affinity of a variety of small molecules for CR.

"One outcome of our studies is that to date we have discovered 12 novel inhibitors of CR, many of which may find clinical use in improving anthracycline therapy. We have also discovered a new class of substrate for CR and these are helping to expand our understanding of how CR recognizes the molecules to which it binds. It is the goal of our lab to use the information from our studies to more completely understand the physiological functions of CR and to reduce the risk of anthracycline cardiotoxicity associated with CR activity."



Jennifer Chase, Ph.D.

Chair and Professor of Chemistry
Chemistry Department
Northwest Nazarene University

Research Focus: *Metabolic control analysis of enzyme pathways*

INBRE Project Title: *Distribution of Flux Control between ADH and ALDH in Liver Ethanol Metabolism*

Dr. Jennifer Chase was raised in Massachusetts and Arizona and received her B.A. in Biology/Chemistry from Point Loma Nazarene College in 1990. Her undergraduate research experience, on the genetics of bacteriophage Epsilon 15, led her to Yale University's Ph.D. program of Molecular Biophysics and Biochemistry with the expectation of studying genetics. As her studies progressed, she did not perform a rotation in a genetics lab as originally planned; instead she completed three biophysics projects.

She followed her new research interest and began research in the group of R. G. Shulman, a physicist-turned-biophysicist who uses in vivo NMR spectroscopy to study a variety of types of muscle and brain metabolism, particularly the dysfunctional metabolism of diabetes.

Chase's graduate research ultimately focused on quantitation of flux control in the muscle glycogen synthesis pathway in rat gastrocnemius using carbon and phosphorus NMR spectroscopy of live rats.

During her years of graduate school, Chase was the lay youth pastor at her local church and became involved in NYI Bible Quizzing for the Church of the Nazarene.

Chase earned her Ph.D. in 1998 and joined Northwest Nazarene University in 1996. She currently holds the position of chair of the Department of Chemistry. She continues to be passionate about ministry to teens and currently serves as the Quiz Director for the InterMountain District of the Church of the Nazarene. She also serves in the Treasure Valley Science-and-Religion Institute, is a voracious reader, an avid board game player, and plays at volleyball and biking. In 2002-03, she received the Eisenhower Award from the Idaho State Department of Education for "Enhanced incorporation of Molecular Biology Techniques into the Idaho High School Science Classroom." Even with her

academic responsibilities and personal pursuits, Chase continues to find the understanding of metabolic control of metabolic pathways to be an ever-engaging academic pursuit.

Chase's INBRE research involves the study of how the consumption of beverage alcohol (ethanol) by humans can disrupt many normal metabolic processes. The cells of liver and other tissues must divert processing enzymes from normal functions to process ethanol, thus reducing the production of important compounds such as the vitamin A (retinol) derivative, retinoic acid. It has been hypothesized that the disruption in synthesis of retinoic acid by ethanol is an underlying cause of fetal alcohol syndrome. The main enzyme responsible for retinoic acid synthesis is alcohol dehydrogenase IV (ADH-IV), most abundant in the stomach and intestines of adults, and essential for proper fetal development.

A second well-recognized impact of ethanol consumption is a decrease in the NAD⁺/NADH ratio, especially in the liver. Because dissociation of NADH is the rate-limiting step in the oxidation of retinol, increases in the level of NADH in the presence of ethanol also could contribute to the decrease in retinoic acid production.

Ultimately, Chase's goal is to develop a computer simulation model of the interactions of ethanol on the liver NAD⁺/NADH ratio and its impact on the retinol oxidation apparatus of the cell. In a preliminary simulation using a model consisting of just ADH-IV and a "NADH oxidase" to reform NAD⁺, she found that the control of the rate of retinol oxidation is distributed between ADH-IV and the "NADH oxidase," with more control by "NADH oxidase" at higher ethanol levels. Thus, quantifying the importance of the cellular capacity to reoxidize NADH as a potential contributor to fetal alcohol syndrome.

Validation of the model is utilizing a human ADH-IV clone. She would like to gain an understanding of the control of all the reaction pathways affecting NADH levels and retinol oxidation. Such information might allow medical professionals to predict who is most likely to be adversely affected by consumption of beverage alcohol and might explain differences in response that currently confound studies on the causes of fetal alcohol syndrome and other alcohol-related diseases.

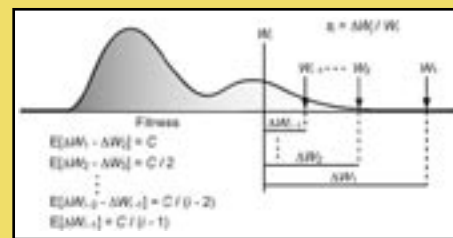


Figure X. The Gillespie-Orr Model: A schematic depiction of the extreme value theory predictions for a single step in adaptive evolution.

RESEARCH SNAP SHOT:

COBRE Research Project Title:

Predictability of Viral Evolution.

Holly Wichman, Ph.D., COBRE Research Project P.I., Professor of Zoology, Department of Biological Sciences, University of Idaho

An empirical test of the mutational landscape model of adaptation using a ssDNA virus.

Description – Using an empirical virus model, we provided the first test of a general model that describes a single step in adaptation. The spectrum of mutations fixed as the first step in adaptation fits the expected array of fitness effects, but the theory requires a simple adjustment for unequal mutation rates. This theory may thus have considerable utility, both for a general understanding of adaptation as well as for applications to the evolution of drug resistance and carcinogenesis.

Synergy – This project is a collaboration between biologists (Wichman and her student Rokyta) and mathematicians (Joyce and his student Biesel).

Deliverables – This research project has already resulted in one publication that was highlighted in a News & Views In Nature Genetics. A grant proposal, titled *Patterns of Adaptive Evolution*, on which Joyce is PI and Wichman the Co-PI ranked in the 6.6 percentile and was funded by the National Institutes of Health for \$1.4 million.



NIC celebrates opening of Health and Sciences Building

By Stacy Hudson

Former Idaho Gov. Dirk Kempthorne and approximately 300 community members helped celebrate the grand opening of the long-awaited North Idaho College Health and Sciences Building Sept. 29.

Construction of the facility kicked off in March 2004, and the building opened with the start of fall semester classes in August.

The \$11.9 million NIC Health and Sciences Building was realized when NIC was one of seven colleges and universities across the state to benefit from the passage of facilities bond legislation in 2003 for new campus buildings.

Although the state of Idaho provided funds for the construction of the NIC Health and Sciences Building, the allocated funds only covered the cost of site preparation, construction, fixed equipment and the basic infrastructure.

NIC and the nonprofit NIC Foundation were responsible for completing the building by funding the latest instructional equipment in the labs and classrooms. A capital fundraising campaign was announced in July 2004 with the initial goal of raising \$2 million, including \$1 million for new technology and equipment and another \$1 million for new endowed scholarships for NIC students. More than 360 donors have contributed to the campaign, bringing the total to more than \$3 million.

With the addition of the new Health and Sciences Building to campus, the college's ability to provide enhanced instruction through state-of-the-art technology in the Health Professions and Nursing and Life Sciences programs has improved significantly.

The opening of the 57,000-square-foot, two-story building increased NIC's general classroom capacity by 25 percent. The



Top, Health and Science Building at dusk. Bottom, NIC Biology Instructor Peter Zao lectures while writing notes to a tablet PC that projects the slide and his notes instantaneously to a large screen in the classroom of the new Health and Sciences Building

state-of-the-art building includes 10 general use classrooms, eight labs, a theater-style auditorium with seating for 100, a smaller auditorium with seating for 60 and two video broadcast classrooms.

New Publications

Dr. Zaid Abdo, Dr. Larry Forney and Dr. Paul Joyce

Department of Biological Sciences and Department of Mathematics, University of Idaho

Abdo, Zaid, Schüette, Ursel M. E., Bent, Stephen J., Williams, Christopher J., Forney, Larry J., and Paul Joyce. (2006). Statistical methods for characterizing diversity of microbial communities by analysis of terminal restriction fragment length polymorphisms of 16S rRNA genes. *Environmental Microbiology*. 8 (5): 929.

Dr. Carolyn J. Hovde Bohach

Department of Microbiology, Molecular Biology and Biochemistry, University of Idaho

Davis, Margaret A., Rice, Daniel H., Sheng, Haiqing, Hancock, Dale D., Besser, Thomas E., Cobbold, Rowland, and Carolyn J. Hovde. (2006). Comparison of Cultures from Rectoanal-Junction Mucosal Swabs and Feces for Detection of *Escherichia coli* O157 in Dairy Heifers. *Appl. Environ. Microbiol.* 72: 3766-3770.

Dobbin, Heather S., Hovde, Carolyn J., Williams, Christopher J., Scott A. Minnich. (2006). The *Escherichia coli* O157 Flagellar Regulatory Gene *flhC* and Not the Flagellin Gene *fljC* Impacts Colonization of Cattle. *Infect. Immun.* 74: 2894-2905.

Ferens, Witold A., Cobbold, Rowland, and Carolyn J. Hovde. (2006). Intestinal Shiga Toxin-Producing *Escherichia coli* Bacteria Mitigate Bovine Leukemia Virus Infection in Experimentally Infected Sheep. *Infect. Immun.* 74: 2906-2916.

Kudva, Indira T., Krastins, Bryan, Sheng, Haiqing, Griffin, Robert W., Sarracino, David A., Tarr, Phillip I., Hovde, Carolyn J., Calderwood, Stephen B., and John, Manohar. (2006). Proteomics-based expression library screening (PELS) - a novel method for rapidly defining microbial immunoproteomes. *Mol Cell Proteomics*. T600013-MCP200.

Dr. Alok Bhushan and Dr. James C.K. Lai

Pharmaceutical Sciences, Idaho State University

Puli, Shilpa, Lai, James C.K., and Alok Bhushan. (2006). Inhibition of matrix degrading enzymes and invasion in human glioblastoma (U87MG) Cells by isoflavones. *Journal of Neuro-Oncology*. 1-8. DOI 10.1007/s11060-006-9126-0.

Dr. Leslie L. Devaud

Department of Pharmaceutical Sciences, Idaho State University

Kristine M. Wiren, Joel G. Hashimoto, Paul E. Alele, Leslie L. Devaud, Kimber L. Price, Lawrence D. Middaugh, Kathleen A. Grant, and Deborah A. Finn. (2006). Impact of Sex: Determination of Alcohol Neuroadaptation and Reinforcement. *Alcoholism: Clinical and Experimental Research*. 30:2 233.

Dr. Elizabeth A. Fortunato

Department of Microbiology, Molecular Biology and Biochemistry, University of Idaho

Rosenke, Kyle, Samuel, Melanie A., McDowell, Eric T., Toerne, Melissa A. Toerne and Elizabeth A. Fortunato. (2006). An intact sequence-specific DNA-binding domain is required for human cytomegalovirus-mediated sequestration of p53 and may promote in vivo binding to the viral genome during infection. *Virology*. 348 (1): 19-34.

Dr. Sara J. Heggland

Department of Biology, Albertson College of Idaho

Coonse, Kendra, Coonts, Allyson, Morrison, Elizabeth, and Sara J. Heggland. (2006). Cadmium induces apoptosis in the osteoblast-like cell line Saos-2. *The Journal of Toxicology and Environmental Health*. (In Press).

Dr. Sara J. Heggland and Dr. Peter R. Craig

Department of Biology and Department of Chemistry, Albertson College of Idaho

Zhukalin Mikhail, Blanksma, Megan K., Silva, T. Dimitri, Suyehira, Samuel W., Harvey, Wendy A., Heggland, Sara J., and Peter R. Craig. (2006). Characterization and in vitro Cytotoxicity Testing of Ethanolamine-derived Cadmium Chelating Agents. *BioMetals*. DOI 10.1007/s10534-006-9015-1.

Dr. Cheryl L. Jorcyk

Department of Biology, Boise State University

Jorcyk, Cheryl L., Holzer, Ryan G., and Randall E. Ryan. (2006). Oncostatin M induces cell detachment and enhances the metastatic capacity of T-47D human breast carcinoma cells. *Cytokine*. 33(6): 323-336.

Dr. Michael Laskowski

Department of Biological Sciences, University of Idaho

Potluri, Srilatha, Lampa, Steven, Norton, Ann, and Michael Laskowski. (2006). Morphometric analysis of neuromuscular topography in the serratus anterior muscle. *Muscle-And-Nerve*. 33(3): 398-408.

Dr. You Qiang

Department of Physics, University of Idaho

Joseph Nutting, Jiji Antony, Daniel Meyer, Amit Sharma, and You Qiang. (2006). The effect of particle size distribution on the usage of the ac susceptibility in biosensors. *Journal of Applied Physics*. 99, 08B319.

Dr. Kenneth Rodnick

Department of Biology, Idaho State University

Farrar, Richard S., Battiprolu, Pavan K., Pierson, Nicholas S., and Kenneth J. Rodnick. (2006). Steroid-induced cardiac contractility requires exogenous glucose, glycolysis and the sarcoplasmic reticulum in rainbow trout. *J Exp Biol*. 209: 2114-2128.

Dr. Troy T. Rohn

Department of Biology, Boise State University

Mouser, Peter E., Head, Elizabeth, Ha, Kwang-Ho, Rohn, and Troy T. Rohn. (2006). Caspase-Mediated Cleavage of Glial Fibrillary Acidic Protein within Degenerating Astrocytes of the Alzheimer's Disease Brain. *Am J Pathol*. 168: 936-946.

Dr. Eva Top

Department of Biological Sciences, University of Idaho

Sota, Masahiro, Yano, Hirokazu, Ono, Akira, Miyazaki, Ryo, Ishii, Hidenori, Genka, Hiroyuki, Top, Eva M., and Tsuda, Masataka. (2006). Genomic and Functional Analysis of the IncP-9 Naphthalene-Catabolic Plasmid NAH7 and Its Transposon Tn4655 Suggests Catabolic Gene Spread by a Tyrosine Recombinase. *J. Bacteriol.* 188: 4057-4067.

INBRE Faculty Release Time for Curriculum Development Yields New Immunology Course at Lewis-Clark State College

By Jacob Hornby

During Spring 2005, INBRE funded a course release for the development of the BIOL443 – Immunology course at Lewis-Clark State College. This course was offered for the first time last fall and was received well by the 11 students enrolled. This course primarily serves as an upper division elective for pre-health professional students, but in the future is likely to draw students from allied health professions, such as the Bachelor of Science in Nursing program, as well as additional students from the growing population of biology majors at LCSC. The course focuses on the expanding field of immunology with emphases on the cells and organs of the immune system, structure and genetics of immunoglobulins,

T cell and B cell maturation and activation, the complement system, inflammatory reaction, hypersensitivities, and immunodeficiencies. Year one funds from INBRE allowed release time for comparison of this course to like courses offered at other peer institutions, as well as the opportunity to begin integrating case studies into the lecture content of LCSC courses to enhance student involvement. The course has provided an exciting and recently emerging content area for students at the upper division level.

COBRE Renewal for \$10.1 Million awarded to the State of Idaho to Continue Biomedical Research on Infectious Diseases

By Bill Loftus

In October 2005, Dr. Gregory Bohach was awarded a \$10.1 million, five-year renewal of the COBRE Grant from the National Institutes of Health (NIH). This renewal grant will expand on the original five-year, \$9.6 million awarded in 2000 by continuing to focus on the study of molecular and cellular basis of host-pathogen interactions and allowing University of Idaho scientists and scientist across the state to continue biomedical research focused on infectious diseases

This renewal of funding for infectious disease research will allow the university to help junior researchers develop competitive research programs. Their work will focus on the parasite that causes toxoplasmosis, staphylococcus diseases, human cytomegalovirus, bubonic plague and fungal infections. It also will allow the university to expand research programs and recruit top students and researchers, said Greg Bohach, associate dean and director of the Idaho Agricultural Experiment Station at the University of Idaho.

This funding will support a team of researchers and provide resources for recruitment efforts for new researchers and graduate students, fund undergraduate research, and enhance equipment and research cores.

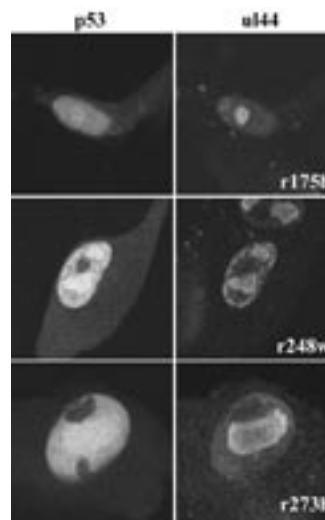
The five research projects support: Dr. Gustavo Arrizabalaga, a molecular parasitologist; Dr. Mark McGuire, an animal scientist and lactation physiologist; Dr. Elizabeth A. Fortunato, a cellular virologist; Dr. Scott Kobayashi, a molecular immunologist;

and Dr. Bruce Miller, a fungal developmental geneticist.

For more information regarding this article, please visit the Idaho website at: www.today.uidaho.edu/Details.aspx?ID=3262.



Gregory Bohach, Ph.D., Director of the Idaho Agricultural Experiment Station and associate dean



Microscopy views of Dr. Elizabeth Fortunato's COBRE Research. Right hand panels (ul44) are pictures of the viral protein ul44, which localizes to the distinct foci where the virus is replicating. On the left of each set is the localization (in the same cell) of a MUTANT p53 (cellular) protein that is engineered and put into the cells. Showing that if you mutate the ability to bind to DNA (for the p53 protein) you loose the ability to be sequestered into the viral replication centers.

DON'T FORGET TO CITE NIH

on your publications and presentations

"This publication was made possible by NIH Grant Number ##### from the (Enter program title here) Program of the National Center for Research Resources."

OR

"The project described was supported by NIH Grant Number ##### from the (Enter program title here) Program of the National Center for Research Resources,"

AND, AS APPROPRIATE

"Its contents are solely the responsibility of the authors and do not necessarily represent the official views of NIH."

Program Titles and Grant Number:

Idaho COBRE Program – P20 RR15587 – Dr. Gregory Bohach, PI.; Idaho COBRE (II) Program – P20 RR16448 – Dr. Larry Forney, PI.; Idaho INBRE Program – P20 RR016454 – Dr. Carolyn Horde Bohach, PI.



Sign-up for the INBRE Electronic Bi-weekly Newsletters

In September 2006, we began distributing an electronic bi-weekly newsletter to all BRIN/INBRE participants. This electronic newsletter includes statewide announcements, upcoming deadlines, funding information, and Kudos for INBRE faculty and students. If you would like to receive the INBRE newsletter, please e-mail inbre@uidaho.edu to be added to our mailing list.



University of Idaho

Idaho INBRE Program
University of Idaho
PO Box 444207
Moscow, ID 83844-4207

