Division of Intramural Research

NAEHS Council Update

February 2010

REORGANIZATION WITHIN THE DIVISION OF INTRAMURAL RESEARCH

The Laboratory of Pharmacology and Laboratory of Molecular Toxicology have been combined into one laboratory within the Environmental Toxicology Program. The new laboratory is called the Laboratory of Toxicology and Pharmacology. This reorganization will enhance scientific working relationships, communications, productivity, and effectiveness by providing an opportunity for the two laboratories to consolidate and assimilate their goals. The Laboratory serves as a nucleus for expanding the Institute's expertise in the mechanisms of the effects of environmental chemicals. Dr. David Miller, the former Acting Director, Environmental Toxicology Program; Acting Chief, Laboratory of Pharmacology; and Acting Chief, Laboratory of Molecular Toxicology; now serves as the Director, Environmental Toxicology Program and the Chief of the newly formed Laboratory of Toxicology and Pharmacology.

The mission of the new laboratory is as follows: The Laboratory of Toxicology and Pharmacology utilizes the approaches and techniques of toxicology, pharmacology, biochemistry, and molecular biology to study the mechanisms of the effects of environmental chemicals to link environmental exposures with the diseases they cause or exacerbate by (1) designing studies to characterize chemical toxicity utilizing genetically tractable models and comparative genomics to focus on gene/environment interactions; (2) applying expression array technology and mathematical models to examine gene networks impacted by chemicals and their metabolites; (3) focusing on the genetic factors predisposing specific individuals to disease upon environmental exposure; (4) studying the processes of exposure and disposition of environmental chemicals; (5) assessing the enzyme systems involved in metabolism of environmental chemicals and drugs, including their genetic heterogeneity; (6) examining the mechanisms responsible for the toxic effects of xenobiotics and their metabolites, including photochemical and free radical mechanisms; (7) evaluating the roles of membrane transporters and receptors in the cellular response to xenobiotic exposure; and (8) providing a focus within NIEHS for the application of alternative model systems from comparative and marine biology to study the pharmacology and toxicity of chemicals and drugs.

DIR RECRUITMENTS

Director Division of Intramural Research
The NIEHS is seeking an exceptional scientific leader interested in being a part of a dynamic management team to fill the position of Director, Division of Intramural Research. In addition, the selectee will also serve as the Scientific Director. The incumbent of this position will direct laboratory and clinical research. The Director, DIR, also serves as a principal advisor to the Institute Director on intramural scientific activities involving interdisciplinary biomedical research programs; develops and recommends policies for the execution of research programs; determines effectiveness of current programs and recommends policies for new programs; and develops new and revised programs to meet national environmental health needs. The Division is organized into five scientific programs, including the Clinical Research Program, which highlight the areas of research excellence of NIEHS. These programs are highly interrelated, interactive and synergistic. Using the interdisciplinary biomedical research approach, the mission of the DIR is to contribute to the basic understanding of biological and chemical processes, understanding of the contributions of environmental agents to human disease and dysfunction and to the underlying mechanisms of environmentally associated diseases. Dr. Patricia Grady, Director, National Institute of Nursing Research, is chair of the search committee.

Tenure-Track Environmental Epidemiologist
The Epidemiology Branch, National Institute of Environmental Health Sciences, NIH, invites applications for a tenure-track epidemiologist to develop an independent investigator-initiated research program. Applicants must have an M.D. and/or Ph.D. in epidemiology or related field, at least two years of post-degree research experience, and a record of accomplishment, including relevant peer-reviewed publications. Expertise is welcome in environmental epidemiology focusing on non-cancer endpoints. Applicants will be evaluated on their demonstrated ability to conduct biologically-based, interdisciplinary, population-level research in environmental epidemiology. Dr. E. Mitch Eddy, Laboratory of Reproductive and Developmental Toxicology, is chair of the search committee.

Tenure-Track Developmental Biologist
A position is available for a Developmental Biologist to establish an independent basic research program and form a research group in the Laboratory of Reproductive and Developmental Toxicology, Division of Intramural Research. Applications are invited from scientists with demonstrated ability for creative and productive research in cellular and molecular mechanisms of mammalian development. Of particular interest are investigators using rodent models to study cell interactions, epigenetics or other basic biomedical problems relating to the impact of the environment on development. The successful candidate will interact with investigators studying diverse problems in reproductive biology, developmental toxicology, hormone mechanisms, signal transduction, cell cycle regulation, cell growth and differentiation, apoptosis, gene regulation, mutagenesis and DNA repair, and cancer biology. Minimum qualifications are an M.D., Ph.D., D.V.M. or equivalent doctoral degree in the biomedical sciences, at least three years of postdoctoral experience, and publications in high quality journals. Dr. Darryl Zeldin,

Acting Clinical Director and Laboratory of Respiratory Biology, is chair of the search committee.

NEW APPOINTMENTS IN THE DIVISION OF INTRAMURAL RESEARCH

Dr. Paul Foster, Chief, Toxicology Branch

Dr. Paul Foster was recently promoted to Senior Scientist and is Chief of the Toxicology Branch in the National Toxicology Program (NTP) at NIEHS. Dr. Foster received his Ph.D. from Brunel University, Uxbridge, England in 1977 and before joining NIEHS he had worked in industry (ICI and Zeneca) and at not-for- profit research institutions (BIBRA and CIIT) as a leader of research programs in endocrine, reproductive and developmental toxicology (ERDT). Dr. Foster is an internationally recognized expert in ERDT and has served on numerous international committees dealing with this subject. His research interests span from understanding the potential human health effects of environmental endocrine disruptors (particularly antiandrogens); mechanisms of testicular toxicity; the study of early testicular Leydig cell dysfunction induced by chemicals as a prelude to hyperplasia and tumors and the toxicokinetic and dynamic parameters affecting the induction of reproductive and developmental toxicity. He also has a broad interest in risk assessment issues in these areas and currently serves as the NTP's senior discipline expert in reproductive and developmental Toxicology.

More recently within the NTP, Dr. Foster has focused his attention on raising the profile of the Program's non-cancer efforts, including the development of specific criteria for the evaluation of reproductive and developmental toxicity studies undertaken by the Program. He has also championed efforts to establish early life exposures as a default exposure paradigm on NTP carcinogenicity studies to better estimate lifetime cancer hazard. This approach arose from an NTP workshop organized by Dr. Foster to explore how well standard approaches to conducting cancer bioassays reflected the newer science emerging on critical windows of exposure for the induction of tumors via hormonal mechanisms (particularly testicular prostate, breast and ovarian cancers). Dr. Foster's recent research is focused on investigating the mechanisms of environmental chemical and drug effects on the development of the reproductive system.

<u>Selected Publications:</u>

Foster, P.M.D. Disruption of reproductive development in male rat offspring following *in utero* exposure to phthalate esters. ***Int. J. Androl.*** 29: 140-147, 2006.

Bell, D.R., Clode, S., Fan, M.Q., Fernandes, A., Foster, P.M.D., Jiang, T., Loizou, G., MacNicoll, A., Miller, B.G., Rose, M., Tran, L and White, S. Toxicity of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) in the developing male Wistar(Han) rat II: Chronic dosing causes developmental delay. ***Toxicol. Sci.*** 99: 224-233, 2007.

Foster, P.M.D and Gray, L.E. Toxic responses to the reproductive system. In: Cassarett & Doull's Toxicology: the basic science of poisons. (Klaassen, C.D, ed). 7th Edition, McGraw Hill, New York. pp 761-806, 2008.

Hotckiss, A.K., Rider, C.V., Blystone, C.R., Wilson, V.S., Hartig, P.C., Ankley, G.T., Foster, P.M.D., Gray, C.L. and Gray, L.E. Fifteen years after "Wingspread"-Environmental endocrine disrupters and human and wildlife health: Where we are today and where we need to go. ***Toxicol. Sci.*** 105: 235-259, 2008.

National Research Council Committee on Health Risks of Phthalates, Phthalates and Cumulative Risk: the Tasks Ahead. National Academies Press, Washington DC, 2008.

Dr. Patricia Jensen, Tenure-Track Investigator, Laboratory of Neurobiology

Dr. Jensen received her Ph.D. in Anatomy and Neurobiology at The University of Tennessee Health Science Center in 2002. Her graduate studies in the laboratory of Dr. Dan Goldowitz focused on the cellular and molecular interactions underlying cerebellar morphogenesis. During her postdoctoral training in the laboratory of Dr. Tom Curran at St. Jude Children's Research Hospital she managed the high-throughput in situ hybridization screen as part of the GENSAT project. In 2005 she joined the laboratory of Dr. Susan Dymecki at Harvard Medical School as a postdoctoral fellow where she carried out molecular and genetic studies focusing on the embryonic and molecular development of individual serotonergic (5-HT) neuron subtypes. Dr. Jensen was recruited to the NIEHS in 2009 to head the Developmental Neurobiology Group in the Laboratory of Neurobiology.

The Developmental Neurobiology Group studies how genetic and environmental perturbations during development alter the fates and functions of specific sets of neurons and how these alterations lead to neurological disorders.

Altered noradrenergic signaling in the prefrontal cortex is implicated in a number of cognitive disorders including autism, attention-deficit/hyperactivity disorder, depression and Alzheimer's disease. To date, much of our understanding about the subpopulation(s) of noradrenergic neurons that project to and modulate prefrontal cortical circuits comes from non-genetic tract tracing and lesioning studies. Little is known about the molecular identity of these subpopulations. Unraveling the genetic pathways that control final noradrenergic subtype identity is critical to our understanding of related developmental and neurodegenerative diseases. In order to fill this knowledge gap my lab uses genetic approaches in the mouse to determine the origins, fates, and functions of the different types of noradrenergic neurons in the mammalian brain. Importantly, this work will provide a means to visualize and genetically manipulate select populations of noradrenergic neurons in vivo and guide the rational generation of mouse models of cognitive disorders.

Major areas of research interests include: Mammalian brain development; neuronal fate specification; and mouse models of neurodevelopmental disorders. Current projects include: Genetic fate-mapping of molecularly defined subdomains within the noradrenergic primordium; molecular profiling of subsets of mouse noradrenergic neurons; and genetic ablation of noradrenergic neurons and analysis of prefrontal target areas.

Selected Publications:

Jensen P, Farago AF, Awatramani RB, Scott MM, Deneris ES, Dymecki SM. Redefining the serotonergic system by genetic lineage. *Nat. Neurosci.* 11: 417-419, 2008.

Du X, Jensen P, Goldowitz D, Hamre KM. Wild-type cells rescue genotypically math1-null hair cells in the inner ears of chimeric mice. *Dev. Biol.* 305: 430-438, 2007.

Jensen P, Magdaleno S, Seal A, Lehman K, Asbury A, Cheung T, Cornelius T, Batten DM, Eden C, Norland S, Rice DS, Dosooye N, Shakya S, Mehta P, Curran T. The brain gene map expression (BGEM): A database containing *in situ* hybridization data of gene expression in the developing and adult mouse nervous system. *PLoS Biol.* 4:e86, 2006.

Jensen P, Magdaleno S, Lehman KM, Rice DS, Lavallie ER, Collins-Racie L, McCoy JM, Curran T. A neurogenomics approach to gene expression analysis in the developing brain. *Brain Res. Mol. Brain Res.* 132: 116-127, 2004.
Jensen P, Smeyne R, Goldowitz D. Analysis of cerebellar development in *math1* null embryos and chimeras. *J. Neurosci.* 24: 2202-2211, 2004.

Dr. Guang Hu, Tenure-Track Investigator, Laboratory of Molecular Carcinogenesis

Dr. Guang Hu recently joined the Laboratory of Molecular Carcinogenesis at NIEHS as a tenure-track stem cell biologist. Dr. Hu earned his Ph.D. in Biochemistry and Molecular Biology in 2003 at Baylor College of Medicine, Houston, Texas. He then completed a postdoctoral fellowship at Harvard Medical School under the supervision of Dr. Stephen Elledge, before joining NIEHS in 2009. His postdoctoral research has made major contributions to two areas: (1) the identification of novel regulators in embryonic stem cell self-renewal through systematic RNAi screens, and (2) the development of novel high-throughput RNAi screen technology.

At the NIEHS, Dr. Hu is now investigating the molecular mechanism of embryonic stem cell self-renewal and differentiation. Embryonic stem cells (ES cells) are pluripotent cells derived from the inner cell mass of the blastocyst-stage embryo. They can be cultured continuously in their pluripotent state, and can also be induced to differentiate into cell types from all three germ layers. Because of these unique properties, ES cells can be used as a model system to study the mechanism of pluripotency and fate-specification during early mammalian development, and they can also be used to derive various types of cells for disease modeling, drug discovery, and the development of cell-based therapies. Dr. Hu is interested in understanding the molecular circuitry that controls self-renewal and lineage-specifications in ES cells. He will continue to use the functional genetic approach he developed to identify and investigate genes and pathways that are involved in these processes. His research will improve our knowledge in developmental biology and facilitate the development of stem cell therapies. It will also contribute to our understanding on how environmental factors can affect human embryogenesis and early development.

Selected Publications:

Hu G, Kim J, Xu Q, Leng Y, Orkin SH, Elledge SJ. A Genome-wide RNAi screen identifies a new transcriptional module required for self renewal. *Genes Dev.* 23: 837-848, 2009
Ali N, Karlsson C, Aspling N, Hu G, Hacohen N, Scadden DT, Larsson J. Forward RNAi screens in primary human hematopoietic stem/progenitor cells. *Blood* 113: 3690-3695, 2009.
Westbrook TF, Hu G, Ang XL, Mulligan P, Pavlova NN, Liang A, Leng Y, Maehr R, Shi Y, Harper JW, Elledge SJ. SCFbeta-TRCP controls oncogenic transformation and neural differentiation through REST degradation. *Nature* 452: 370-374, 2008.
Schlabach MR, Luo J, Solimini NL, Hu G, Xu Q, Li M, Zhao Zh, Smogorzewska A, Sowa ME, Ang XL, Westbrook TF, Liang A, Chang K, Hackett JA, Harper JW, Hannon GJ, Elledge SJ. Cancer proliferation gene discovery through functional genomics. *Science* 319: 620-624, 2008.

Silva JM, Li MZ, Chang K, Ge W, Golding MC, Rickles RJ, Siolas D, Hu G, Paddison PJ, Schlabach MR, Sheth N, Bradshaw J, Burchard J, Kulkarni A, Cavet G, Sachidanandam R, McCombie WR, Cleary MA, Elledge SJ, Hannon GJ. Second-generation shRNA libraries covering the mouse and human genomes. *Nat. Genet.* 37: 1281-1288, 2005.

Stegmeier F, Hu G, Rickles RJ, Hannon GJ, Elledge SJ. A lentiviral microRNA-based system for single-copy polymerase II-regulated RNA interference in mammalian cells. *Proc. Natl. Acad. Sci. U.S.A.* 102: 13212-13217, 2005.

Dr. R. Scott Williams, Tenure-Track Investigator, Laboratory of Structural Biology

Dr. Scott Williams recently joined the Laboratory of Structural Biology at NIEHS as a tenure-track structural biologist. Dr. Williams earned his doctoral degree in Biochemistry with a specialization in macromolecular X-ray crystallography (Ph.D. 2003, University of Alberta, Canada). His postdoctoral training in the use of combined low- and high- resolution structural biological techniques was obtained at the Scripps Research Institute (La Jolla, CA). His research has made major contributions to two different areas: (1) structure/function and mechanisms of inactivation of the BRCA1 tumor suppressor (2) structural and functional roles of the Mre11/Rad50/Nbs1 complex in the maintenance of genomic integrity.

At the NIEHS, Dr. Williams is focusing on understanding the fundamental structural chemistry through which DNA damage is recognized, signaled and repaired. Thousands of times each day, our DNA strands are damaged as our cells duplicate their chromosomes, and as a consequence of oxidation or environmental exposure to chemical mutagens and DNA-damaging radiation. Left un-repaired, harmful forms of DNA damage such as DNA double strand breaks or DSBs, can lead to gross chromosomal rearrangements, genome instability and cancer. The Williams group employs multidisciplinary structural methods (X-ray crystallography and small angle X-ray scattering – SAXS) and biochemical approaches to elucidate: (1) With atomic resolution, how DNA damage is sensed and enzymatically corrected, (2) How DNA repair factors are impacted by heritable mutations in human cancer predisposition syndromes and other diseases, (3) How the cellular DNA repair machinery can be targeted for the improved treatment of human cancers.

<u>Selected Publications:</u>

Williams RS, Dodson GE, Limbo O, Yamada Y, Williams JS, Guenther G, Classen S, Glover JNM, Iwasaki H, Russell P, Tainer JA. Nbs1 is an Extended Flexible Arm Binding to Ctp1 and Mre11-Rad50 to Coordinate dsDNA Break Processing. *Cell* 139: 87-99, 2009.

Williams RS, Moncalian G, Williams JS, Yamada Y, Limbo O, Shin DS, Groocock LM, Cahill D, Hitomi C, Guenther G, Moiani D, Carney JP, Russell P, Tainer JA. Mre11 Dimers Coordinate DNA End Bridging and Nuclease Processing in DNA Double Strand Break Repair. *Cell* 135: 97-109, 2008.

Bernstein N, Williams RS, Rakovszky M, Cui D, Green R, Karimi-Busheri F, Mani R, Galicia S, Koch CA, Cass C, Durocher D, Weinfeld M, Glover JN. The

Molecular architecture of the mammalian DNA repair enzyme, polynucleotide kinase. *Mol. Cell* 17: 657-670, 2005.

Williams RS, Lee MS, Hau DD, Glover JNM. Structural basis of phospho-peptide recognition by the BRCA1 BRCT domain. *Nat. Struct. Mol. Biol.* 11: 519-525, 2004.

Williams RS, Green R, Glover JNM. Crystal structure of the BRCT repeat region from the breast cancer-associated protein BRCA1. *Nat. Struct. Biol.* 8: 838-843, 2001.

DIR RESEARCH UPDATE

Genetic Susceptibility as a Causal Tool in Environmental Epidemiology

Allen Wilcox, MD, PhD, Epidemiology Branch, NIEHS

Genetic susceptibility has been proposed as a tool for clarifying causal associations in environmental epidemiology. If there are subgroups in the population that are genetically susceptible to a particular exposure, then it would be easier (in principle) to detect the association of the exposure with disease in such subgroups. Strong associations in susceptible populations would provide evidence for causation that may not be apparent in the general population. This logic has been the basis for extensive (and expensive) studies in environmental epidemiology. We explored this approach using a case-control study of cleft lip and palate – a common birth defect with both environmental and genetic causes. We have identified several dietary and other factors related to the risk of facial clefts: folic acid supplements and dietary vitamin A reduce risk, and smoking and binge alcohol consumption increase risk. In an effort to add to the evidence for causation, we assayed genes that control the metabolic pathways of these factors, looking for stronger effects of the exposures in genetically-susceptible groups. I will present our results, and discuss the difficulty of conducting and interpreting these studies.

Genome–Wide Association Studies of Asthma and Pulmonary Function.

Stephanie J. London, M.D., Dr.P.H., Epidemiology Branch and Laboratory of Respiratory Biology, NIEHS

While inherited susceptibility clearly influences risk of asthma and variation in pulmonary function, the specific genes involved have largely remained elusive. The genome-wide association study (GWAS) has emerged as a powerful tool to identify novel genetic loci but there have been few studies of asthma or pulmonary function. We undertook two separate GWAS analyses. One GWAS of childhood asthma in Mexico City gives evidence for a novel candidate gene, TLE4. GWAS of pulmonary function based on meta-analysis of 20,000 adults from four population based cohorts (the CHARGE consortium) identified one novel locus for the forced expiratory volume in one second (FEV1) and 7 novel loci for its ratio to the forced vital capacity (FEV1/FVC). Five loci were replicated in a second consortium of comparable size. This work, and future directions, will be presented.

NIEHS SCIENCE AWARDS DAY

The Seventh Annual DIR NIEHS Science Awards Day was held on November 5, 2009, at the Rall Building on the NIEHS Campus to celebrate the achievements of DIR scientists. The event was open to the public and more than 250 attendees from universities and research institutions in the Triangle Area attended. NIEHS Science Awards Day consisted of 8 oral presentations given by fellows, students, and technicians, 87 poster presentations, oral presentations by the Scientist of the Year, Early Career Award and Outstanding Staff Scientist winners, and an Awards Ceremony. Judging for the awards was done by the NIEHS Board of Scientific Counselors, Extramural Scientists from universities in the Triangle Area, Intramural DIR Scientists and the NIEHS Training Assembly.

The following awards were presented at NIEHS Science Awards Day:

Scientist of the Year: Kenneth S. Korach, Ph.D., Laboratory of Reproductive and Developmental Toxicology

Early Career Award: Raja Jothi, Ph.D., Biostatistics Branch

Outstanding Staff Scientist: Dmitry Gordenin, Ph.D., Laboratory of Molecular Genetics

Mentor of the Year: Serena Dudek, Ph.D., Laboratory of Neurobiology

Best Poster Presentation in Environmental Biology: Nisha A. Cavanaugh, Ph.D., Laboratory of Structural Biology

Best Poster Presentation in Environmental Diseases and Medicine: Daniel A. Gilchrist, Ph.D., Laboratory of Molecular Carcinogenesis

Best Poster Presentation in Environmental Toxicology: Brooke Tvermoes, Ph.D., Laboratory of Molecular Toxicology

Best Oral Presentation: Stephen B. Simons, Ph.D., Laboratory of Neurobiology

Paper of the Year: From the Laboratory of Molecular Genetics and Laboratory of Structural Biology: S.A. Nick McElhinny, D.A. Gordenin, C.M. Stith, P.M.J. Burgers and T.A. Kunkel "Division of Labor at the Eukaryotic Replication Fork" *Molecular Cell* 30: 137-144, 2008.

TRAINING AND MENTORING

Department of Defense Congressionally Directed Medical Research Program Fellowship.

Sangmi Kim, Ph.D., a postdoctoral fellow in the Chronic Disease Epidemiology Group, received a Postdoctoral Fellowship Award from the Breast Cancer Research Program (BCRP) of the Department of Defense Congressionally Directed Medical Research Program for 2009. The BCRP funds innovative research that could significantly impact breast cancer research or addresses neglected issues. The program seeks to instigate multidisciplinary and multi-institutional collaborations and seeks to involve early career investigators with a "strong desire to pursue a career in breast cancer research." The BCRP has a two-tiered review process that involves breast cancer survivors as well as scientists in evaluating research projects. Sangmi's mentors are Dr. Dale Sandler (Epidemiology Branch) and Dr. Jack Taylor (Epidemiology Branch and Laboratory of Molecular Carcinogenesis).

Gehring Award from the Society of Toxicology

Scott Auerbach, Ph.D., a former postdoctoral fellow in the Toxicology Branch, received the Perry J. Gehring Award for the Best Presentation in Risk Assessment by a Postdoctoral Scientist at the 2009 Society of Toxicology annual meeting for his presentation titled "Independent Validation of Gene Expression-Based Hepatocarcinogenicity Prediction Models." His mentor was Dr. Richard Irwin (Toxicology Branch).

Laboratory Animal Medicine Award

Coralie Zegre-Cannon, D.V.M., a postdoctoral fellow in the NTP Laboratory Animal Management Group, recently received first-place honors at the 60th annual American Association of Laboratory Animal Science (AALAS) National Meeting held Nov. 8-12 in Denver. Zegre-Cannon received the award for her poster presentation, "Evaluation of Route of Administration and Dosage of Tramadol as an Analgesic in the Rat." Her mentor is Dr. Angela King-Herbert (NTP Laboratory Animal Management Group).

Outstanding New Environmental Scientist (ONES) Awards

Michelle Block, Ph.D. an Assistant Professor of Anatomy and Neurobiology at Virginia Commonwealth University and a former NIEHS Postdoctoral Fellow in the Laboratory of Pharmacology (Dr. Jau-Shyong Hong, mentor) received one of six highly competitive five-year ONES grants awarded this year by NIEHS. Established in 2006, the ONES program identifies outstanding scientists who are in the early, formative stages of their careers and who intend to make a long-term career commitment to research in the mission areas of the NIEHS. The program assists them in launching an innovative research program focusing on problems of environmental exposures and human biology, human pathophysiology and human disease. Dr.

Block will explore the role of protein radicals in microglia in the environmental mechanisms of chronic neurotoxicity.

TOP DIR PUBLICATIONS FOR THE YEAR

Genome-Wide Association Study (GWAS) Identifies Multiple Loci Associated with Lung Function

As part of the Institute's effort to increase our understanding the gene-environment interactions that lead to disease, researchers from the NIEHS directed the CHARGE Consortium in a meta-analysis of GWAS data for two clinically important lung-function measures: forced expiratory volume in the first second (FEV_1) and its ratio to forced vital capacity (FEV_1/FVC), an indicator of airflow obstruction. This meta-analysis included 20,890 participants of European ancestry from four CHARGE Consortium studies: Atherosclerosis Risk in Communities, Cardiovascular Health Study, Framingham Heart Study and Rotterdam Study. The researchers identified eight loci associated with FEV_1/FVC (HHIP, GPR126, ADAM19, AGER-PPT2, FAM13A, PTCH1, PID1 and HTR4) and one locus associated with FEV_1 (INTS12-GSTCD-NPNT). These loci were replicated with the SpiroMeta consortium (16,178 participants). The identified loci are all biologically plausible candidates that are involved in pathways known to be important in lung biology, including immune function, muscle function or inflammation. These findings bring new insight into our understanding of the genetic basis of lung development and the related airway disorders asthma and chronic obstructive pulmonary disease.

Hancock DB, Eijgelsheim M, Wilk JB, Gharib SA, Loehr LR, Marciante KD, Franceschini N, van Durme YM, Chen TH, Barr RG, Schabath MB, Couper DJ, Brusselle GG, Psaty BM, van Duijn CM, Rotter JI, Uitterlinden AG, Hofman A, Punjabi NM, Rivadeneira F, Morrison AC, Enright PL, North KE, Heckbert SR, Lumley T, Stricker BH, O'Connor GT, London SJ. Meta-analyses of genome-wide association studies identify multiple loci associated with pulmonary function. *Nat. Genet.* **42:** 45-52, 2010.

Sun Exposure May Trigger Certain Autoimmune Diseases in Women

NIEHS researchers in collaboration with myositis centers across the country examined whether ultraviolet (UV) radiation from sunlight was associated with the development a form of autoimmune muscle disease known as dermatomyositis, which weaken the muscles and causes distinctive rashes, instead of a form called polymyositis that does not have a rash. The researchers assessed UV exposure at the time of onset of disease with the prevalence of dermatomyositis and myositis antibodies in 380 patients with myositis. An association was found between UV radiation intensity and the proportion of patients with dermatomyositis and with the proportion of patients expressing anti-Mi-2 autoantibodies. These associations were confined to women suggesting that sex influences the effects of UV radiation on autoimmune disorders. Studies are planned to investigate the mechanisms by which these effects are produced, providing insights into the pathogenesis of disease and suggest therapeutic or preventative strategies.

Love LA, Weinberg CR, McConnaughey DR, Oddis CV, Medsger TA Jr, Reveille JD, Arnett FC, Targoff IN, Miller FW. Ultraviolet radiation intensity predicts the

relative distribution of dermatomyositis and anti-Mi-2 autoantibodies in women. *Arthritis Rheum.* 60: 2499-2504, 2009.

Widespread Promoter-Proximal Stalling and Arrest of RNA Polymerase II

It is commonly believed that the recruitment of RNA polymerase II (Pol II) to a promoter is sufficient for gene activation and transcript elongation. However, recent genome-wide studies of Pol II distribution have demonstrated that Pol II accumulates at promoters of many developmentally regulated and stimulus-response genes in their uninduced states and that gene expression in higher organisms is regulated by Pol II stalling during early transcription elongation. Scientist at NIEHS collaborated with an investigator at Virginia Commonwealth University to probe the mechanisms responsible for this regulation. They developed methods to isolate and identify short RNAs derived from stalled Pol II in *Drosophila* cells. Significant levels of these short RNAs were generated from over one-third of all genes, indicating that promoter-proximal stalling is a general feature of early polymerase elongation. The nucleotide composition of the initially transcribed sequence played an important role in promoting transcriptional stalling by rendering polymerase elongation complexes highly susceptible to backtracking and arrest. These results indicate that the intrinsic efficiency of early elongation can greatly affect gene expression.

Nechaev S, Fargo DC, dos Santos G, Liu L, Gao Y, Adelman K. Global analysis of short RNAs reveals widespread promoter-proximal stalling and arrest of Pol II in *Drosophila*. *Science* 327: 335-338, 2010.

Key Regulator of Hepatic Lipid Metabolism Identified

Hepatic metabolic derangements are key components in the development of fatty liver, insulin resistance, and atherosclerosis. SIRT1, a NAD^+-dependent protein deacetylase, is an important regulator of energy homeostasis in response to nutrient availability. NIEHS scientists demonstrated that hepatic SIRT1 regulates lipid homeostasis by positively regulating peroxisome proliferators-activated receptor α (PPARα), a nuclear receptor that mediates the adaptive response to fasting and starvation. Hepatocyte-specific deletion of SIRT1 impaired PPARα signaling and decreased fatty acid β-oxidation, whereas overexpression of SIRT1 induced the expression of PPARα targets. SIRT1 interacts with PPARα and is required to activate PPARα coactivator PGC-1α. When challenged with a high-fat diet, liver-specific SIRT1 knockout mice develop hepatic steatosis, hepatic inflammation, and endoplasmic reticulum stress. Taken together, these findings indicate that SIRT1 plays a vital role in the regulation of hepatic lipid homeostasis and that pharmacological activation of SIRT1 may be important for the prevention of obesity-associated metabolic diseases.

Purushotham A, Schug TT, Xu Q, Surapureddi S, Guo X, Li X. Hepatocyte-specific deletion of SIRT1 alters fatty acid metabolism and results in hepatic steatosis and inflammation. *Cell Metab.* 9: 327-338, 2009.

Calcium Signaling During Mitosis

The role of calcium signaling during cell division has long been a source of mystery. NIEHS researchers determine why store-operated Ca^{2+} entry (SOCE) and Ca^{2+} release-activated Ca^{2+} currents (I(crac)) are strongly suppressed during mitosis, the only known physiological situation in which Ca^{2+} store depletion is uncoupled from the activation of Ca^{2+} influx. The researchers found that the endoplasmic reticulum (ER) Ca^{2+} sensor STIM1 failed to rearrange into near-plasma membrane puncta in mitotic cells, a critical step in the SOCE-activation pathway. They also found that STIM1 from mitotic cells is recognized by the phospho-specific MPM-2 antibody, suggesting that STIM1 is phosphorylated during mitosis. Removal of ten MPM-2 recognition sites by truncation at amino acid 482 abolished MPM-2 recognition of mitotic STIM1, and significantly rescued STIM1 rearrangement and SOCE response in mitosis. Ser 486 and Ser 668 were identified as mitosis-specific phosphorylation sites, and STIM1 containing mutations of these sites to alanine also significantly rescued mitotic SOCE. Therefore, phosphorylation of STIM1 at Ser 486 and Ser 668, and possibly other sites, underlies suppression of SOCE during mitosis.

Smyth JT, Petranka JG, Boyles RR, DeHaven WI, Fukushima M, Johnson KL, Williams JG, Putney JW Jr. Phosphorylation of STIM1 underlies suppression of store-operated calcium entry during mitosis. *Nat. Cell Biol.* 11: 1465-1472, 2009.

DNA Scrunching During Gap Repair Synthesis

Family X polymerases such as DNA polymerase λ (Pol λ) are well suited for filling short gaps during DNA repair because they simultaneously bind both the 5' and 3' ends of short gaps which may occur during environmental exposures. DNA binding and gap filling are well characterized for 1-nucleotide (nt) gaps, but the location of yet-to-be-copied template nucleotides in longer gaps is unknown. NIEHS researchers in collaboration with scientists at the University of North Carolina at Chapel Hill determined crystal structures revealing that, when bound to a 2-nt gap, Pol λ scrunches the template strand and binds the additional uncopied template base in an extrahelical position within a binding pocket that comprises three conserved amino acids. Replacing these amino acids with alanine results in less processive gap filling and less efficient nonhomologous DNA end-joining when 2-nt gaps are involved. This study sheds light on the specific structural changes necessary during DNA repair and, ultimately, to protection against mutations due to environmental exposures.

Garcia-Diaz M, Bebenek K, Larrea AA, Havener JM, Perera L, Krahn JM, Pedersen LC, Ramsden DA, Kunkel TA. Template strand scrunching during DNA gap repair synthesis by human polymerase lambda. *Nat. Struct. Mol. Biol.* 16: 967-972, 2009.

Link Between Serum Cholesterol and Asthma

Cholesterol exerts complex effects on inflammation. However, there has been little investigation of whether serum cholesterol is associated with asthma, an inflammatory airways disease with great public health impact. NIEHS scientists collaborated with investigators at SRA International and Rho Federal Systems Division to determine relationships between levels of 3 serum

cholesterol measures (total cholesterol [TC], high-density lipoprotein cholesterol [HDL-C], and non-HDL-C) and asthma/wheeze in a sample representative of the US population. The researchers conducted a cross-sectional study of 7005 participants age 6 years and older from the 2005 to 2006 National Health and Nutrition Examination Survey. Serum TC and non-HDL-C were lower in patients with current asthma than in subjects without current asthma in the overall population, whereas HDL-C was not different. On racial/ethnic stratification, these relationships reflect marked reductions unique to Mexican Americans. The data may help explain why Mexican Americans have the lowest prevalence of asthma in the country, despite increased asthma risk factors such as low socioeconomic status and obesity.

Fessler MB, Massing MW, Spruell B, Jaramillo R, Draper DW, Madenspacher JH, Arbes SJ, Calatroni A, Zeldin DC. Novel relationship of serum cholesterol with asthma and wheeze in the United States. *J. Allergy Clin. Immunol.* 124: 967-974, 2009.

Requirements for Repair of Radiation-induced DNA Damage

Resection of DNA double-strand break (DSB) ends is considered a critical determinant in pathways of DSB repair and genome stability. Unlike for enzymatically induced site-specific DSBs, little is known about processing of random "dirty-ended" DSBs created by DNA damaging agents such as ionizing radiation. NIEHS researchers in collaboration with a scientist at Indiana University-Perdue University, utilized pulsed field gel electrophoresis (PFGE) to follow the fate of both ends of linear molecules generated by a single random DSB of circular chromosomes in budding yeast. Within 10 min after gamma-irradiation of G2/M arrested wild type (WT) cells, there is a near-synchronous PFGE-shift of the linearized circular molecules, corresponding to resection of a few hundred bases. Resection at the radiation-induced DSBs continues so that by the time of significant repair of DSBs at 1 hr there is about 1-2 kb resection per DSB end. The PFGE-shift is comparable in WT and recombination-defective rad52 and rad51 strains but somewhat delayed in exo1 mutants. However, in rad50 and mre11 null mutants the initiation and generation of resected ends at radiation-induced DSB ends is greatly reduced in G2/M. A similar requirement was found for RAD50 in asynchronously growing cells. Thus, the Rad50/Mre11/Xrs2 complex is responsible for rapid processing of most radiation-induced damaged ends into substrates that subsequently undergo recombinational repair.

Westmoreland J, Ma W, Yan Y, Van Hulle K, Malkova A, Resnick MA. RAD50 is required for efficient initiation of resection and recombinational repair at random, gamma-induced double-strand break ends. *PLos Genet.* 5: e1000656, 2009.

Hippocampal Synaptic Plasticity Can Be Modified by Differential Calcium Handling

Changes in neuronal plasticity play an important role in the normal development of the nervous system. Although much is known about the mechanisms underlying synaptic plasticity, the cellular mechanisms that negatively regulate plasticity in some brain regions are considerably less studied. One region where neurons do not reliably express long-term potentiation (LTP) is the CA2 subfield of the hippocampus. Given the connection between synaptic plasticity and increases in postsynaptic calcium ion concentration ($[Ca^{2+}]$), and that CA2 neurons express a large number of calcium-regulating proteins, scientists at NIEHS and Duke University

collaborated to tested the hypothesis that the relative lack of LTP in CA2 results from differences in the calcium dynamics of these neurons. By measuring calcium-dependent fluorescence transients in dendritic spines, the scientists showed that CA2 neurons have smaller action potential-evoked intracellular Ca^{2+} transients because of a higher endogenous Ca^{2+}-buffering capacity and significantly higher rates of Ca^{2+} extrusion when compared with CA1 and CA3 neurons. Perfusion with higher external [Ca^{2+}] during induction restores LTP to CA2 neurons, suggesting that they possess the cellular machinery required for plasticity, but that the restriction of postsynaptic [Ca^{2+}] limits its expression. Camstatin, an analogue of the calcium-modulating protein Pep-19 strongly expressed in CA2 neurons, blocked LTP and increased Ca^{2+} extrusion in CA1 neurons, suggesting a role for extrusion in the regulation of plasticity in CA2. In agreement with this idea, we found that intracellular introduction of a plasma membrane calcium pump inhibitor (carboxyeosin) allows for the induction of LTP in CA2 neurons. The results indicate that regulation of postsynaptic [Ca^{2+}] through modulation of extrusion and/or buffering regulates expression of LTP in CA2 and potentially other brain regions.

Simons SB, Escobedo Y, Yasuda R, Dudek SM. Regional differences in hippocampal calcium handling provide a cellular mechanism for limiting plasticity. *Proc. Natl. Acad. Sci. U.S.A.* 106: 14080-1408, 2009.

Mechanism of Genistein-Induced Infertility

Female mice treated neonatally with the environmental phytoestrogen genistein have multioocyte follicles, lack regular estrous cyclicity, and are infertile even after superovulation. To determine the cause of their infertility, NIEHS investigators examined oocyte developmental competence and timing of embryo loss. Eggs obtained by superovulation of genistein-treated or control females were equally capable of being fertilized *in vitro* and cultured to the blastocyst stage. However, if eggs were fertilized *in vivo*, retrieved at the pronucleus stage, and cultured, there was a significant reduction in the percentage of embryos from genistein-treated females reaching the blastocyst stage. When these blastocysts were transferred to pseudopregnant recipients, the number of live pups produced was similar to that in controls. Preimplantation embryo development *in vivo* was examined by flushing embryos from the oviduct and/or uterus. Similar numbers of one-cell and two-cell embryos were obtained from genistein-treated and control females. However, significantly fewer embryos were obtained from genistein-treated females. To determine if neonatal genistein treatment altered the ability of the uterus to support implantation, blastocysts from control donors were transferred to control and genistein-treated pseudopregnant recipients. These experiments demonstrated that genistein-treated females are not capable of supporting normal implantation of control embryos. These findings suggest that oocytes from mice treated neonatally with genistein are developmentally competent; however, the oviductal environment and the uterus have abnormalities that contribute to the observed reproductive failure.

Jefferson WN, Padilla-Banks E, Goulding EH, Lao SP, Newbold RR, Williams CJ. Neonatal exposure to genistein disrupts ability of female mouse reproductive tract to support preimplantation embryo development and implantation**.** *Biol. Reprod.* 80: 425-431, 2009.

Methoxyacetic Acid Disrupts Endogenous Estrogen Signaling

Ethylene glycol monomethyl ether (EGME) exposure is associated with impaired reproductive function. The primary metabolite of EGME is methoxyacetic acid (MAA), a short-chain fatty acid that inhibits histone deacetylase activity and alters gene expression. Because estrogen signaling is necessary for normal reproductive function and modulates gene expression, the estrogen-signaling pathway is a likely target for MAA; however, little is known about the effects of MAA in this regard. NIEHS scientists in collaboration with an investigator at the German Cancer Research Center evaluated the mechanistic effects of MAA on estrogen receptor (ER) expression and estrogen signaling using *in vitro* and *in vivo* model systems. MAA potentiates 17β-estradiol (E_2) stimulation of an estrogen-responsive reporter plasmid in HeLa cells transiently transfected with either a human ERα or ERβ expression vector containing a cytomegalovirus (CMV) promoter. This result is attributed to increased exogenous ER expression due to MAA-mediated activation of the CMV promoter. In contrast to its effects on exogenous ER, MAA decreases endogenous ERα expression and attenuates E_2-stimulated endogenous gene expression in both MCF-7 cells and the mouse uterus. These results illustrate the importance of careful experimental design and analysis when assessing the potential endocrine-disrupting properties of a compound to ensure biological responses are in concordance with *in vitro* analyses. Given the established role of the ER in normal reproductive function, the effects of MAA on the endogenous ER are consistent with the reproductive abnormalities observed after EGME exposure and suggest that these toxicities may be due, at least in part, to attenuation of endogenous ER-mediated signaling.

Henley D V, Mueller S, Korach KS. The short-chain fatty acid methoxyacetic acid disrupts endogenous estrogen receptor-α-mediated signaling. *Environ. Hlth. Perspect.* 117: 1702-1706, 2009.

AWARDS AND HONORS

Scientific Awards

Dr. John Cidlowski (Chief, Laboratory of Signal Transduction) received the Edwin B. Astwood award from the US Endocrine Society, and The Keith Harrison award from the Australian Endocrine Society.

Dr. Michael Cunningham (Host Susceptibility Branch) was elected as a fellow of the Academy of Toxicological Sciences.

Dr. Serena Dudek (Laboratory of Neurobiology) received the 2009 A.E. Bennett Research Award from the Society of Biological Psychiatry.

Dr. Ronald Herbert (Cellular and Molecular Pathology Branch) elected as a Fellow of the International Academy of Toxicologic Pathology.

Dr. Fredrick Miller (Office of Clinical Research) was elected to Best Doctors in America.

Dr. Lisa Rider (Office of Clinical Research) was named one of America's Top Pediatricians by the Consumer's Research Council of America.

Dr. Dale Sandler (Epidemiology Branch) received the NIH Director's Award, *"For leadership in developing the Sister Study, an innovative study of environmental and genetic contributors to breast cancer and other diseases in women."*

Dr. Raymond Tice (Chief, Biomolecular Screening Branch) received the Alexander Hollaender Award from the U.S. Environmental Mutagen Society.

Dr. David Umbach (Biostatistics Branch) was named a Fellow of the American Statistical Association.

Dr. Allen Wilcox (Epidemiology Branch) received the 2009 National MCH Epidemiology Award for Advancing Knowledge, *"for your substantial contributions in advancing the knowledge base to improve the health of women, children and families."* He also received the National Maternal and Child Health Award presented by the CDC, NICHD, American Academy of Pediatrics, American Public Health Association, and March of Dimes.

Named Professorships/Lectures

Dr. John Cidlowski (Chief, Laboratory of Signal Transduction) received The Marius Tausk Visiting Professorship from Leiden University in the Netherlands.

Dr. Dori Germolec (Toxicology Branch) was the keynote speaker for the Japanese Society of Immunotoxicology meeting.

Dr. Steven Kleeberger (Acting Deputy Director; Laboratory of Respiratory Biology) was invited to present the 2010 William B. Kinter Memorial Lecture at the Mount Desert Island Biological Laboratory.

Dr. Kenneth Korach (Chief, Laboratory of Reproductive and Developmental Toxicology) gave the Keynote Plenary Address at the Japanese Endocrine Society meeting and at the Finnish Endocrine Society Meeting as well as the Plenary Address at the Japanese Society of Toxicology meeting.

Dr. Thomas Kunkel (Laboratory of Molecular Genetics; Chief, Laboratory of Structural Biology) was the Keynote Speaker at the Sloan-Kettering Institute – London Research Institute of Cancer Research Conference on "Genome Integrity", the Second Erling Seeberg Symposium on DNA Repair and the Gordon Research

Conference on Nucleosides, Nucleotides and Oligonucleotides. Dr. Kunkel also was the Distinguished "Martin Lecturer" at Mount Olive College and The Cell and Developmental Biology Distinguished Speaker Award at Thomas Jefferson University.

Dr. Stephanie London (Epidemiology Branch and Laboratory of Respiratory Biology) was invited to present a Plenary Lecture at the Annual Meeting of the American Academy of Allergy, Asthma and Immunology in February 2010.

Dr. David Malarkey (Cellular and Molecular Pathology Branch) was an invited speaker at the Tenth Annual Meeting of the Institute of Environmental Toxicology.

Dr. Christopher Portier (Laboratory of Molecular Toxicology and Office of the Director) delivered the Keynote Lecture at the Australian College of Toxicology and Risk Assessment Annual Meeting.

Dr. William Stokes (Director, National Toxicology Program Interagency Center for the Evaluation of Alternative Toxicological Methods) presented keynote address at the Annual Meeting of the American Association for the Advancement of Science and at the meeting of the One Health Intellectual Exchange Group.

Dr. Raymond Tice (Chief, Biomolecular Screening Branch) gave the plenary presentations at the 8th International Workshop on the Comet Assay and at the VII World Congress on Alternatives and Animal Use in the Life Sciences.

Dr. Clarice Weinberg (Chief, Biostatistics Branch) gave the Norman E. Breslow lecture at the University of Washington, Seattle.

Dr. Samuel Wilson (Laboratory of Structural Biology) chaired the 2009 Genetic Toxicology Gordon Research Conference; co-chaired the 3rd Biannual US-EU/EU-US DNA Repair Meeting, Galveston, TX; presented the Nakahara Memorial Lecture at the 40th Princess Takamatsu Symposium, Tokyo; and presented the Plenary Lecturer at the 10th International Conference on Environmental Mutagens, Firenze.

Dr. Jerrel Yakel (Laboratory of Neurobiology) presented the E.E. Just Lecture at the Annual Meeting of the American Society of Cell Biology.

Dr. Darryl Zeldin (Acting Clinical Director; Laboratory of Respiratory Biology) presented the State-of-the-Art Lecture at the Annual Meeting of the American Academy of Allergy, Asthma and Immunology.

Scientific Advisory Boards

Dr. William Copeland (Laboratory of Molecular Genetics) was appointed to the Scientific Advisory Board of the United Mitochondrial Disease Foundation.

Dr. Mary Grant (Comparative Medicine Branch) was appointed to the board of directors of The North Carolina Association for Biomedical Research.

Editorial Boards

Dr. Karen Adelman (Laboratory of Molecular Carcinogenesis) was named to the editorial board of the journal *Transcription.*

Dr. Marilyn Diaz (Laboratory of Molecular Genetics) served on the editorial boards of *Autoimmunity* and *The Open Autoimmunity Journal.*

Dr. Gregg Dinse (Biostatistics Branch) served as an associated editor of the journal *Lifetime Data Analysis.*

Dr. Darlene Dixon (Cellular and Molecular Pathology Branch) served on the editorial board of the journal *Veterinary Pathology.*

Dr. E. Mitch Eddy (Laboratory of Reproductive and Developmental Toxicology) served on the editorial boards of *Biology of Reproduction* and *Molecular Reproduction and Development.*

Dr. Stavros Garantziotis (Office of Clinical Research) was elected to the editorial board of the *American Journal of Respiratory Cell Molecular Biology.*

Dr. Joyce Goldstein (Laboratory of Pharmacology) served on the editorial boards of *Drug Metabolism and Disposition* and *Drug Metabolism Reviews.*

Dr. Dmitry Gordenin (Laboratory of Molecular Genetics) served on the editorial board of *Mutation Research, Fundamental and Molecular Mechanisms of Mutagenesis*.

Dr. David Malarkey (Cellular and Molecular Pathology Branch) served as an associated editor of the *Journal of Toxicologic Pathology.*

Dr. Fredrick Miller (Office of Clinical Research) served on the editorial board of *The Open Rheumatology Journal.*

Dr. Richard Paules (Laboratory of Molecular Toxicology) an Associate Editor of the journal *Physiological Genomics.*

Dr. John B. Pritchard (Acting Scientific Director) served on the editorial boards of *American Journal of Physiology: Renal Physiology* and *American Journal of Physiology: Regulatory, Integrative, and Comparative Physiology.*

Dr. Lisa Rider (Office of Clinical Research) served on the editorial board of *The Open Rheumatology Journal*.

Dr. Samuel Wilson (Laboratory of Structural Biology) served as an Associate Editor of the journal *DNA Repair* and as on the editorial board of *Nucleic Acids Research.*

Dr. Jerrel Yakel (Laboratory of Neurobiology) served as senior editor of the *Journal of Physiology.*

Dr. Darryl Zeldin (Acting Clinical Director; Laboratory of Respiratory Biology) served on the editorial boards of the *Journal of Biological Chemistry*, the *American Journal of Physiology: Lung Cellular and Molecular Biology,* the *Journal of Allergy and Clinical Immunology, Prostaglandins and Other Lipid Mediators,* and *Molecular and Cellular Pharmacology.*