THE FRIEDMAN BRAIN INSTITUTE

MOUNT SINAI SCHOOL OF MEDICINE

The Friedman Brain Institute is one of the world's premier institutions dedicated to advancing our understanding of brain and spinal cord disorders, and driving innovative approaches to new treatments and diagnostic tests, through translational research.

FALL 2011

INSIDE THIS ISSUE

The last decade has seen dramatic advances in our understanding of the molecular mechanisms by which genes and environment interact. This new field of biology, epigenetics, describes the intricate and highly complex processes by which the 20,000 or so genes in our bodies are controlled throughout the life cycle. During development, the selective turning on or off of collections of genes govern the differentiation of stem cells into organs, and within the nervous system, determine the generation of 100 billion nerve cells that comprise a human brain and the hundreds of trillion precise synapses among them. Throughout this process, an individual's genes set the template or capacity, while the environment modifies the manner and extent to which that capacity is achieved. We now know that such modulation involves alterations in interactions between DNA and the thousands of proteins in a cell nucleus, which together form chromatin, the material contained in cell nuclei, as described in *Epigenetic Regulation* on page 2.

Such epigenetic mechanisms not only control the formation of the nervous system during development, but continue to mediate adaptation throughout life. An individual's ability to learn and adapt over time, but also his or her vulnerability for a wide range of disease states, involve regulation of chromatin. Such knowledge is revolutionizing our understanding of normal and pathological brain function and promises to fundamentally advance our ability to develop new treatments.

Mount Sinai is at the forefront of epigenetic research of the nervous system. This issue highlights two of the areas where epigenetic analyses have yielded important new insight into disease pathogenesis and potential therapies: work by **Patrizia Casaccia, MD, PhD**, in myelin disorders, and by **Yasmin Hurd, PhD**, in drug addiction.

WWW.MOUNTSINAI.ORG/FBI

Eric J. Nestler, MD, PhD
Nash Family Professor and Chair,
Department of Neuroscience,
Director, The Friedman Brain Institute
eric.nestler@mssm.edu

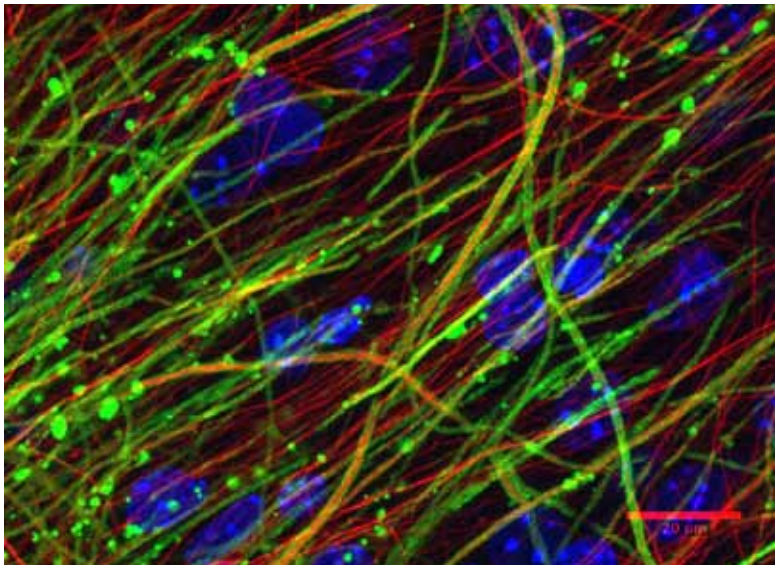


Figure 1. Myelination in brain: The image shows non-myelinated (red) and myelinated (green) axons in the developing mouse brain. Cell nuclei are labeled in blue.

MEDICAL MILESTONES

Epigenetic Approaches to Myelin Biology

Nerve cells (or neurons) form connections with other neurons by extending long processes called axons. Most axons in the brain and spinal cord are surrounded by myelin sheaths, which play crucial roles in the communication among neurons and in their survival.

Myelin pathology, once confined to the field of autoimmune demyelinating disorders, has gained increased attention in recent years, and is implicated in a wide variety of neurological and psychiatric disorders, such as neonatal ventricular leukomalacia, schizophrenia, and inflammatory demyelination as seen in multiple sclerosis. A progressive decrease in myelinated fibers is also seen within the aged human brain and is associated with declining cognitive function. Thus, myelin repair represents an important challenge across all age groups from neonates to elderly individuals, with the identification of new strategies to enhance myelination being relevant not only for specific pathologies, but also for global health in the overall population.

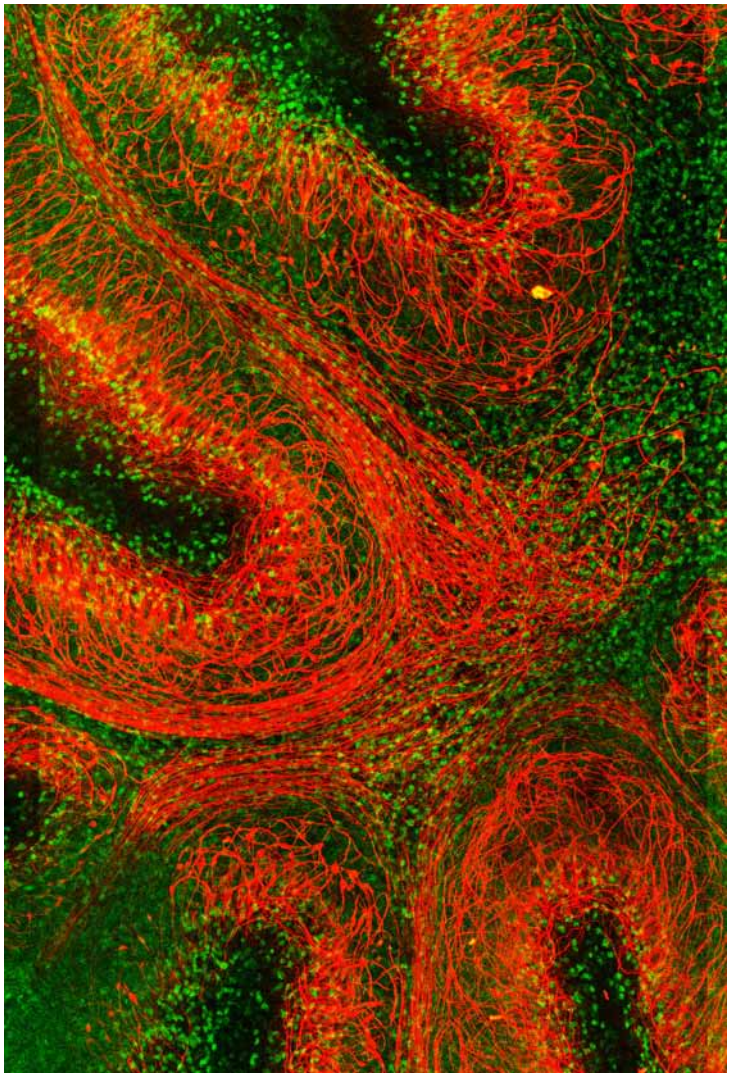
continued on page 2

Epigenetic Approaches to Myelin Biology, continued from page 1

Over the past 12 years, **Patrizia Casaccia, MD, PhD**, Professor of Neuroscience, Genetics and Genomic Sciences, and Neurology, and her colleagues have been working toward a better understanding of the molecular factors that control the health and function of oligodendrocytes, the cell type that makes myelin. Recent research from her laboratory has demonstrated a key role for epigenetic mechanisms, advances that are now paving the way for new treatments.

Early studies using electron microscopy showed that the progression of immature oligodendrocyte progenitor cells toward mature myelin-producing cells is characterized by increased chromatin compaction. In recent groundbreaking publications, the Casaccia laboratory showed that this change in chromatin compaction is mediated by a process called *histone deacetylation*, where acetyl groups are removed from histone proteins that bind DNA, leading to a more repressed (compact) state of chromatin (see *Epigenetic Regulation* below). Immature oligodendrocytes are characterized by high levels of histone acetylation, which drives high levels of expression of genes whose protein products inhibit cell differentiation. As levels of histone acetylation decrease, expression of these inhibitory genes decreases, resulting in the onset of differentiation and eventually myelin formation. The acetylation state of histones is controlled by the equilibrium between two types of enzyme, histone acetyltransferases (HATs), responsible for adding acetyl groups, and histone deacetylases (HDACs) that catalyze the removal of the acetyl groups.

Figure 2. Confocal image of a cerebellar slice in culture: The slice was stained with antibodies specific for neurofilament proteins (a neuronal marker; red) and for histone deacetylase (HDAC1; green) that labels neuronal and glial nuclei. This explant retains all the structural characteristics and cellular networks seen in the intact brain and can be kept in culture for several weeks. It provides an ideal system in which to screen novel small molecules, targeting epigenetic or other mechanisms, for their effects on myelination and neuronal integrity.



In studies involving rats and mice, Dr. Casaccia and colleagues revealed that promoting histone acetylation (by increasing activity of HATs or decreasing activity of HDACs) impairs myelination during development, as well as the ability of the animal to repair myelin after demyelination in multiple sclerosis models. Conversely, experimentally promoting histone deacetylation retards demyelination, enhances myelin repair, and limits nerve injury that occurs secondary to demyelination. Interestingly, high levels of histone acetylation were found in biopsies obtained from humans with multiple sclerosis, supporting the findings in animals. The group also demonstrated elevated histone acetylation in oligodendrocytes isolated from the brains of aged rodents, suggesting general applicability of this finding to other conditions of impaired myelination.

This accumulating evidence provides a novel path for the development of new treatments for myelin-related disorders. HAT inhibitors would be one approach, however the brain contains large numbers of HATs, which are highly similar and difficult to target individually with pharmacological agents. The Casaccia laboratory is taking a different approach, searching for small molecules that antagonize the binding of particular HATs to acetylated histones. Such molecules would promote oligodendrocyte progenitor differentiation and thereby promote myelin formation and repair. Although in the early stages of development, this work, carried out in collaboration with **Ming-Ming Zhou, PhD**, Professor and Chair of the Department of Chemical and Structural Biology, illustrates the therapeutic potential of research at the epigenetic level.

Patrizia Casaccia, MD, PhD
Professor of Neuroscience, Genetics and Genomic Sciences, and Neurology
patrizia.casaccia@mssm.edu

Ming-Ming Zhou, PhD
Professor and Chair of the Department of Chemical and Structural Biology
ming-ming.zhou@mssm.edu

Epigenetic Regulation

Every cell in the body contains roughly 3 billion nucleotides of DNA, which—if stretched out linearly—would be about two meters long. Yet, somehow this DNA is condensed to fit into a microscopic cell nucleus. The field of epigenetics has demonstrated the precise molecular mechanisms by which this extraordinary compaction occurs and is continually regulated during development and throughout adult life to control gene expression, and consequently, a range of adaptive and maladaptive changes in all organs, including the nervous system.

The basic unit of chromatin (the material in a cell nucleus) is the nucleosome, which is composed of DNA wrapped around histone proteins. The structure and function of chromatin is regulated by post-translational modifications of amino acid residues within the N-terminal tails of histones, which project from the nucleosome, as well as post-translational modifications of the DNA itself. The

Prenatal Effects of Marijuana: Epigenetic Mechanisms

Drug addiction is strongly interrelated with neurodevelopment: the initiation of drug use normally begins during adolescence, and addiction susceptibility is influenced by adverse early life events, such as developmental exposure to drugs of abuse. Marijuana (*Cannabis sativa*) is the illicit drug most commonly abused by pregnant women in the United States, and a growing body of evidence suggests that maternal cannabis use can have long-lasting negative consequences on the cannabis-exposed fetus, including increased risk for developing drug addiction and other neuropsychiatric disorders later in life. Understanding the biological basis by which drug exposure early in life promotes this risk for illness is clearly a major public health imperative.

The laboratory of **Yasmin Hurd, PhD**, Professor of Pharmacology and Systems Therapeutics, Psychiatry, and Neuroscience, has approached this challenge by assessing the neurobiological disturbances induced in the human fetal brain in association with drug exposure. The research revealed selective disturbance of genes relevant to reward, motivation, and emotional regulation in limbic-related brain regions of human fetuses with *in utero* exposure to cannabis. In particular, cannabis disrupts dopamine D2 receptor gene regulation in the nucleus accumbens, a key reward region. This finding is highly relevant to addiction disorders: based on brain imaging studies, D2 receptor impairment is characteristic of adult drug addicts. Dr. Hurd and colleagues subsequently observed similar impaired expression of the D2 receptor gene in an animal model in which rats were exposed to cannabis during prenatal development. This D2 receptor impairment persisted into adulthood and was associated with enhanced vulnerability to drugs of abuse: rats exposed to cannabis *in utero* self-administered more heroin as adults, particularly under stressful conditions, and showed enhanced opiate reward.

How does exposure to cannabis *in utero* cause a lifelong change in levels of expression of the D2 receptor gene? Recent studies from the Hurd laboratory have demonstrated the involvement of epigenetic mechanisms. Histones surrounding the D2 gene show increased methylation at particular sites (see *Epigenetic Regulation* on page 2) that are known to repress gene expression as a consequence of cannabis exposure, a change that occurs selectively in the nucleus accumbens. Abnormal histone methylation was also observed at several other genes known to be important in reward and addiction mechanisms. Such chromatin modifications establish the important role of epigenetic regulation in mediating the protracted effects of developmental cannabis exposure on neuronal systems relevant to emotional regulation and critical for drug abuse vulnerability in adulthood.

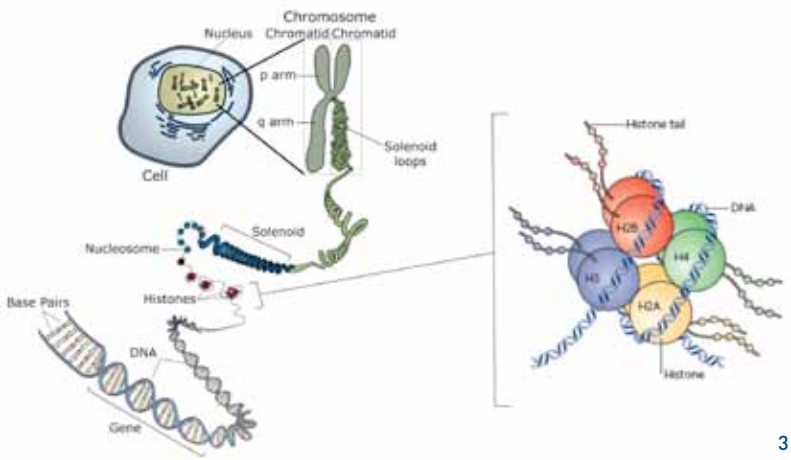
Research in other Mount Sinai laboratories is also contributing to our understanding of epigenetic mechanisms of drug addiction. The laboratories of **Eric J. Nestler, MD, PhD**, Nash Family Professor and Chair of the Department of Neuroscience, and Director of The Friedman Brain Institute, and **Scott J. Russo, PhD**, Assistant Professor of Neuroscience, are using state-of-the-art chromatin and DNA sequencing tools to identify all of the genes in limbic brain regions that are altered at the chromatin level after exposure to a range of drugs of abuse. Together, this research is driving critical approaches to better understanding the biological basis of addiction so that more effective treatments can be developed.

Yasmin Hurd, PhD
Professor of Pharmacology and Systems Therapeudics, Psychiatry, and Neuroscience
yasmin.hurd@mssm.edu

Scott J. Russo, PhD
Assistant Professor of Neuroscience
scott.russo@mssm.edu

best-characterized modifications are acetylation and methylation of lysine residues on histones. Acetylation promotes increased spacing between nucleosomes, and hence, increased gene expression. Methylation can similarly promote gene expression, or induce chromatin compaction and decreased gene expression, depending on the specific lysine affected. DNA methylation likewise promotes chromatin compaction and gene repression. Many hundreds of proteins have been delineated that control these and related processes of chromatin modification, which highlights the astounding complexity of epigenetic regulation.

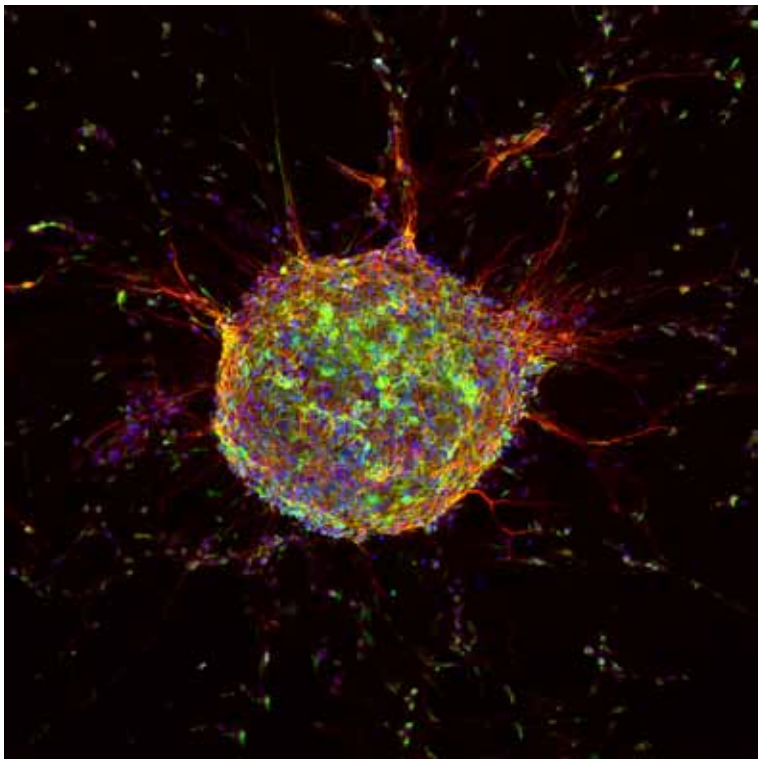
The Friedman Brain Institute has become a leader in discovering how epigenetic mechanisms control nervous system function under normal conditions, contribute to the pathogenesis of diverse disease states, and can be exploited for new therapeutics.



MOUNT SINAI SCHOOL OF MEDICINE
THE FRIEDMAN BRAIN INSTITUTE
ONE GUSTAVE L. LEVY PLACE, BOX 1107
NEW YORK, NY 10029-6574

PHOTO ESSAY

Neural Stem Cells in Culture



Neural stem cells, isolated from fetal mouse brain, aggregate into a spherical structure called a neurosphere when grown in cell culture. The cells are capable of differentiating into mature neurons, as shown by immunostaining for MAP2 (microtubule-associated protein-2), a neuronal marker (red), when grown under particular conditions. Epigenetic mechanisms control this differentiation. Understanding the differentiation and growth of stem cells is being pursued as a repair strategy for spinal cord and brain injury. The blue color stains for DAPI, a marker of cell nuclei, and the green color stains for green fluorescent protein, which marks the cell bodies in the neurosphere.

Hongyan (Jenny) Zou, MD, PhD
Assistant Professor of
Neurosurgery, and Neuroscience
hongyan.zou@mountsinai.org