ICE

A newsletter about faculty and activities at the Institute for Cell Engineering at The Johns Hopkins University School of Medicine

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JOHNS HOPKINS MEDICINE

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Playing Policymaker for a Day

Senator Barbara A. Mikulski visits ICE to learn about the latest in stem cell research.

On April 10, Institute for Cell Engineering researchers traded in pipettes for a pulpit. On the eve of the U.S. Senate vote on the Stem Cell Research Enhancement Act of 2007, which would expand federal stem cell funding to include new cell lines created from discarded embryos at fertility clinics, Maryland senator and hometown girl Barbara Mikulski visited ICE to learn firsthand how loosening current restrictions could help research move forward.

"Tell me what you are doing and tell me what impedes you now, working under the Bush framework," she said, referring to the restrictions President Bush enacted in 2001 that permit federal funding to be used only for pre-existing cell lines, many of which are defective or unsuitable for certain types of research.

Loosening funding restrictions would allow us to do what we do best at Hopkins—work together.

The senator got an earful—of both good and bad.

Hopkins, known for its collegial and collaborative atmosphere, has felt the cramping effects of having to separate equipment and work funded by federal dollars from that of philanthropy. Like having to stock double sets of dishes in a kosher kitchen to separate meat from dairy, researchers often need to buy two pieces of the same equipment, or worse, keep talented, federally funded staff separated from privately funded projects.

"The fundamental basis of Hopkins, from the basic research to the clinical treatments, is our ability to collaborate," said Valina Dawson, professor of neurology, neuroscience and physiology, and co-director of Neuro-ICE, whose own research focuses on whether stem cells can restore brain function in stroke patients and those with Parkinson's disease. "Loosening funding restrictions would allow us to do what we do best at Hopkins—work together."

In her statement to the Senate, Senator Mikulski shared with her colleagues that the current Balkanizing of scientific resources "goes against the way science is studied," and is "slowing down potential breakthroughs."

Despite the impediments, however, ICE has been blessed with generous philanthropic donations and the sharp minds to put those donations to good use [see Spinal Sleuth on next page].

As for ICE's newfound role as policy advocates, Jeffrey Rothstein, neurology professor and director of the Robert Packard Center for ALS Research, admits that "politics and research don't mesh well," but his patients drive him to speak up, for their sake. "In health, there are no sides—we must do what's good for the patient." ■

Senator Mikulski learns about the promises and pitfalls of stem cell research at Hopkins.

Hopkins Researchers Awarded State Research Funds

On May 17, the Maryland Stem Cell Research Commission recommended 24 projects from a total of 86 applications for funding under the Maryland Stem Cell Research Act of 2006. Fifteen recipients are at Johns Hopkins, including four members of ICE:

Angelo All
Whitaker Biomedical Engineering Institute
"Human Embryonic Stem Cell-Derived Oligodendrocytes and Electrophysiological Studies in a Contusion Model of Spinal Cord Injury in the Rat"

Shyam Biswal
Bloomberg School of Public Health
"Nrf2 as a Target for Cancer Stem Cell Chemoresistance"

Jeff Bulte
School of Medicine and ICE
"Human Embryonic Stem Cell-Derived Neurospheres for Treatment of Multiple Sclerosis"

Srinivasan Chandrasegaran
Bloomberg School of Public Health
"Targeted Engineering of the Human Genome in Stem Cells"

Curt Civin
Kimmel Cancer Center
"MicroRNA Regulation of Adult and Embryonic Human Hematopoietic Development"

Nancy Craig
School of Medicine
"Genome Engineering of Human Stem Cells for Gene Therapy"

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Spinal Sleuth

Uncovering clues to help solve the mysteries of motor neuron diseases.

Douglas Kerr is hot on the trail of four deadly diseases.

Douglas Kerr and his research team are unraveling a series of real-life murder mysteries. The victims are motor neurons and neural myelinating cells, and the culprits are amyotrophic lateral sclerosis (ALS), transverse myelitis (TM), spinal muscular atrophy (SMA) and multiple sclerosis (MS). The team has pursued these villains with a dedication worthy of Sherlock Holmes and has been so successful that clinicians might be using their discoveries within five years.

ALS, or Lou Gehrig's disease, was a particularly tough case to crack. "The ALS genetic mutation fills the nervous system with many different inflammatory agents that kill motor neurons," says Kerr, whose team created a cocktail that blocks all the agents at once. "Blocking one or two just didn't halt the disease process." The cocktail is headed for clinical trials as soon as its safety guidelines are determined.

The second culprit is a ticking time bomb. TM, an uncommon inflammation of the spine, occurs when the immune system pumps out excess interleukin-6 (IL-6) in response to stress. The flood of IL-6 leads to rapid limb weakness, sensory disturbances and back pain.

"Time is of the essence with transverse myelitis," says Kerr, who has set up the world's only TM clinic here at Hopkins. "Aggressive treatment during the first three months is critical for better recovery; after three months, recovery slows down." Referring physicians racing against the clock can phone in for weekly clinical conferences or request video telemedicine appointments.

The ALS genetic mutation fills the nervous system with many different inflammatory agents that kill motor neurons.

Two effective treatments, Kerr discovered, are the blood hormone erythropoietin and the anti-inflammatory drug thalidomide. A clinical trial of erythropoietin will begin in the next couple of months, and a trial of thalidomide is planned for the near future. Since both drugs are FDA approved, the therapies could reach the clinic as soon as 18 months after trials start.

Both ALS and SMA are in for murder charges—both destroy motor neurons and cause patients to have floppy, weak muscles. Babies with SMA can die if they can't suckle properly.

SMA is caused by a deficiency of a specific protein, whereas MS kills the protective nerve coat myelin with inflammatory agents. Potential treatments for both, according to Kerr, lie in the promise of embryonic stem cells.

Kerr's team has injected embryonic stem cells into rats bred to have SMA. The injected cells grew into motor neurons, reconnected the rats' muscles to spinal cords, and restored muscle control and some movement. Kerr is chomping at the bit to bring this approach to infants with SMA, but currently that research is stalled, awaiting further funding since federal grants cannot be used for this research.

Kerr has been equally successful in treating MS in animals. His team has coaxed fetal glial stem cells to replace the damaged myelin coating on nerves. They have also discovered that growing the glial cells with a novel nerve protein protects these cells from the inflammatory assault of MS.

Unfortunately, the treatments for SMA and MS may not reach the bedside as soon as Kerr or patients would like. "Every member of my team is passionate about improving human health," he says. "We're a lot closer than people may think, but it would be a shame if we can't make it all the way to the bedside." ■

State Research Funds
(Continued from page 1)

Candace Kerr
School of Medicine and ICE
"Defining Genes Associated with Human Stem Cell Pluripotency and Self-Renewal"

Hai-Quan Mao
Whiting School of Engineering
"Engineering an Artificial Neural Stem Cell Niche"

Andrew McCallion
School of Medicine
"Illuminating Human Cardiac Development and Disease through Transcriptional Analysis in Differentiating Human Embryonic Stem Cells"

Guo-li Ming
School of Medicine and ICE
"Mechanisms Regulating Self Renewal of Human Embryonic Stem Cells"

Akhilesh Pandey
School of Medicine
"Proteomic Characterization of Neural Differentiation in Human Embryonic Stem Cells"

Hamid Rabb
School of Medicine
"Isolation, Expansion and Regenerative Potential of Human Adult Kidney-Derived Stem Cells"

Hongjun Song
School of Medicine and ICE
"Characterization of Neuronal Potentials of Human Embryonic Stem and Adult Neural Stem Cells"

Elias Zambidis
Kimmel Cancer Center
"Human Embryonic Stem Cell Models of Normal and Leukemic Human Stem Cells"

Karen Zeller
School of Medicine
"Myc's Role in Maintenance and Tumorigenicity of Human Embryonic Stem Cells"

A Different Kind of Fish Story

New zebrafish facility will aid understanding of basic cell development.

Don't let the size of the 1-inch, semitransparent specimens fool you—these little zebrafish are big players in the world of cell development. So big, in fact, that ICE recently has established an 1,800-tank zebrafish core facility to share the power of fish with the entire Hopkins research community.

While hardly the first thing that comes to mind when one mentions the words "stem cell biology," these tiny swimmers are more useful in helping uncover how cells choose their fates than one might initially think.

Zebrafish eggs are huge by cell standards (1 to 2 millimeters in diameter), and the growing embryo is completely transparent and hatches into a fingerling only five days after fertilization. Also, one female fish can lay as many as 1,000 eggs at a time.

Shannon Fisher and Andy McCallion check on their tiny charges.

"Mice are still the gold standard for functional genetics," says facility co-director Shannon Fisher, assistant professor of genetic medicine. "But it takes three weeks for a mouse to have a litter of only six to eight pups," which is too slow and too expensive for her own research.

Fisher and fellow geneticist and facility co-director Andy McCallion study the nonprotein coding regions of genomes. Referred to by some as "junk DNA," these noncoding areas are receiving increased scrutiny of late, and the current theory, explains McCallion, is that they frequently dictate the timing, location and quantity of gene expression.

But noncoding regions make up more than 98 percent of the 3 billion DNA building blocks of the human genome—that's a lot of DNA to search through and a timely and expensive endeavor to pursue in mice or any other mammal.

To solve that problem, Fisher and McCallion developed a molecular tool that allows researchers to insert any segment of human DNA into zebrafish embryos to find out where in the body and at what time during development that gene might be turned on.

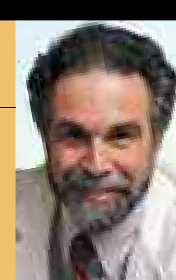
Using fish to streamline research, Fisher explains, will benefit the entire stem cell community, which aims to understand how stem cells transform themselves from undifferentiated precursors to specialized cell types. "The new facility will allow us and others to ask functional and early-stage questions about all aspects of the genome more rapidly and less expensively than ever before," says Fisher. "And we can use those answers to design more time- and cost-effective mouse experiments."

"The more we understand normal development in the embryo, the better we can direct development of stem cells in a culture dish," says McCallion. "All cells are a product of all the instructions they receive from their cellular and extracellular environment during development. What we are trying to do is recreate those instructions outside the body." ■

Conversation

Turn Up the Air

Gregg Semenza has discovered a new therapeutic target for cancer and cardiovascular disease: our cells' oxygen sensor.



Q: How exactly do our cells adapt to changing levels in oxygen?

Semenza: At optimal oxygen levels, cells make energy by shuttling energy-rich electrons through a series of protein relay stations in their mitochondria called cytochromes. The last station, cytochrome c oxidase, or COX, uses oxygen to convert the flow of electrons into ATP, the cell's energy currency.

If oxygen levels in the cell drop, COX doesn't work as well and the cell activates a protein called HIF-1, which directs the cell to replace one of the 13 subunits of COX with another protein that allows COX to function more efficiently under low oxygen conditions. When oxygen levels become even lower, HIF-1 directs the cell to bypass the mitochondria and generate ATP by fermentation, which does not require oxygen at all.

Q: And as it turns out, hypoxia and HIF-1 play a pivotal role in three major diseases, namely cancer, heart attacks and strokes, right?

Correct; heart attacks and strokes occur when a blood vessel becomes blocked, through atherosclerotic build-up or a blood clot. Cells on the other side of the blockage can die because their blood supply is cut off. We've discovered that HIF-1 directs the remodeling of other blood vessels to help restore blood flow. Cancer cells also use HIF-1 to stimulate blood vessel growth.

Q: And what therapeutic avenues does that knowledge now open?

Reducing the amount of HIF-1 in cancer cells may block their growth by preventing both blood vessel formation and changes in energy metabolism that allow the cells to survive under low oxygen conditions.

On the other hand, in tissues that are not getting enough blood flow due to atherosclerosis, such as in the hearts of patients with coronary artery disease or the legs of patients with limb ischemia, we want to increase the amount of HIF-1 to help oxygen-starved cells survive and to stimulate the production of new blood vessels. If these strategies work, they could improve the survival of patients with cardiovascular disease and cancer, which are the major causes of death in the United States. ■

Streamlining Safe Research

Chi V. Dang introduces a few new resources designed to coordinate an amorphous science.

Q: What is the new Stem Cell Resource Center (SCRC)?
The SCRC is a completely new concept in the field, as far as we know. It's an institutional center designed to make stem cell research safer and easier in several different ways. It is a unique ICE initiative, but it will underpin all stem cell research for Johns Hopkins.

Q: What will the SCRC do?
It will initiate and coordinate emerging technology platforms to efficiently and rapidly identify conditions for using human stem cell therapy in patients. For example, it will make a head-to-head comparison of the existing stem cell lines, fully describe them, and check their chromosomes for signs of the changes that accumulate in all living cells as they encounter environment and age. The Center also will find ways to mark the cells so they can be followed as they grow and develop through time. Such centralized services will bring a stunning level of confidence and security to stem cell research at Hopkins.

Q: Why centralize? Can't every researcher do these things as needed?
Centralized services save money and time. Many Hopkins researchers would like to venture into stem cell science but don't because of the immense bureaucratic burden of paperwork required to gain access to individual cell lines. The SCRC will do all that for the entire University, so that individual investigators will basically have free access. The SCRC will be a "one-stop shop" to preserve, create, supply and test high-quality cell lines for our own researchers now, and later for the greater scientific community. We are immensely proud of it and hope it will serve as a model for other institutions so everyone's stem cell research will be as safe and easy as possible.

Q: Will the SCRC use only the stem cell lines approved by President Bush?
No, we can't use any of those lines for research that will end up in human trials because human embryonic stem cell lines created before 9 p.m. EST, Aug. 9, 2001, were initially grown on mouse cells and therefore might harbor mouse-specific viruses. We will use lines derived from extra human embryos from fertility clinics, which is legal under Maryland law. We can't use federal funds to do this, but we can use Maryland state funds or funds from private donors, and Hopkins received a significant anonymous gift earlier this year to set up the SCRC and obtain the cell lines we need.

Q: What else will the SCRC do?
One of the most important initiatives is the creation of a nine-person embryonic stem cell research oversight (ESCRO) committee to make sure that all human embryonic stem cell experiments conducted at the University are safe. ESCRO will function like our current institutional review boards that monitor the safety of any research involving human subjects, but it will be modeled on the 2005 guidelines formulated by the National Academy of Sciences. ■

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