Inside This Issue

Chair’s Message

Faculty Update
Accomplishments and Recognition

Research Spotlight
Tools of Neuroscience Research: Imaging and Modeling

Department News
Department of Neurosurgery conducts retreat
Departments of Neurology and Neurosurgery hold holiday party
Contributor Acknowledgement

Residents’/Fellows’ Corner
Spine Fellowship update

Presentations/Publications
July-December 2007

Conference Schedule
February-July 2008

Upcoming Meetings
January-June 2008

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Research Spotlight: Tools of Neuroscience Research: Imaging and Modeling
Welcome to the Winter 2008 issue of our departmental newsletter Neuroscience Outlook. In this issue we focus on some of the modern tools we are utilizing in the field of neuroscience research in the Synapese lab, the Cerebrovascular lab, and the Human Brain lab. These include a variety of imaging techniques in live animal models, specimens, and individual cells. These techniques and others to be elucidated in upcoming issues of the Outlook, will be critically important in our quest to advance translational research.

Since our last issue, we have had some notable events such as our departmental retreat. This served to bring all the clinicians, researchers, physician extenders and key nursing staff together for a day to assess our progress as a department and to plan for the future. Also in this issue, we highlight notable achievements by our faculty and we chronicle the academic productivity of the department over the past 6 months.

Lastly, we are thankful for the varied donations and funding received. As funding and reimbursement from the typical sources continue to shrink, the charitable donations from individuals and organizations take on a much greater importance. We invite alumni and other well-wishers to make a tax-deductible donation to the Neurosurgery Foundation, the Neurosurgery Resident Education Fund, the Neurosurgery Book Fund or the Marshall Allen Lectureship Endowment at the Medical College of Georgia. A self addressed envelope is conveniently provided. We hope you enjoy this issue.

Cargill H. Alleyne, Jr., M.D.
Associate Professor and Allen Distinguished Chair
Director, Cerebrovascular Service
Residency Program Director
Department of Neurosurgery

Faculty Update

Accomplishments and Recognition

John R. Vender, M.D. served as President of the Georgia Neurosurgical Society at the Fall Meeting of the Georgia Neurosurgical Society in Atlanta, GA in November 2007.

S. Dion Macomson, M.D., was named the Assistant Residency Program Director for the Department of Neurosurgery. Dr. Macomson will assist Dr. Alleyne in his duties as Program Director.

Haroon F. Choudhri, M.D. was the Program Chair for the Fall Meeting of the Georgia Neurosurgical Society in Atlanta, GA in November 2007. Dr. Choudhri was also Visiting Professor at King Fahd Hospital, in Jeddah, Saudi Arabia and at the Saudi National Guard Hospital in King Khalid Medical City, Saudi Arabia, in September 2007.

Cargill H. Alleyne, Jr., M.D. was officially honored at the inaugural Chair Investiture ceremony during which Dr. Norman Chutkan of Orthopedic surgery and Dr. Ana Murphy of Obstetrics and Gynecology were also honored. Dr. Alleyne was also a guest examiner at the Neurosurgical Oral Board Examinations in Houston, Texas from November 6th to 9th, 2007. Additionally, Dr. Alleyne was a member of the Scientific Program Committee of the Congress of Neurological Surgeons in San Diego, CA in October 2007.

Michael A. Jensen, M.S. passed the written portion of the Certified Medical Illustrators examination in November 2007. This examination represents the highest recognition of competence in the field of medical illustration. It tests broad knowledge of science, anatomy and medical conditions and also tests terminology, competency in complex drawing problems, and mastery of ethics and business practices. We congratulate Mike on this impressive achievement.

Joseph R. Smith, M.D. was Visiting Professor at the Department of Neurosurgery of University of Wisconsin, Madison, WI, from October 9th to 12th, 2007.
Brain slices are one of the most important in vitro model systems for studying brain function. Slices are made by quickly cutting live brain tissue to 350-500 µm thickness. In recent years the wide availability of transgenic mice expressing fluorescent proteins of different colors in a subset of neurons and glia greatly facilitated live imaging with 2PLSM (Fig. 1a,b,c). Brain slices contain fully-arborized neurons that can be imaged deep to the cut surface where the native tissue architecture and cellular milieu is preserved. They also provide an experimental model to test potentially useful therapeutics because the preparation lacks the blood-brain barrier that must be bypassed in vivo for drug delivery to injured CNS.

Imaging in vitro
Brain slices are one of the most important in vitro model systems for studying brain function. Slices are made by quickly cutting live brain tissue to 350-500 µm thickness. In recent years the wide availability of transgenic mice expressing fluorescent proteins of different colors in a subset of neurons and glia greatly facilitated live imaging with 2PLSM (Fig. 1a,b,c). Brain slices contain fully-arborized neurons that can be imaged deep to the cut surface where the native tissue architecture and cellular milieu is preserved. Brain slices permit rapid alteration of the bathing media, discrete stimulation of synaptic pathways and resolution of single living neurons (Fig. 1b) and astrocytes (Fig. 1c). They also provide an experimental model to test potentially useful therapeutics because the preparation lacks the blood-brain barrier that must be bypassed in vivo for drug delivery to injured CNS. In experi-
ments involving brain tissue from non-fluorescent transgenic animals or human tissue fluorescent dye can be loaded using a patch pipette (Figure 2a, b, c).

**Imaging in vivo**
The benefits of 2PLSM are numerous, and one advantage of this methodology is deep tissue submicron non-contact and non-invasive imaging. When combined with the inherent high temporal, spatial and three-dimensional resolution, it makes 2PLSM the technique of choice for *in vivo* acute and long-term imaging through a cranial window. Simultaneous imaging of neurons, astrocytes and blood vessels (Figures 3, 4) permits studies that were impossible just few years ago. The use of this methodology will quickly add to our knowledge of stroke and trauma associated neuronal (Figures 5a, b) and astroglial injury. *In vivo* imaging with 2PLSM will help to understand how brain cells are damaged, how they recover and ways to aid their recovery. This technology should greatly facilitate discovery of therapeutic targets to treat pathological outcomes of brain injury.

**Tools of Neuroscience Research: Live Animal Imaging**
The Small Animal Imaging Facility at MCG is a self-contained lab for conducting MRI and optical imaging research, capable of supporting murine and rodent clinical models as well as basic methodological imaging research. The facility is directed by Dr. Tom Hu, who has an established track record in the use of molecular and cellular MR contrast agents to investigate cardiovascular related disease area(s). Other personnel working closely with Dr. Hu include Dr. Nathan Yanasak, who has experience in clinical and murine brain imaging, two graduate students who are pursuing cardiac studies, and three staff members who have expertise in performing small animal cardiac surgery and optical image. The facility consists of several resources:

1) **A Bruker BioSpec 7 Tesla MRI machine** in a fully-shielded room, with detection coils and other apparatus for performing 1H, 31P, and 13C MR imaging and spectroscopy on mice and rats.
2) **A Xenogen optical system**, for imaging various fluorescent and biolumi-
Research Spotlight (continued)

Subarachnoid hemorrhage model
Ventral side of the mouse brain following sham surgery (left) or subarachnoid hemorrhage (right). We use this model to study the molecular and cellular sequelae of SAH to better understand the disease process and to identify novel therapeutics which may be used to treat patients in the future.

Brain injury model
Immunofluorescent image of (Figure 7) glial fibrillary acidic protein (GFAP), a specific marker for astrocytes, in the mouse cerebral cortex following brain injury. By 24 hours post-injury, astrocytes appear damaged (see rounded cells). An improved understanding of the role of astrocytes following neurological injuries may provide novel therapeutic targets.

Tools of Neuroscience Research: Tissue Modeling

Brain tumor model
Detection of membrane damage in human glioblastoma cells following treatment with a novel therapeutic compound. Control cells (Figure 8a) shown no accumulation of propidium iodide whereas low (8b) and high (8c) concentrations show a progressive accumulation of dye (see red color).

Vascular injury model
The hemoglobin oxidation product, hemin, induces cell death in cerebral microvessel cells. This cell culture model (Figures 9a, b) is utilized to better understand the molecular pathogenesis of vascular injury following a brain hemorrhage.

Sergei A. Kirov, Ph.D. and Krishnan Dhandapani, Ph.D.
Department News

Department of Neurosurgery conducts retreat

On October 5th, 2007 the Neurosurgery Department held its first retreat. This took place at the MCG Alumni Center in Augusta. The event was well attended and well received, and provided an opportunity for us to take stock of our five-year history as a full Department and to plan for the future. The Dean of the Medical School, Dr. Doug Miller was present to give some remarks. The full program is detailed here:

Departments of Neurology and Neurosurgery hold holiday party

Our annual Neurology and Neurosurgery Holiday party was held on December 14, 2007 at the Clubhouse in Augusta, Georgia. A good time was had by all!

Contributor acknowledgement

We are honored to accept a donation of several dozen books in the Classics of Neurology and Neurosurgery series by Ernest C. Fokes, M.D. a graduate of the Neurosurgical program in 1969. These books are one of a kind, part of the limited edition of 1000 copies and carry a considerable historical value. These include classic texts such as The Pituitary Body, by Harvey Cushing; The Brain, by Walter Dandy; Anatomy of the Brain and Nerves, by Thomas Willis; Injuries of Nerves, by S. Weir Mitchell; Harvey Cushing, by John Fulton; Pathology of Cerebral Hemorrhage, by Charles Bouchard and texts by many other luminaries such as Sherrington, Cajal, Gowers, Macewen, Mayo, Osler, Head and others. We're deeply grateful to Dr. Fokes.

We also thank the following companies and individuals for their kind monetary donations:

Crosslink Orthopedics, LLC, for an educational grant that helped to defray publication costs of the previous issue of the Neuroscience Outlook.

Cyberonics, for a donation toward the retirement party for Dr. Joseph Smith which took place last July 2007.

Susanne Touchtone, an MCG employee, for her monthly contributions to the Neursurgery foundation.

Residents’ and Fellows’ Corner

Spine Fellowship update

Synthes Spine has generously supported the educational mission of our Department by providing an educational grant to fund the Complex spine fellowship directed by Dr. Haroon Choudhri. This gift will ensure the continued viability of this fellowship.
Presentations and Publications (January 2008 - June 2008)

Presentations


Choudhri H: How to Develop Spine Training Programs (Panelist). Egyptian Neurosurgical Society, Ras Sudr, Egypt, July 2007

Choudhri H: How to Develop Spine Research Programs (Panelist). Egyptian Neurosurgical Society, Ras Sudr, Egypt, July 2007

Kirov SA: Rapid synaptic and astroglial structural plasticity in mature hippocampus. Keio University, Tokyo, Japan, July 2007

Andrew RD and Kirov SA: Two-photon microscopy reveals real-time volume responses by astrocytes to osmotic and ischemic stress in cortical brain slices. Fifth International Conference of Aquaporin, Nara, Japan, July 2007

Kirov SA: Real-time neuronal and astroglial dynamics during osmotic stress and simulated stroke. Nagoya City University, Nagoya, Japan, July 2007


Kirov SA, Rischer WC, Ard DA, Andrew RD: Real-time imaging of single neurons and astrocytes during osmotic and ischemic stress. The International Symposium on Topical Problems of Biophotonics, Nizhniy Novgorod-Moscow, Russia, August 2007


Choudhri H: Spinal Instrumentation. Saudi National Guard Hospital, King Khalid Medical City, Saudi Arabia, September 2007

Choudhri H: Indications for Revision of Spinal Instrumentation, Pan Arab Spine Society, Tunis, Tunisia, October 2007

Kirov SA: Two-photon microscopic imaging of single neurons and glia deep in cortex during ischemia. The 28th Annual Southern Pharmacology Society Meeting, Augusta, GA, October 2007


Smith JR: Closed-Loop Stimulation in the Treatment of Focal Epilepsy. University of Wisconsin, Neurosurgery Grand Rounds, Madison, WI, October, 2007

Alleyne CH: Vascular access. Fundamental Critical Care Support Course (Instructor), Medical College of Georgia, November 2007


Publications


**Conference Schedule (February 2008 - July 2008)**

All grand rounds and conferences take place on Friday in the 3 West amphitheater.

<table>
<thead>
<tr>
<th>Date</th>
<th>Time</th>
<th>Event Title</th>
<th>Speaker</th>
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<td>Feb 01</td>
<td>9:00-10:00</td>
<td>Radiology Review</td>
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<td>10:00-11:00</td>
<td>Spine Conference</td>
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<td>12:00-1:00</td>
<td>Grand Rounds: Dr. Hemant Yagnick</td>
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<td>Feb 15</td>
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