UCSF DEPARTMENT OF SURGERY

27th Annual Resident Research Symposium

Friday, April 25, 2014

UCSF Toland Hall
533 Parnassus Avenue, U-142
San Francisco, CA

Clifford Y. Ko, MD, MS, MSHS, FACS
J. Englebert Dunphy Visiting Professor
Director, Division of Research and Optimal Patient Care
Director, National Surgical Quality Improvement Program
American College of Surgeons
Professor of Surgery, UCLA

UCSF Department of Surgery Education Office
Telephone: (415) 476-1239 Email: EducationOffice@ucsfmedctr.org

This event is sponsored by educational grants from the Howard Naffziger Surgery Fund
J. Englebert Dunphy, M.D.
Professor of Surgery & Chairman of the Department from 1964 to 1975.

Dr. Dunphy graduated from Harvard Medical School and completed his surgical residency training at the Peter Bent Brigham Hospital in Boston. He joined the faculty at Harvard briefly before assuming the Chairmanship of the Department of Surgery at the University of Oregon. In 1964, he became the Chairman of the Department of Surgery at the University of California, San Francisco. Dr. Dunphy was president of the Society of University Surgeons, the American Surgical Association, and the American College of Surgeons. He received honorary fellowships in six foreign colleges of surgeons as recognition of his international stature. Dr. Dunphy was renowned for excellence in many aspects of surgery, with a special interest in the gastrointestinal tract. He was one of the leading surgical educators of his day and was greatly admired and respected by his colleagues and residents. Dr. Dunphy conducted research in wound healing at a basic level.

Dr. Dunphy strongly believed that prospective academic surgeons should become grounded in basic science, and he was one of the first surgical leaders in the United States to obtain an NIH training grant to support residents in the laboratory.
Clifford Y. Ko, MD, MS, MSHS, FACS, FASCRS

J. Englebert Dunphy Visiting Professor

Dr. Clifford Ko is the Director of the Division of Research and Optimal Patient Care at the American College of Surgeons. He oversees all the quality improvement programs, including the Bariatric Surgery Accreditation Program, the Cancer Accreditation program, the Trauma Verification program, the new Surgeon Specific Registry, and the American College of Surgeons National Surgical Quality Improvement Program (ACS NSQIP). He also serves as the Director of ACS NSQIP. Dr. Ko’s work focuses on surgical quality of care, including quality measurement, process improvement, and quality maintenance. He has received millions of dollars in grant funding to study quality of care from sources that include the National Institutes of Health, the Centers for Disease Control and Prevention, the American Cancer Society, the Centers of Medicare and Medicaid Services, the Agency for Healthcare Research and Quality, and the Veterans Administration.

Clinically, Dr. Ko is a double board-certified surgeon with a practice focusing on patients with colorectal cancer. At UCLA, he won the Faculty Teaching Award three times, and is recognized as one of the Best Doctors in America. He is also professor of health services at the UCLA School of Public Health. Dr. Ko received his B.A (Biology), M.S. (Biological/Medical Ethics), and M.D. from the University of Chicago. He also received a Masters of Science Degree (Health Services/Outcomes Research) from the University of California, Los Angeles during his time as a Robert Wood Johnson Clinical Scholars Fellow at UCLA and RAND. Dr. Ko completed his General Surgery Residency at UCLA Medical Center, and obtained specialty training at the Lahey Clinic in Boston in Colon and Rectal Surgery.
Past Visiting Professors

Bernard Langer, M.D.
Professor and Chairman of Surgery, University of Toronto
February 5-6, 1988

William Silen, M.D.
Professor of Surgery, Harvard Medical School
February 3-4, 1989

James Thompson, M.D.
Professor and Chairman of Surgery, University of Texas, Galveston
February 2-3, 1990

Murray Brennan, M.D.
Professor and Chair of Surgery, Memorial Sloan-Kettering Cancer Center
February 3-4, 1991

Richard Simmons, M.D.
Professor and Chairman of Surgery, University of Pittsburgh
January 31-February 1, 1992

Stephen F. Lowry, M.D.
Professor of Surgery, Cornell University Medical College
February 4-5, 1993

Jared Diamond, Ph.D.
Professor of Physiology, UCLA School of Medicine
February 4, 1994

Samuel A. Wells, Jr., M.D.
Professor and Chairman of Surgery, Washington University
February 17, 1995

Jonathon E. Rhoads
Chief of Surgical Oncology, University of Pennsylvania, Philadelphia
February 16, 1996

Patricia K. Donahoe, M.D.
Chief, Pediatric Surgical Services, Massachusetts General Hospital
February 27, 1997

David L. Dunn, M.D., Ph.D.
Professor and Chairman of Surgery, University of Minnesota
February 27, 1998

Ori D. Rotstein, M.D.
Professor of Surgery, Toronto Hospital
February 26, 1999

Olga Jonasson, M.D.
Director of Education and Surgical Services Department
American College of Surgeons
March 17, 2000

Glenn Steele, Jr., M.D. Ph.D.
Dean, School of Medicine, University of Chicago
March 9, 2001

Alexander W. Clowes, M.D.
Professor of Surgery and Chairman, University of Michigan
March 7, 2002

Michael Mulholland, M.D., Ph.D.
Professor of Surgery and Chairman, University of Michigan
March 7, 2003

Christian Larsen, M.D., Ph.D.
Professor of Surgery, Emory University
March 19, 2004

Danny O. Jacobs, M.D., M.P.H.
Chair, Department of Surgery, Duke University Medical Center
March 4, 2005

Steven D. Leach, M.D.
Chief of Surgical Oncology, Johns Hopkins University
March 3, 2006

M. Judah Folkman, M.D.
Professor of Pediatric Surgery & Cell Biology, Harvard Medical School
Director, Vascular Biology Program, Children’s Hospital, Boston
February 15-16, 2007

Sir Peter Morris, AC, FRS, FRCS
Director, Centre for Evidence in Transplantation
Royal College of Surgeons of England
April 4, 2008

George K. Gittes, M.D.
Chair of Pediatric Surgery, University of Pittsburgh
April 3, 2009

Joseph P. Vacanti, M.D.
Chief, Pediatric Surgery, Massachusetts General Hospital
March 12, 2010

Maria Bertagnolli, M.D.
Professor of Surgery, Harvard
Chief, Surgical Oncology, Brigham and Women’s Hospital
April 1, 2011

Michael Harrison, M.D.
Director Emeritus, Fetal Treatment Center, Professor of Pediatric Surgery,
University of California, San Francisco
April 13, 2012

Martin Elliott, M.D.
Professor of Cardiothoracic Surgery, University College London
April 5, 2013
2013 Award Recipients

BEST ABSTRACT: Corticotropin-Releasing Factor (CRF) Receptor Activation Causes Necrotizing Enterocolitis (NEC) in Formula Fed, Neonatal Rats. Presented by Robert Bell, MD

OUTSTANDING ABSTRACT: Task-shifting and the Scope of Surgical Care in Pwani Region, Tanzania. Presented by Jessica Beard, MD, MPH

OUTSTANDING ABSTRACT: Hospital-Based Violence Intervention: Risk Reduction Resources That Are Essential for Success. Presented by Randi Smith, MD, MPH

BEST “QUICK-SHOT” PRESENTATION: Characterization of Human Muscle Stem Cells for Muscle Regeneration. Presented by Xiaoti Xu, MD

HONORABLE MENTION AWARDS:
Cost-Effectiveness Analysis of Intraoperative Radiation Therapy for Early-Stage Breast Cancer. Presented by Cristina O’Donoghue, MD

Generation of Induced Hepatocytes for Autologous Liver Cell Therapy. Presented by Jack Harbell, MD
### SESSION 1: "Trauma, Inflammation, Vascular & Cancer"
**Moderator:** Jade Hiramoto, MD

<table>
<thead>
<tr>
<th>Time</th>
<th>Title</th>
<th>Presenter</th>
<th>Year</th>
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<tbody>
<tr>
<td>9:05 AM</td>
<td>Physiologic and Molecular Mechanisms of Arteriovenus Fistula Maturation.</td>
<td>Joy Walker, MD</td>
<td>2nd</td>
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<tr>
<td>9:20 AM</td>
<td>The Whole is Greater than the Sum of its Parts: Hemostatic Profiles of Whole Blood Variants.</td>
<td>Lucy Kornblith, MD</td>
<td>2nd</td>
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<tr>
<td>9:35 AM</td>
<td>Total Skin Sparing Mastectomy and Immediate Tissue Expander Implant Reconstruction in the Setting of Radiation Therapy</td>
<td>Frederick Wang, MD</td>
<td>2nd</td>
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<tr>
<td>9:50 AM</td>
<td>Clinical Significance of Pneumomediastinum in Moderate to Severe Blunt Chest Trauma.</td>
<td>Wayne Lee, MD</td>
<td>1st</td>
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<tr>
<td>9:55 AM</td>
<td>Inducing Acute Traumatic Coagulopathy In Vitro.</td>
<td>Benjamin Howard, MD</td>
<td>1st</td>
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<td>10:00 AM</td>
<td>20-minute break</td>
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### SESSION 2: "Global Surgery, Public Health, Outcomes & Education"
**Moderator:** Emily Finlayson, MD

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<th>Time</th>
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<tbody>
<tr>
<td>10:20 AM</td>
<td>Depression and Peripheral Arterial Disease: Is Physiology the Link?</td>
<td>Jessica Cohan, MD</td>
<td>1st</td>
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<tr>
<td>10:25 AM</td>
<td>Quality of Trauma Care for Undocumented Latino Immigrants.</td>
<td>Vincent Chong, MD</td>
<td>1st</td>
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<tr>
<td>10:30 AM</td>
<td>Choice of Injury Scoring System in a Resource-Poor Setting: Lessons from Mumbai.</td>
<td>Adam Laytin, MD</td>
<td>1st</td>
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<tr>
<td>10:35 AM</td>
<td>Prognostic Utility of Lymph Node Size, Number, Histology and Extranodal Extension for Papillary Thyroid Cancer Nodal Metastases Following Selective Lymph Node Dissection.</td>
<td>Carolyn Seib, MD</td>
<td>2nd</td>
</tr>
<tr>
<td>10:50 AM</td>
<td>From Novice to Master Surgeon: Improving Feedback with a Descriptive Approach to Intraoperative Assessment.</td>
<td>Emily Huang, MD</td>
<td>2nd</td>
</tr>
<tr>
<td>11:05 AM</td>
<td>Establishing Prospective Trauma Metrics Evaluations for Pre-Hospital Trauma Care in Rwanda.</td>
<td>Rebecca Maine, MD, MPH</td>
<td>2nd</td>
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<tr>
<td>11:30 AM</td>
<td>Buffet Lunch (90 minutes)</td>
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**SESSION 3: "Stem Cells, Transplantation & Immunology"
Moderator: Tippi Mackenzie, MD**

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<th>Time</th>
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<tr>
<td>1:00 PM</td>
<td>Precocious T Cell Maturation In Patients With Gastroschisis.</td>
<td>Cerine Jeanty, MD 1st Year Research Resident</td>
</tr>
<tr>
<td>1:05 PM</td>
<td>Characterization and Transplantation of Human Muscle Stem Cells</td>
<td>Xiaoti Xu, MD 2nd Year Research Resident</td>
</tr>
<tr>
<td>1:20 PM</td>
<td>In Utero Depletion of Fetal Host Hematopoietic Stem Cells Improves Engraftment Following Neonatal Transplantation in Mice.</td>
<td>Chris Derderian, MD 2nd Year Research Resident</td>
</tr>
<tr>
<td>1:35 PM</td>
<td>Pancreatic Antigen Expression in Extrathymic Aire-Expressing Cells Prevents Autoimmune Diabetes.</td>
<td>James Gardner, MD, PhD PGY2, General Surgery Resident</td>
</tr>
<tr>
<td>1:50 PM</td>
<td>10-minute break</td>
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<tr>
<td>2:00 PM</td>
<td>Keynote Presentation: “Observations from the American College of Surgeons Quality Improvement Programs”</td>
<td>Clifford Ko, MD, MS, MSHS, FACS 2014 Dunphy Professor</td>
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<tr>
<td>3:00 PM</td>
<td>Closing Remarks &amp; Awards Presentation</td>
<td>Peter Stock, MD, PhD Research Committee Chair</td>
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<tr>
<td>4:00 PM</td>
<td>Post Symposium Reception for Residents, Faculty and Research Committee (4-7pm)</td>
<td>Social Kitchen &amp; Brewery 1326 9th Avenue (@ Irving), San Francisco</td>
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= Quick Shot presentations | = Standard presentations

A special thanks to all who participated in this year’s Resident Research Symposium and to the UCSF Department of Surgery Research Committee. This event is sponsored by an Educational Grant from the Howard C. Naffziger Surgical Fund.
Physiologic and Molecular Mechanisms of Arteriovenous Fistula Maturation
Joy P. Walker, Robert Toy, Hugh Alley, Michael S. Conte, Charles M. Eichler, Joseph H. Rapp, David Lovett, Warren J. Gasper, Christopher D. Owens

Introduction: Arteriovenous fistulas (AVF) are the most durable method of permanent hemodialysis access, yet only 50% mature sufficiently to initiate high quality hemodialysis. Beyond arterial or venous morphometrics, factors associated with maturation are poorly understood and characterized. We hypothesized that hemodynamic and physiological parameters of the blood vessels are critical to AVF maturation and are mediated via immune-dependent genetic pathways.

Methods: Prospective cohort study of patients undergoing upper extremity AVF. Preoperative measurements included patient characteristics and biomarkers of inflammation. Vascular wall characteristics included morphometry, structural (stiffness), and functional (endothelium-dependent flow mediated vasodilation, FMD) physiologic measurements. In a subset of patients hemodynamics were directly measured by computational fluid dynamics and differential gene expression was computed from temporally distinct, paired vein specimens from the same patient. Structural staining and confirmatory qPCR were performed in selected genes. The primary endpoint was AVF maturation.

Results: Twenty-nine patients were enrolled. Median follow-up was 87 days and 1 patient died prior to conclusion of follow-up. Median hsCRP was 8.0mg/L. Mean arterial stiffness (AIx) was 26.6% (healthy age-matched controls 20-22%). The mean FMD was 4.3% (healthy controls 10-12%). The overall maturation rate was 46.4%. Stiffer arteries (higher AIx) were significantly associated with a lower percent change in the venous diameter at three months (p=.02) as well as increased AVF failure at three months (p=.05). Hemodynamic measurement indicated that wall shear stress remained elevated at 3 months post AVF creation despite stabilization of vascular remodeling. Similarly gene ontogeny analysis revealed that inflammatory and fibrosis gene programs remained up regulated 3 months following AVF creation (p <<.05, see Figure). Structural stains confirmed an increase in lumen and total vessel area and deposition of elastin and collagen with significant intimal hyperplasia between baseline and 3 month samples (p<.05).

Conclusions: Renal failure is associated with increased brachial and central arterial stiffness and impaired endothelial function. Arterial and venous remodeling appears to be an endothelium-independent process. AVF maturation is associated with vascular compliance. Shear stress remains elevated and inflammatory and fibrosis gene pathways remain activated in mature fistulas despite stabilization of geometric remodeling.
INTRODUCTION: Mounting evidence highlighting the benefits of hemostatic resuscitation has led to a renewed interest in whole blood (WB) and reconstituted whole blood (RWB). However, no data exists to characterize the clotting profiles of these variants. This study characterizes banked WB variants and RWB in standard 1:1:1 and 2:1:1 transfusion ratios of packed red blood cells (RBC), fresh frozen plasma (FFP), and platelets (PLTS). We hypothesized that the global hemostatic profile of 1:1:1 RWB is superior to 2:1:1 RWB, and platelet-modified WB (MWB) is superior to 1:1:1 RWB.

METHODS: 23 units of RBC, FFP, and PLTS were obtained from the regional blood collection center and mixed to create 23 1:1:1 and 23 2:1:1 RWB units. Freshly donated WB units were obtained and used to create 11 of each: non-modified (NMWB) (room temperature and cooled) and platelet-modified (MWB) (room temperature and cooled) variants. MWB units were created by adding PLTS in a 6 WB:1 PLT ratio. INR/PTT, CBC, functional studies, and an extensive panel of pro- and anti-coagulant factor assays were performed on all products.

RESULTS: 1:1:1 RWB had significantly lower INR and PTT (1.31 vs. 1.55, \( p = 0.0029 \), Figure; 42s vs. 50s, \( p = 0.0008 \)) and higher activity of factors II, V, VII, VIII, IX, X, anti-thrombin III, protein C, and higher fibrinogen levels than 2:1:1 RWB (factor IX 86% vs. 70%, \( p = 0.0313 \); fibrinogen 242mg/dL vs. 202mg/dL, \( p = 0.0385 \)). There were no differences in INR/PTT or factor activity between MWB and NMWB. However, MWB had greater maximum clot firmness (MCF) by EXTEM than NMWB (MCF 61mm vs. 50mm, \( p = 0.0031 \)). MWB also had greater MCF by EXTEM than 1:1:1 RWB (MCF 61mm vs. 45mm, \( p = 0.0005 \)).

CONCLUSIONS: Although 1:1:1 RWB had a superior clotting profile relative to 2:1:1 RWB, MWB exhibited even better global hemostasis than 1:1:1 RWB. Characterization of factor-level and functional clotting differences between WB variants is imperative for understanding the clinical benefits of hemostatic resuscitation.
Characterization and Transplantation of Human Muscle Stem Cells
Xiaoti Xu MD, Karlijn Wilschut PhD, Gayle Kouklis BS, Catharine Garland MD, Jason H Pomerantz MD

INTRODUCTION:
Satellite cells are endogenous skeletal muscle stem cells that regenerate muscle, and are therefore appropriate targets to enhance muscle repair after injury or to be used as building blocks for muscle engineering. In mice, heterogeneous satellite cell populations have been well characterized to engraft and expand after transplantation. Although satellite cells have been identified in humans, their heterogeneity has not been determined nor have they been successfully transplanted. To date, attempts to transplant human muscle cells experimentally and in clinical trials have not used freshly isolated satellite cells and failed because of inefficient engraftment and poor survival. Therefore human muscle stem cells remain unavailable for clinical use. The purpose of this study is to develop approaches to characterize and transplant endogenous human satellite cells for regenerative applications.

METHODS:
Biopsies were taken from diverse human skeletal muscles during reconstructive procedures at UCSF. Individual samples were fixed or frozen for analysis of satellite cell content. Other samples were enzymatically digested for transplantation. Satellite cells were identified and counted on individual fixed fibers using antibodies against transcription factors and surface proteins in conjunction with anatomic localization beneath the basal lamina. Satellite cell content was determined by the following formula:

$$\text{Satellite cells per mm}^3 = \text{(average observed number of fibers in 1 mm}^2\text{) X (#satellite cells per mm of fiber)}$$

Samples for transplantation either underwent A) partial digestion with collagenase followed by preparation of live myofibers or B) digestion and staining for satellite cell surface markers followed by fluorescence activated cell sorting (FACS) purification of satellite cells. Whole fibers or purified satellite cells were transplanted into the tibialis anterior (TA) of NOD-SCID Gamma (NSG) mice after irradiation and injury with myotoxin to promote muscle regeneration. Mice were sacrificed at 5 weeks for histological analysis of engraftment or reinjured with myotoxin and sacrificed 5 weeks later. Muscle cryosections were analyzed for presence of human muscle cells. Human derived nuclei and fibers were visualized using hematoxylin and eosin and antibodies specific for human lamin a/c and spectrin. The number of human derived fibers was evaluated on cross sections of transplanted muscles.

RESULTS:
43 human muscle samples representing 7 muscle types, including rectus abdominis, sartorius, vastus lateralis, pectoralis major, latissimus dorsi, temporalis and gracilis, were collected and processed. The frequency of satellite cells per muscle ranged from 1 to 4 per mm of fiber and 500 to 1200 per mm$^3$ of muscle with frequency being similar in most muscles except for the temporalis, which had approximately two-fold greater frequency than the other muscles analyzed. After transplantation, both purified satellite cells and individual fibers consistently gave rise to human muscle fibers, whereas similar numbers of transplanted unsorted cells, or passaged myoblasts failed to engraft or yield human fibers. Transplantation of 5,000 isolated human satellite cells produced an average of 34.25 (range 11 to 56) human spectrin positive fibers after 5 weeks, each with human derived nuclei dispersed over 1-2cm lengths of the newly formed muscle fiber. Transplantation of 7 single human myofibers generated an average of 32 (range 5 to 53) human spectrin positive fibers at 5 weeks. Re-injury at 5 weeks resulted in generation of large clusters of human fibers, indicating proliferation and expansion of engrafted muscle cells. Engrafted human PAX-7 positive sublaminar cells identified on the periphery of human fibers indicated population of the satellite cell niche by transplanted cells.

CONCLUSION:
We have characterized muscle satellite cell frequency in diverse muscles and found it to be relatively constant with the exception of temporalis that had twice as many satellite cells compared to body and limb muscles. Our data demonstrate two successful approaches for muscle stem cell transplantation: isolation of fibers with niche-protected satellite cells and FACS purification of muscle stem cells. Using surface markers we enriched for muscle cells that demonstrated the capacity for engraftment into damaged muscle and formation of human-derived fibers. Progeny of transplanted human cells proliferated and participated in muscle repair after repeated injury, generating new muscle fibers of human origin and repopulating the satellite cell niche. Together our data identify a population of human muscle cells with properties of bona-fide stem cells and establish the feasibility of their transplantation, thus enabling development of future clinical applications.
Clinical Significance of Pneumomediastinum in Moderate to Severe Blunt Chest Trauma

Wayne S. Lee, MD; Vincent E. Chong, MD, MS; Gregory P. Victorino, MD
Department of Surgery, University of California, San Francisco – East Bay, Oakland, California, USA

Introduction: Because pneumomediastinum (PNM) is not commonly associated with severe injury, management for PNM can vary from observation, to panendoscopy, to operative intervention. It may be appropriate to observe mildly injured blunt trauma patients, but the clinical significance of PNM in more severely injured patients remains controversial. Our hypothesis was that there are identifiable risk factors associated with PNM in moderately to severely injured blunt trauma patients that portends an increase in mortality.

Methods: A retrospective review was performed on blunt trauma patients with injury severity score (ISS) ≥20 and abbreviated injury score (AIS) chest ≥3 who underwent initial chest CT scan admitted to our university based urban trauma center from 2002 to 2011. The presence and characteristics of PNM were analyzed.

Results: Of 441 patients, 35 patients (7.9%) had PNM on chest CT. Patients with PNM had higher ISS (p<0.05), and AIS chest (p<0.001). PNM was not associated with mortality, hospital stay, or ventilator days (p>0.1). PNM was associated with a longer intensive care unit (ICU) stay compared to no PNM (13.6 days vs. 8.9 days; p<0.05). PNM size was not associated with an increased in-hospital mortality (p=0.11); however posterior location and PNM in all mediastinal compartments were associated with increased mortality (p<0.05).

Conclusions: In moderate to severe blunt chest trauma patients, PNM is fairly uncommon. Patients with PNM were more severely injured and with longer ICU stay. Posterior PNM location and PNM in all mediastinal compartments were associated with increased mortality and should alert the trauma surgeon to a potentially lethal injury.
Inducing Acute Traumatic Coagulopathy *In Vitro*

BM Howard, LZ Kornblith, CK Cheung, RF Vilardi, B Miyazawa, MJ Cohen

**Introduction**
Nearly one third of critically injured patients present with acute traumatic coagulopathy independent of iatrogenic causes. We have previously reported that this coagulopathy is associated with shock and tissue injury, and is mediated via activation of the protein C pathway. Patients in ATC have hypocoagulable thromboelastometry (ROTEM) tracings as well as prolonged partial thromboplastin time (PTT) and decreased levels of factor V and VIII. To further test the etiology of this phenomenon, we hypothesized that such coagulopathy could be induced *in vitro* in healthy whole blood with the addition of activated protein C (APC).

**Methods**
Whole blood was collected from ten healthy subjects, and was “spiked” with increasing concentrations of APC (control, 75 ng/mL, 300 ng/mL, 2000 ng/mL). PT/PTT, factor activity assays, and ROTEM were performed on each sample. Analysis of variance testing was performed to assess differences between variables based on APC concentration, with Bonferroni correction used to assess significance in multiple comparisons. Linear regression was performed to assess the association of APC concentration with viscoelastic testing parameters, conventional coagulation tests, and specific factor activity levels.

**Progress/Results**
In all ten healthy subjects’ whole blood, increasing concentrations of APC produced findings consistent with traumatic coagulopathy. Analysis of variance testing showed that ROTEM parameters differed significantly by APC concentration, with stepwise prolongation of clotting time (CT) and clot formation time (CFT), decreased alpha angle (α), and reduced maximum clot firmness (MCF). Similar analysis revealed significant between-group differences in PTT and activity levels of Factor V and VIII. Linear regression demonstrated that for every 100 ng/mL increase in APC concentration, CT increased by __ seconds (CI __-, p= __), CFT increased by __ seconds (CI __-, p= __), α decreased by __ degrees (CI __-, p= __), and MCF decreased by __ mm (CI __-, p= __); PTT was prolonged by __ seconds (CI __-, p= __), factor V activity decreased by __ % (CI __-, p= __), and factor VIII activity decreased by __ % (CI __-, p= __).

**Conclusions**
In this study, we were able to reproduce a phenotype of acute traumatic coagulopathy in healthy whole blood by the addition of APC alone, as shown in both functional viscoelastic testing and in conventional and factor-based laboratory measures. This lends further mechanistic insight to the etiology of coagulation abnormalities in trauma, and provides a novel model for future research in the diagnosis and treatment of ATC.
Depression And Peripheral Arterial Disease: Is Physiology The Link?

Jessica N. Cohan, Emily Nosova, Karen Chong, Hugh Alley, Christopher Owens, and S. Marlene Grenon

Introduction: Depression is associated with the development of peripheral arterial disease (PAD). Among patients with PAD, depression is a risk factor for progression of disease and poor outcomes after surgical treatment. The reason for this association remains unclear. The goal of the present study was to assess the physiological differences between depressed and non-depressed patients in a cohort with PAD.

Methods: This study was a cross-sectional analysis using baseline data from the OMEGA-PAD Cohort, which enrolled consecutive patients presenting to the vascular surgery clinic at the San Francisco Veteran’s Affairs Medical Center to determine the impact of omega-3 fatty acids on PAD. Subjects underwent a history and physical exam, laboratory testing, and non-invasive studies including flow-mediated brachial artery vasodilation to evaluate endothelial function and applanation tonometry to assess arterial stiffness. Depression was defined by a Patient Health Questionnaire-8 (PHQ-8) score ≥10 and Post Traumatic Stress Disorder (PTSD) was defined as a PTSD Checklist score ≥50. Functional impairment was measured using the Walking Impairment Questionnaire (WIQ).

Progress: Eighty PAD patients with complete depression screens were included in this analysis. Sixty-seven (96%) were male and 17 (21%) met criteria for depression. Patients with depression were more likely to have co-morbid PTSD (41 vs 3%, p<.001) and previous vascular surgery (59 vs 32%, p=.04). There was also a trend towards increased tobacco use (41 vs 29%, p=.32), diabetes (59 vs 35%, p=.07), and coronary artery disease (53 vs 43%, p=.46) among depressed patients. When compared with their non-depressed counterparts, there was a trend towards increased inflammatory biomarkers and functional impairment in depressed patients. Non-invasive testing revealed greater endothelial dysfunction and arterial stiffness in patients with depression, although these differences did not reach statistical significance (Table 1).

Conclusions: This preliminary analysis shows that among PAD patients with depression, there is a trend towards greater comorbidity, inflammation, functional impairment, arterial stiffness, and endothelial dysfunction. We will continue to our goal enrollment of 200 patients, at which time we will be better powered to detect differences between these groups if they exist.

Table 1: Results of laboratory testing, WIQ, and arterial studies in PAD patients with and without depression. Mean and SD reported for parametric data; Median and IQR reported if non-parametric.

<table>
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<th>No Depression</th>
<th>Depression</th>
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<tr>
<td></td>
<td>N=63</td>
<td>N=17</td>
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<tr>
<td>Serum Inflammatory Markers</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>C-Reactive Protein</td>
<td>2.5 (3.4)</td>
<td>3.9 (5.8)</td>
<td>.32</td>
</tr>
<tr>
<td>Fibrinogen</td>
<td>395 (161)</td>
<td>417 (47)</td>
<td>.74</td>
</tr>
<tr>
<td>IL-6</td>
<td>1.1 (.72)</td>
<td>1.6 (.83)</td>
<td>.10</td>
</tr>
<tr>
<td>TNF-α</td>
<td>2.0 (.59)</td>
<td>2.5 (.79)</td>
<td>.09</td>
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<tr>
<td>Walking Impairment Questionnaire</td>
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<tr>
<td>Walking Distance Score</td>
<td>26 (46)</td>
<td>16 (28)</td>
<td>.72</td>
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<tr>
<td>Walking Speed Score</td>
<td>22 (43)</td>
<td>13 (24)</td>
<td>.18</td>
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<tr>
<td>Non-Invasive Arterial Testing</td>
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<tr>
<td>Flow Mediated Dilation %</td>
<td>7.4 (6.6)</td>
<td>4.2 (5.3)</td>
<td>.17</td>
</tr>
<tr>
<td>Pulse Wave Velocity (m/s)</td>
<td>10.8 (3.9)</td>
<td>12.9 (5.9)</td>
<td>.19</td>
</tr>
<tr>
<td>Aortic Pressure (mmHg)</td>
<td>129.0 (28.7)</td>
<td>137.3 (28.0)</td>
<td>.07</td>
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IL-6 Interleukin-6; TNF-α Tumor Necrosis Factor-α. Analyses performed using T-test or Wilcoxon Rank Sum.
Quality of Trauma Care for Undocumented Latino Immigrants

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Introduction: Undocumented immigrants rely on safety net hospitals and trauma centers for their emergency health care needs. However, little is known about the quality of care this population receives at urban trauma centers. There is also reason to believe that immigration status contributes to the health vulnerability of undocumented patients. As such, the purpose of this study was to measure quality of care at an urban hospital with a high volume of undocumented immigrant patients. We hypothesized that in-hospital mortality would be worse for undocumented Latino immigrants when compared to Latino patients with legal status.

Methods: The medical records for Latino trauma patients at our center between 2007 and 2012 were retrospectively reviewed. Undocumented status was defined using two inclusion criteria: 1) lack of social security number and 2) insurance status as either “self pay” or “county insurance” that undocumented immigrants are eligible for. Using multivariate logistic regression, patient demographics (age, gender, documentation status, mechanism of injury, and injury severity score) were analyzed in reference to their association with in-hospital mortality.

Results: During the study period, 2,441 Latino trauma patients were treated at our center, of which 465 (19%) were undocumented. Undocumented Latino immigrants made up 3.4% of the hospital’s trauma population. In contrast, this population is 8.4% of the total Alameda County population.

There was no difference between undocumented Latino immigrants and the control population in regards to in-hospital mortality (3.9% vs 3.4%; p = 0.61). Undocumented Latino immigrants were younger (mean 31 vs 33; p < 0.001) and presented with lower ISS (mean 7.5 vs 8.5; p < 0.001). Undocumented patients were also more likely to be uninsured (54% vs 29%; p < 0.001). The only predictors of in-hospital mortality by logistic regression were age (OR = 1.04; p = 0.002), ISS (OR = 1.24; p < 0.001), penetrating injury (OR = 5.57; p < 0.001), and lack of insurance (OR 5.37; p = 0.001).

Conclusions: The undocumented Latino immigrant population treated at our trauma center differed from the control population in regards to insurance status and age. Specifically, they were younger and more likely to be uninsured. This is consistent with data from previous research. Furthermore, in our county, undocumented Latino immigrants utilize trauma care less than what might be expected given their proportion in the population. Lastly, though being undocumented was not found to place Latino patients at increased risk of in-hospital mortality after trauma, having no insurance was a strong predictor. As healthcare and potential immigration reform change utilization patterns and federal reimbursement for urban trauma centers, it will be important to continue tracking outcomes for this potentially vulnerable patient population.
Choice of Injury Scoring System in a Resource-Poor Setting: Lessons from Mumbai.

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Introduction:
In India, 11% of deaths and 13% of disability-adjusted life years (DALYs) lost are due to injury. Accurate assessment of injury severity is crucial for clinical practice, research and quality improvement. Multiple injury severity scoring systems exist including anatomic scoring systems such as Injury Severity Score (ISS)—which is the current standard measure in Indian hospitals—and physiological scoring systems including Revised Trauma Score (RTS)—which is calculated based on Glasgow Coma Score (GCS), respiratory rate and systolic blood pressure—and MGAP score—a simple scoring system based on mechanism, GCS, age and systolic blood pressure but not respiratory rate (Sartorius 2010). We hypothesize that in a low resource setting in Mumbai, India ISS is less effective in assessing the severity of injuries than RTS or MGAP score.

Methods:
We performed a retrospective cohort study of patients with life- or limb-threatening injuries presenting to Lokmanya Tilak Municipal General Hospital in Mumbai, India between October 2010 and December 2011. Chi squared analysis was used to identify injury trends, and bivariate logistic regression models were used to correlate overall hospital mortality with ISS, RTS and MGAP, with receiver operating characteristic curves and Pearson chi squared analysis to assess goodness of fit.

Results:
Of the 1117 patients analyzed, 88% were male. The mean age was 31 years (SD 17). Road traffic injuries (32%) and falls (24%) were the most common causes of injuries. Overall in-hospital mortality rate was 32%, and 8% died within a day of presentation.

Missing data prevented calculation of ISS in 76% of patients and RTS in 65%, compared to only 12% for MGAP. In patients with sufficient data to calculate scores, RTS and MGAP scores were statistically significantly correlated with mortality with area under ROC curve 0.85 for each, while ISS was not with area under ROC curve 0.50. RTS demonstrated better goodness of fit than MGAP.

Conclusion:
The reality of clinical practice and research at trauma centers in low and middle-income countries (LMICs) makes accurate data collection challenging. In this cohort, RTS and MGAP correlated well with in-hospital mortality. While RTS was superior in predicting individual mortality, MGAP was a useful alternative that should be considered in LMICs when exhaustive data collection is not feasible.
Prognostic utility of lymph node size, number, clinical presentation and extranodal extension for papillary thyroid cancer nodal metastases following selective lymph node dissection.

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Introduction: A recent update of the American Thyroid Association (ATA) risk recurrence staging system recommended the inclusion of lymph node (1) size; (2) clinical presentation; (3) number; and (4) extranodal extension to risk stratify patients with papillary thyroid cancer (PTC) nodal metastases. The evidence for these recommendations is largely based on literature review, including many studies from institutions that perform routine prophylactic lymph node dissections. An assessment of these recommendations at institutions that do not perform prophylactic lymph node dissections is lacking.

Methods: We performed a cohort study of patients 18 years and older with PTC who underwent definitive surgical management and were found to have nodal metastases over a ten year period at a tertiary referral center that does not perform routine prophylactic lymph node dissections. Patient demographics, clinicopathologic data, and recurrence/survival outcomes were reviewed. The Kaplan-Meier method, Wilcoxon-Breslow tests, and Cox proportional hazards regression models were used to estimate survival functions and adjusted risk of recurrence/mortality.

Results: The median follow-up for all 228 patients was 9.1 years (interquartile range 7.4 to 12.3). Overall, 80 patients (35%) developed recurrences and 15 patients (7%) died from thyroid cancer during follow-up. On univariate analysis, patients with recurrences were older, more likely to be men, and to have a higher T stage, clinically positive and more involved overall and lateral lymph nodes, extranodal extension, and distant metastases. Of the four criteria recommended for the new ATA classification system, Wilcoxon-Breslow tests comparing survival functions found that extranodal extension and clinically positive lymph nodes were independent predictors of recurrence and decreased survival. Having greater than five positive lymph nodes and size of largest lymph node metastasis were predictors of recurrence only. A Cox proportional hazards regression model adjusting for age greater than 45, gender, pathologic stages and the above factors demonstrated that extranodal extension was the only of the four criteria that was predictive of recurrence.

Conclusion: Of the four criteria recommended for the new ATA risk recurrence staging system for PTC nodal metastases, extranodal extension is most predictive of recurrence and survival. Clinical presentation, number, and size of nodal metastases may not be as predictive of prognosis at institutions that do not perform routine prophylactic lymph node dissection.
From Novice to Master Surgeon: Improving Feedback with a Descriptive Approach to Intraoperative Assessment


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Background: Most tools for intraoperative assessment of surgical trainees are based on deconstruction of performance into isolated scales: technical skills, interpersonal ("nontechnical") skills, and conceptual knowledge. However, these skills interact in a complex and nonlinear way in the operating room, and our understanding of how surgeons learn to integrate them in practice is incomplete. This study explores a more comprehensive, developmental, and descriptive approach to assessing trainee intraoperative performance, which can improve communication and feedback.

Methods: We recorded semi-structured interviews with twenty surgeon educators and asked them to characterize how surgeons develop an integrated practice in the operating room. Interviews were transcribed, deidentified, and analyzed using a grounded theory approach to identify emergent themes. Two researchers achieved concurrence and then independently coded the transcripts. Emergent themes were also compared to existing theories of skill acquisition.

Results: Despite varied backgrounds and levels of expertise, all surgeon educators characterized intraoperative surgical performance as an integrated practice of multiple skill categories. These skills included anticipating, planning for contingencies, monitoring progress, self-efficacy, and "working knowledge" (applied understanding of conceptual knowledge, such as the ability to discern planes of dissection). Although specific developmental timing for these skills may depend on factors such as case complexity, surgeon educators described stages of development, broadly characterized as "technician," "anticipator," "dissector," "strategist," and "executive." Distilling these data, we constructed narrative, descriptive profiles of each stage of development.

Discussion: Viewing surgical performance as integrated practice rather than the conglomerate of isolated skills allows for more authentic assessment. The stage described in our qualitative data aligned well with existing models of skill acquisition, while providing a more integrated picture. The narrative, descriptive profiles of surgeons working towards mastery can help both educators and trainees better understand how to categorize, communicate, and correct deficiencies. They provide a standardized vocabulary for communicating feedback, while fostering reflection on trainee progress.
Establishing Prospective Trauma Metrics Evaluations for Pre-Hospital Trauma Care in Rwanda.

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Pre-hospital services in resource-limited setting lack guidelines about which indicators to measure. We hypothesize that simple metrics from routinely collected data can support pre-hospital quality improvement.

In November 2013, the pre-hospital emergency program in Rwanda, SAMU, established a prospective database, digitizing all paper-based run sheets. We analyzed metrics for injured patients to identify current performance and areas for improvement. Proportions between groups were compared using equality of proportion tests, using significance level or 0.05.

A total of 789 patients used SAMU services between November 2013 and January 2014. Injuries were the most common problem, affecting 447 (56.7%) patients, while medical and OB/Neonatology problems represented 232 (29.4%) and 109 (13.8%) patients, respectively. Road traffic crashes caused 70.3% of injuries. The extremities (54.7%) and head (40.8%) were the most commonly injured anatomic regions. Neurological problems (24.6%) and gastrointestinal problems (21.1%) were the most-common medical diagnoses.

Oxygen saturation was recorded in 99.8% of injured patients, including all 47 patients with thoracic injury. For medical patient oxygen saturation was recorded in 97.4% of patients. Injured patients were more likely to have saturations recorded (p = 0.004). While saturation was recorded, only 25.3% of injured patients with any indication for supplemental oxygen received it (21 of 83), whereas 47.6% (30 of 63) of patients with a primary medical diagnosis who required oxygen received it (p=0.005). Hemodynamic instability was noted in 28 injured patients, of whom 60.7% (17) received intra-venous (IV) fluids. Medical patients with hemodynamic instability were as likely to receive IV fluids as injured patients (p = 0.245). Of the 49 medical patients who were hemodynamically unstable, 23 (46.9%) received IV fluids. Hemorrhage was controlled with local pressure in 74% of bleeding patients (134 of 184). Glasgow Coma Scale (GCS) was documented for 96.6% of injured patients, and 97.2% with head injuries; 100% of patients with GCS ≤8 received oxygen during transport (n=9).

Preliminary evaluation of the SAMU system shows good performance, with some room for improvement, as well as differences in the response to injured and medical patients. In resource-limited settings, routinely collected pre-hospital data can become key performance metrics. Investment in accurate and reliable data collection systems is essential to improving care.
Precocious T Cell Maturation In Patients With Gastroschisis

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Introduction: The pathophysiology of intestinal damage in gastroschisis (GS) is poorly understood. We studied inflammatory biomarkers and T cell profiles in patients and mice with GS to understand the etiology of intestinal inflammation.

Methods: We prospectively enrolled patients with GS and healthy controls for analysis of maternal and cord blood cytokines and T cell profiles. Naïve, effector memory (EM), central memory (CM), and regulatory T cell subsets were examined using flow cytometry. Human and murine intestinal specimens were studied for T cell infiltration.

Results: Levels of eotaxin, IL-1Rα, IL-6, IL-8, and MCP-1 were higher in patients with GS compared to controls and increased with disease severity. Patients with GS also had higher percentages of EM CD4 and CD8 T cells, Th1 and Th17 cells, and CD8 cells producing IFN-γ, indicating T cell activation. Patient intestinal specimens demonstrated increased T cell and eosinophil infiltration into the intestine compared to controls. Eosinophils were also increased in mice with GS, which was more apparent in the herniated intestine and liver compared to the intraabdominal portion.

Conclusions: Fetuses with gastroschisis express high systemic levels of inflammatory biomarkers, polarization of T cells toward effector subsets, and infiltration of inflamed intestine with T cells and eosinophils, as seen in inflammatory bowel disease. These findings suggest that activated T cells may contribute to intestinal damage in GS and that anti-inflammatory agents may improve outcomes.
Total skin-sparing mastectomy and immediate tissue expander-implant reconstruction in the setting of radiation therapy.

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Introduction:
Radiation therapy is an increasingly common adjuvant therapy for treatment of locally advanced breast cancer and in patients with extensive nodal involvement. While autologous reconstruction is generally preferred in these situations, not all patients are candidates and some must undergo tissue expander-implant reconstruction. Few studies have investigated the impact of radiation therapy on reconstructive outcomes in the setting of mastectomy techniques that spare the nipple skin.

Methods:
We identified all patients who underwent total skin-sparing mastectomy (TSSM) with immediate tissue expander/implant based reconstruction from January 2006 to December 2012 from our breast reconstruction outcomes database. Patients were divided into 3 cohorts for analysis. The “No Radiation” cohort included all cases undergoing TSSM and reconstruction that did not receive radiation. The “Prior Radiation” cohort included cases that had a history of prior radiation therapy to the breast. The “Post-mastectomy Radiation” cohort included cases that received radiation therapy after TSSM and reconstruction. Because the use of acellular dermal matrix (ADM) has been associated with some postoperative complications, we also performed stratified analysis based on the use of acellular dermal matrix (ADM) at time of immediate reconstruction.

Results:
We identified 903 cases of TSSM and immediate tissue-expander placement in 581 patients. There were 727 cases in the “No Radiation” cohort, 63 cases in the “Prior Radiation” cohort, and 113 cases in the “Post-mastectomy Radiation” cohort. Patients in the “Prior Radiation” cohort had a higher risk of developing infections requiring PO (RR 2.06, p=0.04) or IV antibiotics (RR 2.83, p=0.001), infections requiring procedures for resolution (RR 2.88, p=0.026), wound breakdown (RR 3.32, p=0.012), and loss of prosthetic reconstruction (RR 4.05, p<0.005) when compared to patients in the “No Radiation” cohort. Patients in the “Post-mastectomy Radiation” cohort had a higher risk of developing infections requiring PO (RR 2.03, p<0.005) or IV antibiotics (RR 3.03, p<0.005), full-thickness skin necrosis requiring debridement (RR 3.10, p=0.001), and loss of prosthetic reconstruction (RR 3.48, p<0.005). The use of ADM was shown to modify the risk of postoperative complications. In the “Prior Radiation” cohort, there was a trend toward a decreased risk of infections requiring procedures for resolution with the use of ADM (p=0.143) and a trend toward an increased risk of skin and nipple necrosis complications with the use of ADM. In the “Post-mastectomy Radiation” cohort, there was a significant decreased risk of infections requiring procedures for resolution with ADM (p=0.041) and trends toward a decreased risk of infections requiring IV antibiotics (p=0.054), implant exposure (p=0.119), and implant loss (p=0.262).

Conclusions:
In patients undergoing TSSM and immediate tissue expander-implant based reconstruction, exposure to radiation therapy increases the risk of postoperative infections, necrotic complications, and implant loss. In patients who undergo post-mastectomy radiation, placement of ADM at the time of mastectomy may decrease the risk of postoperative infections, skin breakdown, and implant loss.
In Utero Depletion of Fetal Host Hematopoietic Stem Cells Improves Engraftment Following Neonatal Transplantation in Mice
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Introduction: In utero hematopoietic cell transplantation (IUHCTx) is a promising strategy to treat congenital disorders as the fetal host can potentially be tolerized to transplanted cells early in gestation. However, levels of engraftment have been low and fetal host conditioning strategies to increase space in hematopoietic niches have not been widely explored. We hypothesized that depletion of fetal host hematopoietic stem cells (HSC) using an antibody against the c-kit receptor (ACK2), a strategy which selectively depletes HSC by disrupting stem cell factor (SCF) signaling, would improve engraftment after HSC transplantation.

Methods: Fetal mice were injected with increasing doses of ACK2 (2.5-50 µg/fetus) or isotype control antibody on E14.5 and surviving pups were transplanted with congenic fetal liver mononuclear cells on day of life 1 (P1, 7 days after in utero injection). Host HSC depletion and residual serum ACK2 concentrations were examined on P1. Peripheral blood chimerism and the lineage distribution of chimeric cells were determined beginning 4 weeks after transplantation.

Results: Survival to birth among fetuses injected with 2.5, 5, or 10 µg of ACK2 was similar to controls (control: 74%; 2.5 µg: 80%; 5 µg: 71%; 10 µg: 60%, p=0.2 by chi-square test, n≥45/group) but was significantly lower at higher concentrations (20 µg: 37%; 50 µg: 31%, p<0.001 vs. control, n≥70/group). Transient anemia and leukopenia were observed on P1 with doses ≥ 5 µg which resolved by P7 (n=17). In utero ACK2 treatment resulted in a 10-fold depletion of host HSCs (defined as Lin-Sca-1+C-kit+, KLS) in the bone marrow of treated animals by P1 (5 µg: 2.45×10⁵ vs. control: 2.3×10⁶, p<0.0001, n≥17). Residual ACK2 antibody was undetectable in the serum by P1, validating our strategy of in utero depletion and neonatal transplantation. In animals receiving neonatal transplantation, ACK2 depletion resulted in a significant increase in levels of engraftment 4 weeks after transplantation compared to controls (control: 3.3±0.3%; 2.5 µg: 13±1.4%; 5 µg: 10±2.4%; 10 µg: 11±2.0%, p<0.05 for each dose vs control by ANOVA). Accordingly, we detected an increased number total bone marrow KLS cells 7 days after transplantation in ACK2 treated animals compared to controls (412±45.9 vs. 933±112 cells, p=0.01, n≥3/group). Moreover, levels of chimerism increased over time in treated animals (2.5 µg: 190%; 5 µg: 170%; 10 µg: 160%) while they remained unchanged in controls. Lineage analysis of peripheral blood for granulocytes, B cells, and T cells indicated an equal increase in all lineages, suggesting ACK2 depletes true HSCs and not committed progenitors. Interestingly, ACK2 depletion at doses 2.5-10 µg did not result in engraftment of allogeneic BALB/c cells (n=11), indicating that allogeneic neonatal transplantation, unlike in utero transplantation, is limited by a host immune response which is unaffected by ACK2.

Conclusion: We have demonstrated that fetal HSC depletion using ACK2 can lead to clinically relevant levels of donor cell engraftment with minimal toxicity. In previous studies with this antibody, host HSC depletion required either immunodeficient animals or concurrent irradiation, whereas we achieved depletion in wild-type fetal hosts, suggesting differences in fetal vs. adult HSC sensitivity to SCF signaling. Future studies should explore this strategy to improve engraftment in large animals models of IUHCTx.
Pancreatic Antigen Expression in Extrathymic Aire-Expressing Cells Prevents Autoimmune Diabetes  James M. Gardner, MD, PhD

Introduction:

The AutoImmune REgulator (Aire) gene is essential to immune tolerance. Aire is expressed in the thymus, where it allows specialized medullary thymic epithelial cells (mTECs) to express a diverse range of otherwise tissue-restricted self-antigens like insulin and thyroglobulin, thus exposing developing T cells to a more complete picture of immunologic self. Recently we described a novel population of extraThymic Aire-expressing Cells (eTACs) in the secondary lymphoid organs that also express a range of self-antigens, and which we speculate may play a complementary role in enforcing self-tolerance in the peripheral immune system. However no substantial evidence exists that eTACs are in fact tolerogenic. Here we investigate whether targeted expression of pancreatic self-antigens in eTACs is sufficient to induce immunologic tolerance and prevent autoimmune diabetes in two mouse models of disease.

Methods:

Two novel transgenic mouse strains were generated expressing the pancreatic antigens islet-specific glucose-6-phosphatase related protein (IGRP) and the Barbara Davis Center 2.5 antigen (BDC2.5) under control of the Aire promoter to study eTAC-mediated prevention of autoimmune diabetes by CD4+ and CD8+ T cells, respectively. Impact on autoimmune diabetes was studied using the nonobese diabetic (NOD) mouse in both intrinsic and adoptive transfer models of disease. The Aire-driven IGRP-GFP (Adig) mouse expresses a GFP-tagged copy of the islet antigen IGRP in eTACs and allows study of interaction between eTACs and the IGRP-specific CD8+ T-cell clone 8.3, and the Aire-driven BDC antigen (AdBDC) mouse permits study of eTAC interactions with islet-specific CD4+ T cell clone BDC2.5. Diabetes incidence was tracked in cohorts of 8-12 mice in two replicates with serial blood glucose measurement. Functional analysis was done with immunofluorescent and two-photon microscopy and flow cytometry, as well as in vitro cytokine release assays.

Results:

Targeted expression the pancreatic antigens IGRP or BDC peptide in eTACs entirely prevents T cell-mediated autoimmune diabetes in two distinct animal models of disease. Interaction between eTACs and CD4+ or CD8+ T cells causes deletion, functional inactivation, or conversion of those interacting T cells into a regulatory phenotype, but disease prevention does not require regulatory T cells. Further, unlike other putative tolerogenic peripheral APC populations, eTAC-mediated tolerance is uniquely stable and resistant to conversion from tolerance to activation in the presence of inflammatory stimuli.

Conclusions:

Together these results suggest that eTACs play an important role in maintaining immunologic self-tolerance, and suggest this population as an attractive therapeutic target for the prevention and treatment of autoimmune diabetes, and potentially for diverse applications in autoimmunity and transplantation.