Fluid Management in Septic Shock

Magdalena Boelsterl, M.D., M.P.H., Mark Siegel, M.D.

**Background:** Treating hypovolemia remains a central tenet of early management of severe sepsis. Goal directed fluid therapy has been shown to enhance survival when given within the first 6 hours after presentation. However, additional fluids in a resuscitated state can lead to respiratory failure, and it has been shown that a positive fluid balance in sepsis patients increases mortality and the risk for and duration of intubation. Precise end points for fluid resuscitation are yet to be defined, and there is recent evidence that aggressive fluid therapy in resuscitated sepsis after the time limit of 6 hours, can lead to a higher incidence of respiratory failure.

**Hypothesis:** Continued aggressive intravenous fluid resuscitation after the time threshold of 6 hours after presentation leads to increased incidence of intubation in patients with septic shock.

**Methods:** All patients admitted to the medical intensive care unit (MICU) between April 1st and September 30th 2010 were screened to meet the following inclusion criteria: Adult older than 18 years, admitted from the ED, 3 or more SIRS criteria and shock documented in the ED, as well as a code status that allowed for intubation within the first 24 hours. Patients who were retrospectively thought to not have been septic and those whose medical records were incomplete were excluded. Extensive data on demographics, severity of disease, and management of the patient both in the ED and in the MICU were obtained with a specific focus on hourly fluid rates and hourly urine output. Cumulative amounts of intravenous fluids, blood products and medications up until the time of intubation were documented. Totals for the group of patients intubated after 6 hours of early goal directed therapy (EGDT) were compared to the average cumulative amount of fluids not-intubated patients had received within the same timeframe. The differences were tested for significance with the Wilcoxon signed-rank test.

**Results:** Out of a total of 1152 MICU admissions over 6 consecutive months, 80 patients (=6%) met inclusion criteria; 13 of these patients were excluded. Of the remaining 67 patients with an average age of 65 years and a gender distribution of 34 women and 32 men, 24 (=36%) were intubated. The majority of intubations (n=15 or 62.5%) occurred before or during the patients’ ED stay. Patients intubated after EGDT (n=9) received an average of 233 ml/hr prior to intubation, whereas patients who were not intubated received on average 194 ml/hr between hours 6 to 24. 6 of the 9 intubated patients had received larger total amounts of intravenous volume than the average of patients who were not intubated. The difference between the groups did not reach statistical significance with a p-value of 0.2481.

**Conclusions:** So far, our data could not support the hypothesis that intubations in patients with septic shock after completion of EGDT are associated with an increased cumulative amount of intravenous volume preceding the intubation. Our study may not have been adequately powered, mostly due to the low number of intubations in our patient population and in part due to the small sample size. We will continue data collection to repeat the statistical analysis with greater power. Further analysis is ongoing and includes comparisons of net volume balances as opposed to net intravenous volume intake in the two groups and comparisons after adjustment for severity of disease. These results will be presented in the near future.
Background: Obstructive sleep apnea is a highly prevalent disorder characterized by recurrent upper airway collapse during sleep that manifests clinically through symptoms of excessive daytime sleepiness. Pathophysiologic sequelae of OSA, including intermittent hypoxia, sleep arousal, and daytime sleepiness can all be effectively treated using continuous positive airway pressure (CPAP). Recent analysis of observational studies has shown that CPAP use can reduce the cardiovascular morbidity and mortality associated with OSA. One common critique of these findings has been the potential for bias from the “healthy user effect,” which postulates that patients who are compliant with CPAP have a tendency to be healthier than others in observational cohorts in ways not accounted for by usual modeling. In particular, CPAP users may be more compliant with other medical therapies, including anti-lipid, anti-hypertensive, and anti-platelet agents that would predispose them to improved cardiovascular outcomes. To date, two studies have examined the association between CPAP compliance and medication adherence, but offer conflicting results. No long-term analysis of contemporaneous CPAP and medication usage has been undertaken to date.

Specific Aim: To determine whether compliance with Continuous Positive Airway Pressure (CPAP) therapy will be associated with adherence to cardiovascular medication regimens in a cohort of male veterans undergoing CPAP after stroke or transient ischemic attack.

Hypothesis: CPAP compliance will be associated with higher adherence to cardiac medications.

Methods: Subjects were taken from the ongoing VA GoToSLEEP study, which consists of veterans 1) with a history of stroke or TIA within the last 18 months who 2) carry a diagnosis of hypertension and 3) lack a previous diagnosis of sleep apnea. Patients diagnosed with sleep apnea and receiving CPAP during this trial were eligible for inclusion. All patients were assessed for CPAP compliance at 6 months and after study completion using “gold standard” directly downloadable data cards from their autotitrating CPAP devices. Acceptable CPAP compliance was based on an average usage of more than 4 hours per night, accompanied by device usage during >70% of all nights. Adherence to medications was assessed by tracking the log of medication refills in the VA electronic pharmacy. The proportion of days covered by each class of medication was calculated over the contemporaneous period of CPAP use at both 6 month and study completion time points. Subjects were classified as having adequate vs. low medication adherence for each regimen based on a threshold of 80% or more days covered during the exposure period.

Results: In the set of 46 patients meeting study criteria, 33% were adherent to CPAP at 6 months and 39% were adherent over the total time of device usage. The percentage of patients adherent to cardiovascular medications were 83%, 62% and 70% for anti-platelet, anti-lipid, and antihypertensive drug classes, respectively. At 6 months, there was a positive association between CPAP compliance and total cardiovascular medication adherence (p=0.017), with a 1.7-fold relative risk (95%CI [1.2,2.4]) for CPAP compliance given medication adherence, and a 6.8 fold relative risk for medication adherence given CPAP compliance [0.98,47]. For total study time, an association of similar strength was seen with near statistical significance (p=0.057). Analysis of individual drug classes revealed a significant association between adherence to lipid therapy and CPAP compliance at 6 months (p=0.02), with a similar trend observed over the total course of the study (p=0.18).

Conclusions: In this study on a set of VA patients with detailed long-term CPAP compliance data after stroke or TIA, adherence to cardiovascular medications is associated with CPAP compliance at 6 months, with a likely association at one year as well. The association is most significant for anti-lipid medication classes, likely because patients were least adherent to this medication regimen. A limitation of the study is a small sample size, which hampers our ability to estimate the strength of the association observed. The current study provides evidence of a significant healthy-user effect in patients compliant with CPAP, but further work is needed to examine temporal relationships between CPAP and medication compliance and adjust for additional confounders.
**Circadian Variation of Uncoupling Protein 3 Expression and its Influence on Oxidative Stress in Doxorubicin Induced Cardiomyopathy**

Kevin Dougherty, M.D., Raymond Russell, M.D., Ph.D.

**Background:** Cardiomyocytes contain set of proteins that are self-regulated and operate on a 24 hour cycle to help cardiomyocytes prepare for and react to expected daily stressors. It has previously been shown that *uncoupling protein 3* (UCP3), a mitochondrial membrane protein that helps to regulate the mitochondrial transmembrane potential is expressed in a circadian rhythm, likely through the role of PPAR. Furthermore, based on the function of UCP3, it is believed that perturbation of UCP can result in increased levels of reactive oxygen species (ROS) which are widely accepted as cytotoxic. Hemodynamic changes, particularly volume overload, has been shown to result in down-regulation of UCP3. Alternatively, ischemic preconditioning can increase UCP expression and there is a clearly established inverse correlation between UCP expression and both infarct size and reactive oxygen species (ROS) levels.

**Specific Aims:** (1) Investigate the correlation between the UCP3 gene expression and UCP3 protein levels. (2) Compare the cardioprotective effects of maximal versus minimal UCP3 circadian expression in response to doxorubicin.

**Hypothesis:** UCP3 protein levels will correspond to the mRNA levels previously described, however, there will be a time lag. Subsequently, elevated levels of UCP3 protein will provide a protective effect against oxidative stress as manifested by preserved cardiac function.

**Methods:** FVB mice were maintained on a 12-hour light:dark cycle and sacrificed in 3 hour increments over a 24-hour period (n=4/time point). Hearts were harvested and homogenized. UCP3 protein was isolated using gel electrophoresis and relative quantification was achieved using immunoblot technique with UCP3 and VDAC1/Portin antibodies. Total RNA was isolated using RNeasy. PCR reactions using 18S for normalization and gene-specific primers for UCP3 were performed according to Taqman protocols. A second cohort of animals (n=6/group), received doxorubicin (3mg/kg i.p., 3 times/week) either at 10 am (doxo10) or 7 pm (doxo7). Prior to and following doxorubicin administration, animals underwent echocardiography. Fractional shortening (FS) was calculated to assess cardiac function. Animals were then sacrificed and hearts were harvested for future assessment of apoptosis, oxidative stress and myocardial fibrosis.

**Results:** After normalization for total protein concentration, UCP3 protein expression was greatest at the 7 am and 10 am time points and lowest at 7 pm and 10 pm time points. Statistically significant differences between all subgroups were seen except 7:10, 13:16, 19:22 and 1:4 (p<0.05). No statistically significant different levels of UCP3 mRNA were detected between any time points. Doxorubicin administration resulted in 1.47±2.8gm and 1.93±2.4gm weight loss in the 10am and 7pm groups respectively. The control group (normal saline) gained 2.97±1.5gms. Doxo10 and doxo7 weights differed significantly from control, but not from each other (p<0.05). FS of the control, doxo10 and doxo7 groups were 42.9±4.6%, 39.5±5.4% and 30.7±5.1% respectively, with significant between both doxo10 and NS compared to doxo7 (p<0.05, p<0.01 respectively).

**Conclusions:** Circadian rhythms in UCP3 expression are associated with the susceptibility to doxorubicin cardiotoxicity, which has implications with respect to scheduling cycles of chemotherapy to minimize myocardial damage in patients receiving doxorubicin. Further studies will focus on histologic and biochemical analysis of tissue samples and determination of circadian changes in other antioxidant genes.
Information Needs and Sign-out Utilization Habits of Cross Covering Physicians

Robert L. Fogerty, MD, MPH. Leora I. Horwitz, MD, MHS

**Background:** Patients who are cared for by covering physicians have higher adverse event rates and increased delays to treatment. These adverse events can be the result of inaccurate or inadequate sign-out. However, the actual data needs of covering physicians, and where they obtain these data, are currently unknown.

**Specific Aim:** Describe the data set required by cross covering physicians and to describe how cross covering physicians use physician-physician sign-out.

**Methods:** During a four-week period from July 7 to August 3, 2010, interns on general medical services were asked to prospectively record data about their cross cover experience during a traditional 30 hour on-call shift or a night float shift. Each intern was provided with a pocket card and asked to record data directly on the card, which was collected the next day. Each time the intern was contacted regarding a patient received during sign-out (one call), the intern was asked to record who initiated the contact (ie, nursing, patient), what the situation was about (ie medications, test results), where the intern found the desired information (ie written sign-out, Electronic Medical Record[EMR]), whether all required data was located, whether the call was anticipated by the primary team, whether the call could have been anticipated by the primary team, and if the received sign-out was sufficient. Each intern was eligible to participate once during the study period.

**Results:** A total of 14/24 (58%) of eligible interns completed data collection, with 123 unique calls recorded. Interns were able to find all desired information for 91% of calls. Information was found most often in the written sign-out (44%), followed by the EMR (24%), and verbal sign-out (21%). Questions regarding orders (25%) were most common, followed by medications (20%), plan of care (17%), and test results (11%). Interns sought information in different places depending on the questions they were asked. Interns were more likely to use the written sign-out rather than the EMR when asked about medications or test results, and were equally likely to use the written sign-out or the EMR for questions regarding orders and plan of care (P=0.01). Nursing staff were responsible for 89% of calls. Interns judged 66% of all events as possible to anticipate, yet only 40% of all events were anticipated by the primary team. Calls regarding plan of care and medications were most likely to be anticipated, whereas events regarding test results were least likely to have been anticipated (p=0.009).

**Conclusion:** Even with widespread EMR usage, covering physicians remain most likely to reference information received during physician-to-physician sign-out. With 2/3 of cross cover calls being predictable, accurate sign-out remains vital to safe patient handoff and should include an emphasis on expected overnight events.

Sandi-Jo Galati, M.D., Karrie C. Hendrickson, Ph.D, RN, Kasia J. Lipska, M.D., Janis E. Bozzo, RN, MSN, Zhenqiu Lin, Ph.D, Silvio E. Inzucchi, M.D.

Background: Studies have shown an association between inpatient hypoglycemia and mortality. Several clinical factors predict inpatient hypoglycemia (illness severity, changes in nutrition, and type of anti-hyperglycemic therapy), but hospital blood glucose (BG) management, and specifically the value of point-of-care (POC) BG monitoring in the prediction of severe hypoglycemia, has not been investigated. Nonetheless, in their 2009 inpatient hyperglycemia consensus statement, the American Association of Clinical Endocrinologists and the American Diabetes Association (AACE/ADA) advised a reassessment of a patient’s anti-hyperglycemic regimen once the BG falls below 100 mg/dl.

Hypothesis: In diabetic inpatients with severe hypoglycemia (defined as a BG<50 mg/dl), there is a significant difference in the number of low-normal and mild-moderately hypoglycemic BG readings in the preceding 48 hours compared to a control population of diabetic inpatients without severe hypoglycemia.

Methods: We examined POC BG levels in all diabetic patients hospitalized on general wards at Yale-New Haven Hospital during one year. Of 480 patients with at least one BG <50 mg/dl (severe hypoglycemia), we identified 365 patients on subcutaneous insulin or a sulfonylurea drug whose event occurred ≥48 hrs from admission and who had ≥2 BG values recorded prior to the event. All BGs in the 48-hrs prior to the hypoglycemic event (mean 8.2), were categorized into five low-normal to mild-moderately low glycemic windows: <60, 60-69, 70-79, 80-89, 90-99 mg/dl. Similarly, a randomly selected 48-hr period of BGs (mean 6.5) from DM patients admitted during the same year without severe hypoglycemia (n=2387) was sorted into the same ranges. Chi-square tests were used to compare the %BGs catalogued and the proportion of patients with BG<100 between the 2 groups.

Results: The frequency of antecedent low-normal to mild-moderate hypoglycemia was nearly 2-fold higher in patients with severe hypoglycemia compared to randomly selected 48-hr periods in patients without severe hypoglycemia (21.7% vs. 11.3%; p<0.0001), becoming increasingly disparate with BG values <90 mg/dl (Table). In addition, a greater proportion of patients with severe hypoglycemia (61.1%) had BG <100 mg/dl in the 48-hrs preceding their hypoglycemic event compared to those without (37.6%; p<0.0001).

<table>
<thead>
<tr>
<th>BGs* over 48-h (mg/dl)</th>
<th>&gt;100</th>
<th>90-99</th>
<th>80-89</th>
<th>70-79</th>
<th>60-69</th>
<th>&lt;60</th>
</tr>
</thead>
<tbody>
<tr>
<td>Severe hypoglycemia</td>
<td>78.3%</td>
<td>5.6%</td>
<td>5.0%</td>
<td>4.7%</td>
<td>3.2%</td>
<td>3.3%</td>
</tr>
<tr>
<td>(2970 BGs)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No severe hypoglycemia</td>
<td>88.7%</td>
<td>5.7%</td>
<td>2.8%</td>
<td>1.5%</td>
<td>1.0%</td>
<td>0.3%</td>
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<tr>
<td>(15,576 BGs)</td>
<td></td>
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*antecedent in patients with severe hypoglycemia; random in patients with no severe hypoglycemia.

Conclusions: We conclude that diabetic inpatients with severe hypoglycemia frequently have low-normal and mild-moderately hypoglycemic BG readings in the 48 hours preceding the event. Accordingly, a BG value <100 mg/dl, and specifically <90 mg/dl, may serve as an important clue for providers to consider adjustment of anti-hyperglycemic therapy, supporting recent AACE/ADA recommendations.
Background: Anterior cruciate ligament (ACL) injuries are more common in female athletes. Previous studies have identified a higher risk of injury in the preovulatory phase and women taking oral contraceptive pills (OCP’s) show a higher risk at the start of the menstrual cycle. It is believed that progesterone may be protective for women with respect to ACL tears; however progesterone does not reach maximal levels in the serum until day 11. Previous research has demonstrated that KT measurements are correlated to ACL injury risk.

Specific Aim: To investigate the effect of OCP’s across the menstrual cycle on KT-2000 measures to determine if there is a difference in anterior translation between the initial and last week on OCPs.

Hypothesis: There will be increased anterior translation when comparing KT-2000 measures from week 1 to week 3.

Methods: Eighteen females (18-30 years of age), taking OCPs for at least 3 months were included in the study. KT-2000 measurements were tested 2 times weekly for 4 weeks after an initial familiarization visit. Entry point into the study was randomized with respect to the day of the menstrual cycle and the evaluator was blinded to the time of the menstrual cycle.

Results: The KT-2000 measurements were: Week 1 = 5.28mm, Week 2 = 4.72, Week 3 = 4.87, Week 4 = 4.64; Week 1 had significantly greater translation versus Week 3 (P<0.01).

Conclusions: The KT-2000 measurements vary across the menstrual cycle in women taking OCP’s for at least 3 months, with week 1 having the longest KT-2000 measurements. Week 1 is significantly different from the measurements on week 3 and 4. In this study, we documented that KT-2000 levels are higher at the start of the menstrual cycle. These findings may help explain previous research findings that women on OCP’s are more likely to tear their ACL at the start of the menstrual cycle.
Pilot Prospective Evaluation of the Oral Microbiome and Pneumonia Prevention Practices in Mechanically Ventilated Adults.

Sachin Jain MD MPH, Samit Joshi, DO, Sukanya Naramsimhan PhD, Erol Fikrig MD, Vincent Quagliarello, MD

**Background:** Ventilator-associated pneumonia (VAP) has a prevalence of 9-27% and is responsible for significant morbidity and mortality. Effective prevention methods include: oral Chlorhexidine rinses, elevation of the head of the bed to 30 degrees, and continuous post-glottic suctioning. Despite these measures, ventilator-associated pneumonia continues to occur in critically ill patients. National societies and the CDC have not yet instituted a formal “bundle package” of prevention measures. Our project attempts to take a novel upstream approach to understanding the pathogenesis of ventilator-associated pneumonia. Based on published data that critical illness alters innate and adaptive immunity, we are studying how the composition of the microbial community of the oral cavity changes during intubation, and proximate to the development of pneumonia.

**Specific Aims:** 1) Ascertain if and how the oral microbiome evolves over the course of intubation. 2) Measure the frequency of oral care, head of bed elevation, continuous post-glottic suctioning on intubated patients. 3) Measure the incidence of ventilator-associated pneumonia. **Hypothesis:** The oral microbiome changes during intubation and selects for the predominance of organisms commonly associated with ventilator-associated pneumonia.

**Methods:** Informed consent was obtained to recruit patients of both genders and all races of adult 18 years and older if they had been mechanically ventilated for 24 hours or less. Potential subjects were excluded if they: 1) had clinical or radiographic evidence for active pneumonia; 2) had a life expectancy <1 month; or 3) they or their designated proxy were unable or unwilling to give informed consent. In addition to a baseline sample collected within 24 hours of intubation, a sample was collected every 48 hours thereafter until 14 days after being intubated or until the patient was extubated or died, whichever occurred first. An oral swab was used to collect specimens from the gingivae, buccal mucosa, soft and hard palate. The specimens were placed in a sterile container containing buffer and labeled with a unique identifier. Samples were stored in a -80°C freezer. DNA extraction and PCR was performed on the samples. Other information was collected, including age, gender, race, reason for admission, reason for intubation, antibiotics administered during hospital course, temperature, white blood cell count, radiographic results, and microbiological data. DNA samples will be sent to an outside facility for bioinformatics analysis to determine the diversity of the oral microbiome. IRB approval was obtained by the Yale University HIC.

**Results:** Samples were collected from 9 intubated patients in the ICU that met inclusion criteria representing a range of demographics. Of these 9, none of them developed ventilator-associated pneumonia (VAP) by CDC/NHSN criteria. Samples were collected from as short as within 24 hours of intubation to as far out as 12 days post-intubation for any given patient. Anti-bacterial antibiotics were being given to 7/9 patients at the time of sampling. One patient was on an anti-viral agent, and one patient was on no antibiotics. Oral Chlorhexidine was prescribed for all (9/9) patients in the study during the course of their intubation. Head of the bed was elevated to 30 degrees in all patients.

**Conclusion:** There were no VAP episodes identified, and there was high adherence to evidence-based practices of the use of Chlorhexidine and head of bed elevation in intubated patients. Further correlations between the timing/type of antibiotics and changes of the oral microbiome are pending final DNA analysis at this time.
Changes of the Oral Microbiome in Aging Adults at Risk for Pneumonia

Joseph Y. Kim, MD, Sukanya Narasimhan, PhD, Can Bruce, PhD, Juliana Mastronunzio, PhD, Erol Fikrig, MD, Vincent Quagliarello, MD

BACKGROUND: The diverse populations of microbial organisms colonizing human hosts affect both health and disease. The colonizing microbes of the oral cavity (i.e. the oral microbiome) can plausibly affect the risk of respiratory tract disease, particularly in aging adults.

OBJECTIVE: This study examines changes in the human oral microbiome in the context of aging.

METHODS: Oral cavity samples were obtained from the gingival crevices, buccal-mucosa, tongue, and the soft and hard palate from 14 adults aged 27 to 97 years old. The bacterial composition of each of these samples was determined by amplifying and sequencing 16S rRNA gene segments and aligning them with 16S rRNA sequences stored in the Ribosomal Database Project. They were then hierarchically clustered into operational taxonomic units (OTU) at dissimilarity levels of 3%, 5%, and 20%, corresponding approximately to species, genus, and phylum ranks.

RESULTS: Among all 14 subjects, an average of 50 distinct OTUs corresponding to phyla and an average of 359 OTUs corresponding to genus were seen. Linear regression analysis of OTUs corresponding to species showed a significant decrease in the complexity and diversity of the oral microbiome with increasing age, as measured by Chao index, entropy, and evenness ($p$ values=0.016, 0.007, 0.021, respectively) among subjects aged 25-75. Further shifts in composition of the oral microbiome related to age were seen with principle component analysis of bacterial genera, which revealed a distinct clustering of the data points for subjects aged 25-75 and data points for subjects older than 75 scattered outside this group.

CONCLUSION: These observations suggest a pattern of shifting composition of the human oral microbiome in the context of aging. We hypothesize that these changes in the oral microbiome may be involved in the pathogenesis of respiratory tract infection in older adults. These results provide a framework for future studies in larger cohorts to examine the relationship between aging, the oral microbiome, and the development of pneumonia in older adults.
Transitional zone length is associated with non-obstructive dysphagia in patients with normal high resolution esophageal manometry

Shakthi Dharan Kumar MD, Xiangyu Cong, James Dzuira PhD, MPH, Anish Sheth MD

**Background:** In the past few years, high-resolution manometry (HRM) has become the principle tool to aid in the analysis of esophageal motility disorders. New parameters for classifying functional abnormalities can now be identified with this technology. The visual output in topographic format provides detailed information on the various anatomic segments of the esophagus. Recently there has been interest in the proximal esophageal transitional zone (TZ), which represents the junction between the skeletal and smooth muscle components of the esophageal body. Ghosh et al. demonstrated that TZ defects greater than 1 cm in duration and 2 cm in length were associated with unexplained dysphagia. Like the TZ, other parameters of esophageal manometry may be associated with symptoms of unexplained dysphagia.

**Specific Aim:** The purpose of this study is to identify novel manometric parameters in patients with dysphagia and conventionally normal HRM studies.

**Hypothesis:** Abnormalities in upper esophageal sphincter (UES) mean basal pressure, transitional zone parameters, and peristaltic wave velocity may be seen in patients with unexplained dysphagia.

**Methods:** A review of 300 consecutive patients who underwent HRM at our institution from 2008 to 2010 was performed to identify patients with dysphagia and normal HRM studies. All patients had normal endoscopic evaluations and underwent HRM with a 36-channel ManoView HRM (Sierra Scientific, Inc). Patients with symptoms of reflux without dysphagia and normal HRM studies were chosen to serve as 1:1 matched controls. UES pressure and peristaltic wave velocity were obtained from ManoView software analysis. Transitional zone segment was measured using the 30 mm Hg isobaric contour in a standardized method as outlined by Ghosh and colleagues in both the dysphagia and control groups.

**Results:** A total of 53 patients with unexplained dysphagia and normal HRM were identified, as well as 53 patients in the control group without symptoms of dysphagia. UES mean basal pressure and peristaltic wave velocity were not significantly different compared to the control group (p=0.055 and p=0.50 respectively). The TZ length was longer in the dysphagia group compared to controls (3.21 vs 2.35 cm, p=0.014) using the Wilcoxon rank sum test.

**Conclusions:** Our study indicates patients with dysphagia and normal HRM studies had a significant increase in the length of the proximal esophageal transitional zone compared to patients without dysphagia, even when adjusting for age, gender and height. These findings support the notion that defects in the TZ are associated with impaired bolus transit and symptoms of unexplained dysphagia.
Racial Disparity in EUS-FNA Referral Patterns of African American Patients with Solid Pancreatic Masses

Peter Le, M.D., Uzma Siddiqui, M.D.

**Background:** Pancreatic cancer is the fourth leading cause of cancer death overall. Compared to Caucasian patients, epidemiologic studies indicate that African Americans have disproportionately higher incidences (27-40%), rates of mortality (29-38%), more advanced disease, and lower rates of surgical resection of pancreatic carcinoma. Surgical resection remains the only potential cure for pancreatic cancer, but only 20% of tumors are resectable at the time of diagnosis. Endoscopic ultrasound (EUS) is a highly sensitive modality for visualizing pancreatic masses and provides real-time tissue sampling through fine-needle aspiration (FNA). Recent studies indicate that patients with pancreatic masses undergoing EUS have improved stage-appropriate management and improved survival. Currently, there is no clear data assessing if African Americans, a high-risk group, undergo EUS workup of pancreatic masses as frequently as other ethnicities.

**Specific Aim:** Investigate if a racial disparity exists for EUS-FNA in the pre-operative evaluation of patients with suspected or known pancreatic masses.

**Hypothesis:** African American patients undergo EUS-FNA less frequently than Caucasian patients for known pancreatic masses.

**Methods:** Patients were collected at two large tertiary referral centers (800-100 annual EUS procedures per center) between 2003 and 2008. Eligible patients had a pancreatic mass seen on imaging study (CT, MRI, US), were older than 18 years of age, and did not have a tissue diagnosis of the mass at the time of EUS referral. Exclusion criteria included age under 18 and known diagnosis of pancreatic cancer. The primary outcome was to identify the rate of African American patients who underwent EUS-FNA compared to other ethnicities. This was compared to the baseline rates of pancreatic cancer obtained from the Connecticut Surveillance Epidemiology and End Results (SEER) database over this time period. Statistical analysis to compare two groups (patients receiving EUS-FNA vs. patients in the SEER database) was performed using a z-test for two proportions. Conversion from the z-score to p-value was used to determine statistical significance.

**Results:** 205 patients were analyzed. By ethnicity, 89.7% were Caucasian, 3.9% African American, 3.4% Hispanic, and 2.9% did not report their ethnicity. The rate of pancreatic cancer recorded by the SEER database was 89% for Caucasians and 8.6% for African Americans. Z-test analysis of these two groups showed a p-value of 0.61 in the Caucasian group and 0.047 in the African American group. Diagnostic yield during EUS-FNA was 91.6%. All but 2 Caucasian patients had medical insurance at time of EUS-FNA.

**Conclusions:** Despite being at higher risk for pancreatic carcinoma, our data from two large EUS referral centers suggests that EUS is significantly underutilized in the African American population (p=0.047). There was no difference in the Caucasian group (p=0.61). The etiology for this disparity remains unclear and is likely multifactorial. Possible etiologies include referral bias at the time abnormal pancreatic imaging is obtained, advanced stage of disease at time of pancreas imaging prompting percutaneous biopsy of a metastatic lesion instead of EUS-FNA, or referral to interventional radiology biopsy of the pancreatic mass. Further studies are needed to identify and examine the limiting factors to EUS access in this population.
Improved detection of malignant biliary strictures with addition of K-ras gene mutation analysis to brushing cytology and biopsies.

Veena Nandigam MD, Priya Jamidar MD

**Background:** Accurate pre operative diagnosis of malignant biliary strictures is important for treatment and prognosis. Endoscopic retrograde cholangio pancreaticography (ERCP) in addition to evaluating the location and morphology of biliary strictures allows for tissue sampling. In this study the diagnostic yield of biliary brushing cytology and biopsies in combination with K-ras gene mutation was evaluated.

**Methods:** 231 consecutive specimens from 181 patients who underwent ERCP with biliary brushing cytology and biopsy from 2001 to 2009 at our institution were evaluated. 61 cases were excluded from final analysis because of inadequate follow up. 32 cases had the K-ras gene mutation analysis performed. Statistical analysis was performed using SPSS software.

**Results:** The mean age of patients in the study was 65 years, 53% were male and 73 out of 170 cases (43%) had malignant biliary strictures confirmed at subsequent surgery, core needle biopsy or endoscopic ultrasound guided fine needle aspiration cytology. The sensitivities for brush cytology and biopsy for detection of malignant strictures were 30% and 42%. When brush cytology and biopsies were combined, sensitivity slightly improved to 47%. Specificity was 100% for both modalities. Addition of K-ras gene mutation analysis improved the sensitivity of biliary brushings cytology to 72%.

**Conclusion:** Biliary brush cytology and biopsy have mediocre sensitivity in diagnosing malignant strictures and additional analysis of K-ras gene mutation markedly improved the diagnostic yield. Additional prospective studies are needed to determine if the K-ras gene mutation testing should be performed selectively or routinely in biliary strictures.

Table 1: Performance characteristics of cytology, biopsy and K-ras gene mutation in detecting malignant strictures.

<table>
<thead>
<tr>
<th>Diagnostic Modality</th>
<th>Sensitivity (95% CI)</th>
<th>Specificity (95% CI)</th>
<th>PPV (95% CI)</th>
<th>NPV (95% CI)</th>
<th>Accuracy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brushing Cytology (N=170)</td>
<td>30 (19 - 41%)</td>
<td>100 (96 - 100%)</td>
<td>100% (84 - 100%)</td>
<td>65% (57 - 73%)</td>
<td>70%</td>
</tr>
<tr>
<td>Biopsy (N=170)</td>
<td>42 (30 - 55%)</td>
<td>100 (96 - 100%)</td>
<td>100 (88 - 100%)</td>
<td>70 (62 -78%)</td>
<td>75%</td>
</tr>
<tr>
<td>Cytology+Biopsy (N=170)</td>
<td>47 (35 - 59%)</td>
<td>100 (96 - 100%)</td>
<td>100 (90 - 100%)</td>
<td>71 (63 – 79%)</td>
<td>77%</td>
</tr>
<tr>
<td>Cytology+K-ras gene mutation (N=32)</td>
<td>72 (47 - 90%)</td>
<td>93 (66 - 100%)</td>
<td>93 (66 - 100%)</td>
<td>72 (47 - 90%)</td>
<td>81%</td>
</tr>
</tbody>
</table>

*PPV- Positive predictive value, NPV- Negative predictive value, CI- Confidence Interval*
Adherence to the Surviving Sepsis Campaign’s delivery of antibiotics goals: Examining the management of severe sepsis/septic shock patients at YNHH
Olusegun Olusesi, MD., Mark Siegel, MD.

Background: The Surviving Sepsis Campaign guidelines have been promoted as a practice standard to improve outcomes for patients with severe sepsis/septic shock. Evidence shows that delays in the delivery of antibiotics increase the likelihood of death and complications. One of the aims of the campaign is to decrease the time to administration of antibiotics to within 1 hour for severe sepsis/septic shock.

Aim: To examine adherence to the goal of antibiotics delivery to severe sepsis/septic shock patients within 1 hour of presentation.

Methods: We examined the YNHH MICU admission registry from April 2010 until September 2010 for patients admitted directly from the YNHH ED who met criteria for severe sepsis and septic shock on presentation to triage. Inclusion criteria were admission to the YNHH MICU from the ED, age >18, and patients who met criteria for severe sepsis/septic shock with positive cultures on samples obtained within 24 hours of admission. Exclusion criteria were patients who were DNR and had decided not to receive vasopressors. Timing of antibiotics administration was confirmed using ED nursing medication administration documentation. Patients were classified into Group 1 (antibiotics <1 hour) and Group 2 (antibiotics >1 hour). The primary outcome was adherence to antibiotics delivery within 1 hour of presentation. Secondary outcomes compared and examined MICU and Hospital Length of stay between Group 1 and Group 2.

Results: 58 patients who had complete medical records were examined. 44 were identified as white, 11 as black and 2 as Hispanic. 21 were males and 27 were females. Median time for antibiotics delivery for all patients was 97 minutes (IQR 63-143). 14 patients (0.24), received antibiotics within 1 hour and 44 patients (0.76) patients received antibiotics >1 hour. Overall mortality rate was 0.2 (10/58). Mortality rate in Group 1 was 0.28 and in Group 2 was 0.14. 98% of the patients received antibiotics for which the infecting organism was sensitive to. For Group 1, median MICU length of stay was 2 days (IQR 1-3) vs. 3 days (IQR 2-5) in Group 2. Median hospital length of stay was 5 days (IQR 1-8) in Group 1 vs. 9 days (IQR 4-12) in Group 2.

Discussion: At YNHH, appropriate antibiotics are delivered to most patients presenting with severe sepsis/septic shock, but time to delivery rarely meets suggested goal of less than 1 hour from presentation. Early antibiotics administration may be associated with shorter length of stay in the hospital and MICU. Many factors could impact time to antibiotics administration, including ability to diagnosis severe sepsis/septic shock promptly, and time delays in ordering, receiving, and delivering antibiotics. Advantages associated with decreased length of stay could include fewer hospital associated complications and lower cost. Further studies will be required to look at the potential approaches to shortening time to antibiotics in this population.
Outcomes for Geriatric Patients Following a Rapid Response Team Call

Megan Panico MD, Grace Jenq MD, Shyoko Honiden MD, Katy Araujo MPH, Margaret Pisani MD, MPH

Background: There is conflicting data on the impact of Rapid Response Teams (RRT) on outcomes. There is no data on process of care after patients undergo an RRT. Specifically outcomes of importance to older patients have not been examined. Examining outcomes from RRT’s are crucial to optimizing valuable resources in the hospital. Understanding process of care and outcomes related to RRT’s will help physicians to better triage patients during an RRT as well as improve discussions with patients and families on code status and what it means to transfer to an ICU level of care.

Specific Aim: Describe the outcomes of the Rapid Response Team Calls at YNHH specifically looking at transfer to a higher level of care, change in code status, length of hospital stay, mortality and discharge location stratified by age.

Hypothesis: Increasing age will be associated with increased transfers to the ICU and increased adverse outcomes including mortality and the inability to return to previous level of function as measured by discharge location.

Methods: Retrospective descriptive study of 355 RRT calls over four and a half months in 2009. Reason for RRT call, clinical characteristics, and changes in code status were collected. We examined which patients were being transferred to the ICU based on age, acuity (APACHE II) and reason for RRT. We evaluated changes in code status, hospital mortality and discharge location.

Results: Of the 355 patient’s evaluated 178 remained on the floor, 24 were transferred to the step down unit and 153 were transferred to the ICU. Illness severity as measured by the APACHE II score was a significant predictor of transfer to the ICU (p<0.001). The average APACHE II score for patients who remained on the floor versus those transferred to the ICU was 14.6 versus 21.1. Patient’s with RRT’s called for hypoxia and “staff worried” were significantly more likely to be transferred to a higher level of care. Age was not found to be a predictor of transfer to the ICU or death. Patients with dementia were less likely to be transferred to the ICU (8% vs 21%), although this trend was not significantly significant (p = 0.09). Of patients admitted from home that were greater than 65, who were transferred to the ICU, only 23% were able to return home at discharge from the hospital compared to 38% of patients who remained on the floor. In patients less than sixty-five, 54% returned home after ICU transfer compared to 63% of patients who remained on the floor after an RRT.

Conclusion: RRT’s identify sicker patients who may benefit from ICU care. Age was not found to be a predictor of adverse outcomes including mortality in this patient population. Patients with dementia were less likely to be transferred to the ICU after an RRT. Despite age not being a predictor of adverse events in the hospital it clearly impacted the ability to return home following an ICU admission. Further research is needed regarding which patients receive benefit from ICU transfer after RRT’s and the impact of age on functional status after an ICU stay.
Effect of Estrogen Receptor-α Gene Mutations on Rapid and Genomic Responses to Estrogen

Amir H. Sadrzadeh Rafie, M.D., Jeffrey R. Bender, M.D.

Background: There are two known estrogen receptor (ER) genes encoding ERα and ERβ. Both ERα and ERβ isoforms are expressed in endothelial cells (ECs), mediating both genomic cardiovascular effects of estrogen and rapid membrane-initiated ER-dependent activation of endothelial nitric oxide synthase (eNOS), resulting in the elaboration of the athero-protective product nitric oxide (NO). Although the cardiovascular protective effect of estrogen has more recently come into question based on large scale randomized clinical trials, the potential favorable biological effects of estrogen, and of ER engagement, in the cardiovascular system has been recognized. The definition of an N-terminus truncated isoform of the ERα, ER46, in human EC, and its critical role in membrane-initiated rapid responses to 17β-estradiol (E2), opened a new chapter on estrogen effects in the vasculature as it produces a more favorable endothelial response to estrogen than full-length ERα(66). The molecular mechanism driving expression of this truncated receptor has not been elucidated.

Specific Aim: Generate ERα expression constructs with point mutations in a translation initiation (Kozak) sequence and neighboring splice acceptor site; and express these altered ERα’s in ER-negative COS-7 cells, evaluating whether these nucleotide changes result in loss of ER46 expression, and/or reduced rapid responses to E2.

Hypothesis: Polymorphisms or mutations within a Kozak sequence or a neighboring splice site that would normally direct ER46 expression reduce ER46 levels or function and confer an endothelial dysfunction state consistent with the development of vascular pathology.

Methods: Wild-type (WT) ERα and mutant constructs of ER-736 (A→T, ER Thr168Ser) and ER-754 (A→T, ER Met174Leu) were generated by mutagenesis PCR and verified by sequencing. COS-7 cells transfected with either WT or mutant ERαs and eNOS expression constructs were E2 stimulated for 15 minutes, following which eNOS and phospho(p)eNOS immunoblotting were performed. To assess nuclear (genomic) signaling driven by ligand-activated ERs, an estrogen response element (ERE)-luciferase construct was co-transfected with the constructs above and E2-stimulated induction was measured. Finally, surface E2 binding sites were assessed in transfectants using membrane impermeable E2-BSA-FITC (fluorescein isothiocyanate) and immunofluorescence microscopy.

Results: Results in COS-7 transfectants are displayed in the table below as E2-stimulated fold change relative to DMSO control responses (assigned value of 1) in the same transfectants* and as mean ± standard deviation for surface E2 fluorescent intensity in the transfectants¥.

<table>
<thead>
<tr>
<th>Transfectants</th>
<th>ER46</th>
<th>ERα</th>
<th>ER-736</th>
<th>ER-754</th>
</tr>
</thead>
<tbody>
<tr>
<td>peNOS*</td>
<td>1.55</td>
<td>0.94</td>
<td>0.87</td>
<td>0.95</td>
</tr>
<tr>
<td>Luciferase activity*</td>
<td>3.31</td>
<td>1.19</td>
<td>1.51</td>
<td>2.43</td>
</tr>
<tr>
<td>E2-BSA-FITC binding¥</td>
<td>Not done</td>
<td>4.18±0.52</td>
<td>5.04±0.78</td>
<td>15.09±0.98</td>
</tr>
</tbody>
</table>

Conclusions: ER46+ transfectants exhibit significantly greater receptor responses to estrogen than do ERα(66)+ transfectants. Alteration in potentially critical translation initiation and splice sites does not affect those differential ER responses.
Patient Decision Making in Total Knee Replacement
Lauren Wong, MD; Liana Fraenkel, MD, MPH

Background: Osteoarthritis is a common illness and the leading cause of lower extremity disability among older adults, resulting in extensive use of healthcare resources. As the population ages, the prevalence of OA is expected to increase. When knee osteoarthritis causes significant pain, has an adverse effect on a patient’s life, and has failed medical management, total knee replacement (TKR) is indicated as a treatment option. TKR has been shown to be a cost effective intervention that improves long-term pain, functioning, and quality of life. Despite this, many studies have found variability in the use of TKR based on sex, race and ethnicity, and geography, as well as unwillingness among patients with severe osteoarthritis to have TKR. The objective of this study is to improve our understanding of the decision making process underlying TKR in patients with severe knee osteoarthritis.

Specific Aims and Hypothesis: The goal of this study was to determine which priorities patients with severe knee osteoarthritis value most and how they make trade-offs when deciding whether or not to pursue TKR, and how these priorities correlate with different baseline characteristics.

Methods: HIC exemption was obtained. Inclusion criteria included age 60 years or older, ability to understand English, knee pain most days of the week for the past two months, and diagnosis of knee osteoarthritis. Exclusion criteria included diagnosis of inflammatory arthritis or prior TKR. Our goal was to recruit 100 subjects. Potential subjects would be identified via billing codes for knee osteoarthritis from the Yale Orthopedic Clinic, given that they see a high number of patients with severe osteoarthritis. Those meeting eligibility criteria would be asked to participate in the study, which involved answering a series of questionnaires. No sensitive data would be collected. The survey consisted of three parts. The first series of questions was designed to collect baseline characteristic data, such as severity of pain and its impact on function and quality of life, the subject’s feelings regarding knee replacement, and their baseline level of health. The second part consisted of a series of computer generated questions designed to assess how patients make trade-offs among different values related to TKR, such as expectation of surgical outcome, opinion of friends, and expected duration of rehabilitation. The last part collected basic demographic data. These data generate an overall rank-list of values, from most to least important, which would then be correlated with baseline characteristics.

Results: We were unable to successfully come up with a methodology to recruit subjects that was agreeable with the Orthopedics Department. Modifications of the subject recruitment process, including broadening to private orthopedic practices affiliated with Yale, were unsuccessful.

Conclusions: We successfully developed a series of questionnaires designed to better understand how patients with severe knee arthritis of different demographic groups with different views and emotions regarding knee arthritis and knee replacement surgery make trade-offs to arrive at a decision about whether or not to pursue TKR as a treatment option. However, we were unable to successfully recruit subjects to participate in the study via the Orthopedics Department. Alternative recruitment methods might include primary care physician referrals to the study, though this would likely require a longer length of time in order to recruit sufficient numbers.
Three Single Nucleotide Polymorphisms of Genes Involved in Innate Immunity are Associated With Early Viral Response in Caucasian Patients Treated for Hepatitis C

Xiao Zhao M.D., Wajahat Mehal, M.D. D.Phil.

Background: Chronic hepatitis C infection is the leading cause of cirrhosis in North America. The standard treatment for chronic infection includes a 48-week course of peginterferon-α combined with ribavirin, with a response rate of 50%. It is well known that early viral reduction predicts sustained viral response and African American patients have a lower response rate. Recently, a single nucleotide polymorphism (SNP) near the IL28B gene, has been shown in a genome wide association study to be associated with treatment response. This SNP, more prevalent within the caucasian population, demonstrates a mechanistic link between innate immunity and viral clearance.

Specific aim: Identify SNPs of genes within the innate immunity pathway that influence the efficacy of anti-viral immune response.

Hypothesis: Innate immunity plays an important role in clearing hepatitis C; therefore, genetic polymorphism of genes involved in this pathway is likely to affect treatment response.

Methods: We first identified 15 genes involved in innate immunity (Card8, Card16, Caspase1, Caspase12, CISH1, IL1B, IL28B, MyD88, Nalp3, NFKB1, Nod2, P2rx7, SLC2A9, Serpina9,TLR9) and identified SNPs within them using the 1000 genome database. We then narrow down the list based on the difference in racial frequency, literature search, linkage disequilibrium, and functional significance. We genotyped the selected SNPs using Sequenome on DNA samples obtained from the participants of the ViraHep-C study, which consisted of 188 Caucasians and 171 African Americans undergoing standard hepatitis C treatment. Chi-square test was used to calculate for association between each SNP and early viral response (EVR), defined as undetectable viral load at week 12 of treatment. All P values less than 0.05 by the two-tailed test were considered statistically significant.

Results: We have tested 34 SNPs of the above mentioned genes for association with EVR. In addition to IL28B, we identified three other SNPs that significantly impact treatment response at 12 weeks in caucasians. CARD8 SNP (rs2043211) was shown to be a positive predictor of response, whereas IL1 SNP (rs 1143634) and Nalp3 SNPs (rs6672995) were found to be negative predictors.

Conclusions: CARD8, Nalp3, IL1 and IL28 are all important members of the innate immunity pathway. Their involvement in hepatitis C treatment response further supports that innate immunity plays an important role in viral clearance, These findings can help us design appropriate genetic tests prior to initiation of treatment and can also potentially lead to future therapeutic targets.
Medication Errors And Misunderstandings On Hospital Discharge For Patients With Acute Coronary Syndrome, Heart Failure, or Pneumonia.

Authors: Boback Ziaeian, M.D.; Leora Horwitz, M.D.

Background: An appropriate medication regimen after hospital discharge is an essential component of effective care following hospitalization. Yet, adverse drug events post-discharge are exceedingly common. One study estimates that 12.5% of patients suffer an adverse drug event within 30 days of discharge, of which 62% are preventable or ameliorable. These preventable adverse events may be attributable to two factors: first, provider error, and second patient misunderstanding. Prescription errors and medication reconciliation have been studied during various transition points of hospitalization. On admission, medication reconciliation is prone to error with an estimate of 67% of medication histories being inaccurate and contributing to 27% of inpatient prescribing errors. A smaller number of studies that have identified medications errors at discharge. One study found 50% of discharge medication reconciliations are inaccurate or 16% of all medications prescribed. Equally important is patient understanding of the discharge medication regimen. Prior studies indicate that patients have a poor understanding of their medications lists upon discharge from the hospital.

Specific Aim: In this study, we analyze both provider errors in discharge medication regimens and patient understanding of discharge lists in order to understand their prevalence and distribution respectively.

Hypothesis: Provider errors may contribute to unintended modifications to medication regimens. After hospitalization, many patients do not understand modifications to their home medications.

Methods: We undertook a prospective, observational cohort study of patients older than 18 years, discharged from a medicine service at Yale New Haven Hospital with a diagnosis of acute coronary syndrome (ACS), heart failure (HF), and pneumonia. Patients underwent a structured, telephone interview within one week of discharge and asked about changes to their medications. Medications were classified as being relevant or non-relevant to their admission diagnosis. The primary outcome measure was the concordance between patient understanding and the hospital discharge medication list. Suspected provider errors were classified after physician chart review.

Results: A total of 377 patients were studied. 87.8% patients received at least one new prescription. The average number of medications continued per patient was 5.4 (SD 3.4) or 75.0%, re-dosed 0.7 (SD 0.9) or 13.3%, and 0.8 (SD 1.1) or 14.9% discontinued. Of all medication modifications, 10.3% were suspected provider errors. Errors were more common for non-relevant medications than relevant (6.6% vs. 19.5%). A higher proportion of suspected provider errors were for medications that were re-dosed or discontinued (21.8% and 26.3%). Within the discontinued and re-dosed category, the proportion of errors for non-relevant medications was significantly higher (38.3% and 41.0%) compared to the relevant medications (15.1% and 15.2%). Patients fully understood only 21.6% of the medication modifications across all medications. They fully understood 25.7% of the new prescription medications received, only 16.1% of the medications re-dosed, and 17.6% of the medications discontinued.

Conclusions: Numerous changes are made to discharge medication lists for patients upon discharge, many of them unintentional provider errors. Patients do not understand the vast majority of the intentional changes. Both these facts may place patients at increased risk of adverse medication events post-discharge. Working to establish systems that improve patient education and reduce provider errors upon discharge may improve patient outcomes.