

ASCLS Annual Meeting 2015: Official Abstracts of Submitted Papers, Case Studies and Posters

Atlanta, GA

The following abstracts have been accepted for presentation at the 2015 American Society for Clinical Laboratory Science (ASCLS) Annual Meeting and Clinical Laboratory Exposition to be held July 28 through August 1 in Atlanta, GA. Abstracts are reviewed by members of the ASCLS Abstract and Proposal Review Committee. They are the final authority in selecting or rejecting an abstract.

Papers, case studies and posters will be presented during the following times at the annual meeting.

POSTER PRESENTATIONS

Tuesday and Wednesday, July 28 and 29, 10:00am-4:30pm at the Georgia World Congress Center in the Exhibit Hall. *Authors will be present on Wednesday, July 29 from 10:30am to Noon to discuss their work and answer questions.*

ORAL RESEARCH PRESENTATIONS

Wednesday, July 29, 3:30-5:00pm at the Georgia World Congress Center.

Poster Presentation Abstracts

The Detection of HPV-Infected Cells Using Real-time PCR

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The family *Papillomaviridae* comprises a variety of non-enveloped circular double-stranded DNA viruses that have the ability to infect mammals and birds. Most human papillomavirus (HPV) infections are asymptomatic and go unrecognized in a clinical setting. Oncogenic or high-risk HPV types 16 and 18, however, are the causative viruses for at least 90% of cervical

cancer which is considered the most common neoplastic condition that affects women. Nononcogenic, or low risk, HPV types such as HPV types 6 and 11 are the cause of anogenital warts and other sexually transmitted infections. This indicates the importance of detecting the virus for the purpose of monitoring viral load and treating infection. The HeLa cell line is a model system for positive detection and examination of HPV viral DNA infection because it has been transformed by HPV-18 and consequently carries portions of the viral genome. HEK-293 cells are derived from human embryonic kidney cells and do not have HPV DNA present in their genome. There are currently four FDA approved tests used for detecting HPV including probe assays and PCR amplification assays. Real-time PCR is considered a sensitive method for the detection and quantification of HPV DNA. The objective of this research project was to detect HPV using real-time PCR and determine a cutoff value to establish clinical sensitivity and specificity. It was demonstrated that HPV DNA was detected in the HeLa cell specimens at a concentration of 0.156 nanograms of DNA per reaction with a mean C_t cutoff value of 33. Analytical specificity and sensitivity of this assay were validated through statistical analysis using an ROC curve. These findings indicate that real-time PCR is a usable assay for the detection of HPV DNA.

Interprofessional Opportunities for Medical Laboratory Science, Nursing, and Physical Therapy Students in a Diabetes Self-Management Education Program

Karen Golemboski, PhD, MLS(ASCP)^{CM}, Bellarmine University, Louisville, KY

The Institute of Medicine has established that interprofessional practice is an essential competency for all healthcare professionals; however, healthcare education still emphasizes individual preparation. Including interprofessional experiences during

education will help to prepare graduates for integrated, cooperative practice opportunities. Through a university-based community outreach program, undergraduate and post-baccalaureate Medical Laboratory Science (MLS) students participated in a diabetes self-management education program (*Active Steps for Diabetes*) for mobility-impaired adults with Type 2 diabetes, along with students in a Doctor of Physical Therapy program and with both graduate and undergraduate Nursing students. Over a semester, MLS students met with the other healthcare students, as well as with patients, to discuss pertinent laboratory tests. At the conclusion of the program, students and faculty from all 3 programs participated in Grand Rounds presentations to review patient management and outcomes. After conclusion of the interprofessional program, a survey indicated that 100% of the MLS students agreed (strongly or somewhat) that meeting with other healthcare students helped them to recognize different aspects of MLS practice; 80% agreed that meeting with patients did the same. Ninety percent agreed that communicating about laboratory testing with students in other programs helped them to learn more about the laboratory's role in healthcare; the response was even higher (100% agreement) that communicating with patients helped them to learn more about the laboratory's role. Additionally, 90% indicated they would like to participate in additional interprofessional activities.

Nonthrombocytopenic Purpura and Arthralgia in a Child: A Case Report

Jessica Ming, Kristin Landis-Piowar, PhD MLS (ASCP)^{CM}, Oakland University, Rochester, MI

We describe a case of male child who presented with palpable purpura on the legs, and arthralgic and arthritic knees. Clinical history revealed an ear infection four days prior. Chemistry, hematology, and hemostasis laboratory evaluation was performed and all parameters were found to fall within the normal reference interval. A diagnosis of Immunoglobulin A vasculitis (IgA-V), formerly known as Henoch-Schönlein purpura (HSP), was given for the patient. IgA-V is the most common small-vessel vasculitis seen in children and is frequently reported following respiratory tract infections both viral and bacterial. It is a systemic condition characterized by the tetrad of nonthrombocytopenic palpable purpura,

arthritis or arthralgias, gastrointestinal involvement and nephritis. IgA-V is mediated by antigen-IgA immune complexes that deposit in small vessel walls and that leads to activation of the alternate complement pathway. Identification and diagnosis of IgA-V are of particular importance since the purpuric lesions are easily confused with the hemorrhagic rash seen in idiopathic thrombocytopenic purpura (ITP). IgA-V is considered to be self-limiting with mild cases resolving spontaneously. Treatment is generally supportive, although corticosteroids have been indicated for relief from severe joint pain. The prognosis is good, except in children with renal failure. The symptoms experienced by the patient in this case resolved after two weeks.

Effect of Leptin on Human Mononuclear Cell Production of IL-8, IL-12p70, and MIP-1 α

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Leptin is an adipokine produced primarily in white adipose tissue. Its blood concentration is directly proportional to body fat percentage and is therefore higher in obese individuals. Leptin is known to stimulate circulating monocytes to secrete certain pro-inflammatory cytokines such as TNF α . Leptin's role in inflammation is still not completely understood. We hypothesized that leptin would be a non-selective stimulus of the cytokines IL-8, IL-12p70, and MIP-1 α . Blood samples were drawn from 34 healthy subjects (body fat range: 11-43%) for serum analysis and cell culture. Peripheral blood mononuclear cells were isolated by density gradient centrifugation and cultured using leptin concentrations of 0, 50, and 100 ng/dL. Cytokine concentrations were measured by immunoassay. Serum MIP-1 α was positively correlated with percent body fat ($p = 0.01$) and serum leptin ($p = 0.01$). In vitro, leptin induced MIP-1 α in a dose-related manner ($p = 0.0003$), but had no significant effect on IL-8 or IL-12 secretion. The magnitude of the leptin-induced response was positively related to the subjects' age ($p = 0.01$) and serum leptin concentration ($p = 0.004$). Therefore, we conclude that leptin selectively induces some pro-inflammatory cytokines, such as MIP-1 α , but not others. MIP-1 α may contribute to the chronic inflammation associated with obesity. This work was supported by NIH grant HL 093663.

Stable Dabigatran Levels in Anticoagulation Clinic Patients: An Unexpected Finding!

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The dabigatran effect is rapid. Its half-life is 12–17 hours. Experts believe missed doses may lead to thrombosis. We evaluated plasma dabigatran levels on anticoagulation clinic patients monthly for 6 months. Subjects took 150 mg BID beginning one month prior. All subjects were >18 years old and had creatinine clearances >30 mL/min. No subjects were excluded for parallel medications or pre-existing conditions. We enrolled 64 males, mean age 77, and 38 females, mean age 76. Mean CHAD₂DS₂-VASc score was 3.2; mean BMI 29.8. Specimens from 72 subjects who completed the six-month cycle and 30 healthy controls taking no medication were analyzed. A 3.2% sodium citrate specimen from each subject on a monthly date within \pm 5 days of the start date was collected. There was no monitoring the time subjects took medication. We prepared platelet poor plasma by immediate centrifugation and stored at -70°C . We assayed specimens using the APTT (APTT Automated, Stago); Ecarin Chromogenic Assay (ECA-T, Stago); Hemoclot Thrombin Inhibitor (HTI, Aniaira); and Prothrombinase-induced clotting time (PiCT, Enzyme Research Laboratory). Testing was performed on a STAR-Evolution coagulometer. Reference intervals were computed from controls for the APTT (mean 30.7 s; \pm 2SD: 26.1–35.3 s); PiCT (mean 40.1 s; \pm 2SD: 29.4–50.7 s). Dabigatran subjects' APTT range was 26.2–>300 s, mean 53.7 s (>300 was assigned 301 s); PiCT range was 60.8–>301 s, mean 186.7 s; ECA-T range: 10–950 ng/mL, mean 177.6 ng/mL; HTI range 0.0–770 ng/mL, mean 185.3. A two-factor ANOVA (dabigatran, time) with repeated measures on all tests revealed no significant difference between monthly results within subjects ($p=0.234$) for any assay. Our data imply reproducibility in the absence of strict dosage time versus specimen collection time. This unexpected consequence implies a need for studies that determine dabigatran anticoagulant effect versus time subsequent to dosage.

Measurement of Blood Lead Levels in the Workplace

Deborah Sammons, BS, Christine Toennis, BS, Elena Page, MD, Barbara MacKenzie, BS, Jerome Smith, PhD, John Snawder, PhD, Marissa Alexander-Scott, DVM, Shirley Robertson, Centers for Disease Control and Prevention, National Institute for Occupational Safety and Health, Cincinnati, OH

Acute and chronic lead exposure is still a health risk for firing range, construction and foundry workers, workers recycling electronics and batteries, and many others involved in manipulating lead. Routine monitoring of blood lead levels (BLLs) in occupational settings is used to indicate recent exposure and often involves invasive venous blood draws and time consuming, laboratory based analysis. Point of care instruments, like the LeadCare® II, use a small quantity of blood and provide results within minutes, making biomonitoring and evaluation of work controls and practices quicker and easier. Traditionally, finger-sticks are not recommended due to external contamination. We investigated the feasibility of measuring BLLs, using the LeadCare® II, from three sample types, all types collected from 22 workers in an electronic scrap recycling facility: 1) blood from finger-sticks after cleansing with lead removal wipes, Hygenall® Field Wipes, 2) blood from finger-sticks after cleansing with Castile soap towelettes and 3) venous blood from the arm. Each participant's LeadCare® II results were compared to a venous blood sample analyzed by graphite furnace atomic absorption spectrometry (GFAAS). BLLs obtained after cleaning fingers with Hygenall® Field Wipes or Castile towelettes were correlated ($r=0.95$, $P<0.01$); however, the former were higher (mean difference = 1.0 micrograms per deciliter [$\mu\text{g}/\text{dL}$], $P<0.01$). Both finger-stick BLL samples correlated with the venous samples as measured by GFAAS ($r=0.96$ and 0.97 respectively, $P<0.01$), but were higher (mean difference= 3.2 $\mu\text{g}/\text{dL}$ and 2.2 $\mu\text{g}/\text{dL}$, respectively, $P<0.01$). Venous BLLs analyzed by LeadCare® II correlated with GFAAS ($r=0.98$, $P<0.01$) and have the potential to be useful in monitoring lead exposure as a screening tool, but were higher (mean difference = 1.7 $\mu\text{g}/\text{dL}$, $P<0.01$). Additional work needs to be done to examine the effectiveness of using blood from finger-sticks for monitoring BLLs in workers.

The findings and conclusions in this abstract have not been formally disseminated by the National Institute for

Occupational Safety and Health (NIOSH) and should not be construed to represent any agency determination or policy. Mention of company names and/or products does not constitute endorsement by NIOSH.

Expression of RAGE on Mononuclear Cells

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Receptor for Advanced Glycation End (RAGE) products is a pattern recognition receptor that binds advanced glycation end (AGE) products and high mobility box group-1 (HMGB-1, a protein released by damaged cells). Diabetes is associated with chronic inflammation: increased expression of RAGE receptors may be one mechanism by which inflammatory cells such as peripheral blood mononuclear cells (PBMCs) become activated, since type II diabetics have increased expression of RAGE on their PMBCs. Atherosclerosis is often associated with diabetes, developing as a result of vascular injury, inflammation and vessel remodeling. Factors affecting the risk for diabetes include weight, sex, and blood concentrations of cholesterol, glucose, and insulin. Our study's objective was to determine what risk factors for type II diabetes are associated with increased RAGE expression on PBMCs. We hypothesized that these risk factors would be directly to RAGE expression on PBMCs. To test our hypothesis, blood was collected from healthy individuals, PBMCs were isolated by density gradient centrifugation and RAGE expression (as well as serum HMGB-1) was analyzed by western blot. Risk factors were measured using an automated clinical analyzer, and interleukin-6 (IL-6) secretion was measured in cell culture supernatants as a maker of activation. RAGE expression correlated positively with serum triglyceride ($R = 0.43$, $P = 0.01$), and the strength of this correlation was associated with the number of CD14-dim, CD16+ cells ($P = 0.01$). These CD16+ cells reportedly congregate in atherosclerotic plaques. Furthermore, IL-6 secretion positively correlated with HMGB-1 ($P = 0.006$) and was twice as high for cells with above-median RAGE expression, compared to those below the median ($P = 0.04$). These results are consistent with the concept that serum triglycerides increase RAGE expression, which augments HMGB-1-induced IL-6 and promotes inflammation. This work was supported by NIH grant

HL 093663.

Refining Evaluation Measures of a Continuing Education Activity for Laboratory Practice Recommendations

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Providing effective training on good laboratory practices is a continued initiative of the Centers for Disease Control and Prevention (CDC) to improve the quality of clinical and public health laboratories. In April 2012, CDC published the "Good Laboratory Practices for Biochemical Genetic Testing and Newborn Screening for Inherited Metabolic Disorders" document (<http://www.cdc.gov/mmwr/pdf/rr/rr6102.pdf>) with a companion online continuing education (CE) activity. The CE activity included a post-test with most questions containing 'choose all that apply' answer choices from April 2012-May 2014 (Phase I). In June 2014, the post-test questions were revised into a 'single-best answer' format to better assess the participants' learning of the recommended practices (Phase II). The 319 participants who completed Phase I and the 69 participants who completed Phase II as of December 2014 presented similar professional activities and work settings, with nurses and laboratory professionals accounting for the top two participant categories. Phase II participants experienced more than a 60% increase in passing the post-test on the first attempt (28.8% and 46.4%, Phases I and II, respectively). In addition, Phase II participants reported a greater degree of satisfaction with the CE activity, including a higher percentage agreeing that the difficulty level was appropriate and that the stated learning objectives were met (89.1% vs. 94.4% and 89.7% vs. 94.2% respectively, Phase I vs. Phase II). However, it is difficult to attribute the improved responses to the revised post-test because the participants' baseline knowledge and reasons for seeking CE credits are not currently assessed. A follow-up evaluation will be conducted to determine factors associated with the improved participant feedback and address the additional educational needs identified in this CE activity. These additional evaluation measures will complement the current CE evaluation, and be used to improve the delivery of this and future CE trainings on quality laboratory practices.

Potential Financial Impact of Implementing Screening Tests for the Diagnosis of Cold Agglutinin Disease

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As healthcare continues to evolve, there are financial pressures to contain rising costs while providing quality patient care. An IRB approved retrospective audit was conducted on the appropriateness of laboratory testing for the diagnosis of cold agglutinin disease (CAD). Provider ordering practices were evaluated prior to and since the implementation of a cold agglutinin screen (CAS) in April 2012 which includes a direct antiglobulin test (DAT-IgG and C3d) and saline antibody screens with auto controls at RT and 30°C along with cold agglutinin titers (CAT). Moreover, testing results were compared to the ultimate diagnosis rendered. A retrospective chart review from January 1, 2010 through October 31, 2013 was performed identifying all CAT and all CAS since the inception of the test in April 2012. The electronic medical record of each patient was reviewed for diagnosis and preliminary testing. A direct cost analysis including reagents, consumables and labor was performed for CAT and CAS. Cold agglutinin titers of >1:64 were considered clinically significant in the diagnosis of CAD. There were 80 cold agglutinin titers (CAT) ordered prior to inception of CAS, of which, 5 (6.3%) were clinically significant with titers of >1:64. Since the inception of the cold agglutinin screen (CAS) in April 2012 there were 17 CAS ordered. Ultimately, three (17.6%) were diagnosed with CAD. Since April 2012, 38 CAT were ordered independent of the CAS, of which, 3 (7.8%) were true positive with titers >1:64 and all previously diagnosed with CAD. Our study demonstrated the pretest probability went from 6.3% using the CAT to 17.6% using the CAS. The 35 (92.1%) independently ordered CAT revealed no new diagnosis of CAD at a total cost of \$562.10.

Should Babesiosis Testing be Considered in Blood Donor Screens?

Anietie Uko, MLS (ASCP)^{CM}, Tufts Medical Center, Boston, MA

The purpose of this study is to determine if the Food

and Drug Administration and the American Association of Blood Banking should incorporate testing for the apicomplexan piroplasm parasite *Babesia microti* with correlation to recent studies done by the American Red Cross. In early August a 40 year old patient was seen in a Level One trauma facility in Massachusetts due to a motorcycle accident. The patient was quickly taken into surgery for a splenectomy and a nephrectomy. During his surgery he received units of Red Blood Cells, Fresh Frozen Plasma, and Platelets. In the beginning of September he was re-admitted due to a fever and abdominal pain. The patient was then diagnosed with Transfusion Transmitted Babesiosis. During his stay the patient was transfused with eight units of Red cells and administered Atovaquone and Azithromycin. The patient endured a transfusion reaction a day after his second emergency room visit and was also diagnosed with T.A.C.O. Due to the transfusion related Babesiosis, Blood donor suppliers were contacted and performed a follow-up from the 6 different patient samples they issued. One donor reported she tested positive for the Babesia parasite in a Massachusetts Hospital after she sought treatment for a fever and chills. Currently donors are not tested for the parasite, but are asked if they have ever been diagnosed with Babesiosis in their pre-donor screen.

Implementing Patient Safety into Medical Laboratory Science Curriculum

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The Institute of Medicine has identified quality aims and patient safety competencies that are needed to improve U.S. healthcare. Most healthcare professions have introduced patient safety initiatives and educational curricula into their practices and educational programs to respond to these reports, but the medical laboratory profession has been slow to adopt these concepts into our curriculum. Many medical laboratory professionals are not familiar with patient safety vocabulary and concepts, nor are most able to recognize the laboratory's role in ensuring patient safety. The purpose of our study is to

demonstrate the effectiveness of a patient safety curriculum in three different medical laboratory science programs in the United States. A patient safety module was added to one course in each program during the first and last semesters of each program. Analysis of student learning was assessed after each educational intervention. During the first semester of the two year study, a 10 question quiz was administered to medical laboratory science students one week prior to the presentation, along with assigned reading on patient safety, and again, 1-3 weeks after the presentation. The number of correct answers on the pre-quiz was compared to the number correct on the post-quiz. Data for 32 students at three MLS programs indicate a statistically significant (p value 0.000) improvement of 1.5 points on the post-quiz as compared to the pre-quiz, improving an average score of 3.9 to 5.4 out of 10. These data indicate that introduction of a short, standardized curriculum on patient safety in medical laboratory science education programs can improve students' awareness and knowledge of the laboratory's role in ensuring patient safety. This is a preliminary step in the process of creating a culture of patient safety among practicing and future medical laboratory professionals.

Predictive Value of Reverse Triiodothyronine in Diagnosis of Subclinical Hypothyroidism

Stacy E. Walz, PhD, MT(ASCP), Pam Towery, EdS, RD, LD, Arkansas State University, Jonesboro, Arkansas

Subclinical hypothyroidism (SCH) affects close to 10% of the general population, and can be difficult to diagnose. Although symptoms are often mild and non-specific, they can negatively impact a person's quality of life. Most cases of SCH do not necessarily require treatment, but diagnosis is important so affected individuals can be more closely monitored and further testing can be performed to identify the underlying cause. There are many laboratory tests available to physicians to aid in diagnosing dysfunction of the thyroid gland. Thyroid stimulating hormone (TSH) is the recommended screening test for thyroid dysfunction. Our study's purpose was to evaluate the utility of an esoteric test called reverse triiodothyronine (rT3) as a possible addition to the laboratory testing menu for diagnosis of SCH. Reverse T3 values over 32

nanograms per deciliter can indicate suppression of the thyroid gland, even when all other tests of thyroid function come back normal. We utilized anonymous, retroactive data on 322 patients from a local physician's office to assess the rT3 test's predictive value in the diagnosis of SCH. The study sample consisted of non-pregnant women between the ages of 18 and 65 who had been evaluated at this physician's office. The positive predictive value, or specificity, of rT3 was found to be 100%, and its negative predictive value, or sensitivity, was found to be 81.2%. The purpose of this study was not to suggest replacement of the TSH with rT3 as the screening test for assessment of thyroid dysfunction. Rather, we wish to increase laboratory professionals' awareness of this esoteric test such that they can have informed conversations with their physician customers about appropriate test selection for patients with possible subclinical hypothyroidism.

Control Material Comparability Assessed by Sigma-metrics

Sten Westgard, MS, Westgard QC, Inc., Madison, Wisconsin, Daryush Mirlohi, BS, Thermo Fisher Scientific, Fremont, California

The goal of this study was to determine the Sigma-metrics of two control materials on the same instrument. For twelve analytes (albumin, alanine aminotransferase, aspartate aminotransferase, creatinine kinase, glucose, creatinine, triglycerides, total protein, lactate dehydrogenase, digoxin, magnesium, and amylase) control materials from two different vendors (MAS, Bio-Rad) were run on the same instrument (Dade Dimension RxL) at the ICM (Cardiac Institute of Montreal) from July 2nd to December 8th, 2014.

Control materials are a critical analytical quality monitor for laboratories. Vendors of control material often market their products based on price and convenience of use. Too often the analytical quality of the control material is assumed and overlooked. Rarely is the actual analytical performance measured. Six Sigma metrics provide an objective comparative technique. For laboratories seeking consolidation or reduction in costs, the first step should be to confirm that no quality is compromised when switching between control vendors. Imprecision was calculated from at least 20 control measurements. Bias was

calculated as the difference between the observed mean and the target value. Quality Requirements were selected mainly from CLIA proficiency testing criteria. The Sigma-metric equation $[(TEa - Bias) / CV]$ was used to calculate the Sigma-metric at each level, and an average Sigma-metric was then calculated for each analyte. The results showed that the controls displayed comparable Sigma-metrics for a large majority of the analytes (75% or 9 out of 12). However, for a significant number of analytes (3 out of twelve or 25%), the MAS controls had higher Sigma-metrics than the Bio-Rad controls and would indicate different QC designs. This finding proves that control materials cannot be assumed to be identical. Sigma-metrics allow an objective comparison of the performance of control materials and can identify when it is safe or even ideal to switch vendors.

Design and Validation of a Survey Questionnaire for the Assessment of Physician Transfusion Medicine Knowledge

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The purpose of this study was to design and validate survey questionnaire for the assessment of baseline transfusion medicine knowledge of attending physicians, residents and medical students of Georgia Regents Medical Center (GRMC). Blood transfusion is a lifesaving intervention but also poses increased and unnecessary risk to transfusion recipients. Furthermore, variability in transfusion practice between hospitals and within hospitals is common. Physician education is one of the key elements which determines the effectiveness of any blood system. To assess baseline transfusion medicine knowledge, a survey instrument was developed and validated using expert construct opinion. The survey was then electronically administered to physicians at GRMC. Seventy-eight completed physician surveys were received and analyzed. The internal consistency of survey items was established using Cronbach's alpha which yielded a value of 0.77, indicating good reliability of the survey as a measurement tool. The overall mean score for the knowledge assessment ranged from 41% to 82%. The lowest and highest correct scores were from the family

medicine and pathology specialty groups respectively. Of the respondents, 28% indicated no transfusion medicine education in the past five years while 32% had less than an hour of transfusion medical education during the same time period. All specialty groups indicated the need for more transfusion medicine education.

A Case Study on Histoplasmosis

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A 45-year old male with a past medical history of kidney/pancreas transplant 10 years prior with primary failure of the pancreatic allograft, presented to an outside hospital with bright red blood per rectum. He progressed to multi-system organ failure (MSOF) due to massive GI bleed with poor localization. He was transferred to Ochsner Medical Center in severe acute respiratory distress, on 100% fraction inspired oxygen (FiO₂) and 20 of positive end-expiratory pressure (PEEP). His lactic acid was over 20. The complete blood count revealed severe leukocytopenia with decreased hemoglobin and hematocrit. Upon examination of the peripheral blood smear by a Medical Laboratory Scientist, intracellular yeast was identified. The smear was reviewed by the pathologist, who commented that the morphology was compatible with histoplasma, and a histoplasma serological test was ordered. The result of the histoplasma test was positive. The clinical impression was that he contracted the yeast infection by an injury of the knee, and due to the patient's immunosuppression, he began to decompensate with respiratory failure, and was unable to fight off the infection. After several weeks, he succumbed despite intensive intervention.

Associations between Toll-Like Receptor 4 Expression, Angiotensin II and Oxidized Low Density Lipoprotein in Human Subjects

Angela Zaki, MHS-CLS, Shaina Mccaskill, MHS-CLS, Gloria Slaon, MS, MT(ASCP), Joseph Cannon, PhD, Georgia Regents University, Augusta, GA

Toll-like receptor 4 (TLR4) is a pattern recognition receptor that induces an inflammatory response when activated by lipopolysaccharides or damage-associated

molecular pattern molecules. Expression of TLR4 on peripheral blood mononuclear cells has been associated with increased body fat content. Since adipose tissue produces leptin, and women produce more leptin per gram of adipose tissue than men, we hypothesized that TLR4 expression would be related to circulating leptin concentrations, with greater expression in women. Blood samples from 26 healthy men and women, 20-60 years of age, were collected and tested. The expression of TLR4 was evaluated by western blot. Leptin and other relevant blood parameters were measured by immunoassay. TLR4 expression was not related to serum leptin concentration, and although serum leptin was >3x higher in women ($P = 0.01$), there were no sex differences in TLR4 expression. Therefore, our primary hypothesis was disproved. However, high density lipoprotein and estradiol were negatively correlated with TLR4 expression in a multiple regression ($R = 0.53$, $P = 0.03$). In addition, the known association between angiotensin II and oxidized LDL was augmented when TLR4 expression was higher ($P = 0.03$). Overall, these data support the concept that angiotensin II-induced reactive oxygen species production is influenced, in part, by TLR4 expression. This work was supported by NIH grant HL 093663.

Oral Presentation Abstracts

Misdiagnosis of Acute Lymphoblastic Leukemia – The Need for the DCLS on Grand Rounds

Deborah Josko, PhD, Rutgers, The State University of New Jersey – School of Health Related Professions, Newark, NJ

A 56 year old female was seen at a walk up clinic for an infected ingrown hair in the inguinal area. The boil was lanced and drained and 14 days of Penicillin was prescribed. Two weeks later the infection did not subside and appeared to be worsening. The patient went to the emergency department where she was later admitted and placed on combination IV antibiotics. Routine blood work including blood cultures were drawn. Day three after admission the infection did not improve despite the antibiotics and topical ointment. In addition, she was diagnosed with idiopathic thrombocytopenic purpura due to her low platelet count of 25,000. During clinical rounds all hospital personnel on the case: physicians, physician assistants,

PharmDs as well as the nursing staff were perplexed as to why her platelet count was so low and attributed it to the infection. The patient's family member was a PhD Clinical Laboratory Scientist (CLS) and was called in as a consult. Within 30 seconds, the CLS identified the problem. Although the top portion of the CBC was within normal limits, the differential count revealed 53-55% reactive lymphocytes that went unnoticed for three days. The follow up revealed the presence of blast cells which was later confirmed as Acute Lymphoblastic Leukemia by flow cytometry. The patient was transported to another facility where she began aggressive chemotherapy. Unfortunately the patient expired two months after diagnosis.

If a DCLS was present and consulted on the case with the healthcare team, the abnormality would have been detected on admission. Would the outcome have been different? Most likely no, but at least the patient would have had an accurate diagnosis in a timelier manner. This case validates the importance of having laboratory representation on the floors during grand rounds.

Effects of *Ligusticum porteri* on Human Peripheral Lymphocytes

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Ligusticum porteri has been implicated in boosting the immune responses and treatments for diseases. However, little information about its medicinal effects have been validated. This study investigated the cytotoxicity, anti-inflammatory, and anti-oxidant effects of *L. porteri* ethanolic root extracts on peripheral blood lymphocytes (PBLs). The lymphocytes were incubated with different concentrations of the root extracts (0, 50, 100, 200, and 400µg/ml) and harvested every 6 hours for 3 days. The protective effect of the herb against oxidative damage was determined by inducing oxidative stress with the administration of 50µM of hydrogen peroxide (H_2O_2). The proliferation of PBLs after 2 days incubated with 200µg/ml and 400µg/ml of *L. porteri* were 2 and 2.5 fold higher than the control ($p < 0.05$). Treatments at lower concentrations (50, 100µg/ml) did not boost cell viability. Addition of 400µg/ml *L. porteri* to PBLs significantly enhanced levels of Interleukin-2, Interleukin-10, and Interferon- γ ($p < 0.05$). There were no significant changes in malondialdehyde and

glutathione levels, and superoxide dismutase and catalase activities in the PBLs non-treated and treated groups ($p>0.05$). The addition of 400 μ g/ml of *L. porteri* significantly reduced lipid peroxidation and protein oxidation in H₂O₂-challenged PBLs by 94.34% and 26.45% respectively after 2 days. There were noticeable increases of 17.46% and 55.23% in antioxidant enzyme superoxide dismutase and catalase activities induced by oxidative-damaged PBLs after treatment with 400 μ g/ml of *L. porteri* for 2 days. Our data suggests that *L. porteri* may enhance the immune responses and protect PBLs against oxidative stress.

Characterization of Laboratory Professionals' Consultation Role in Clinical Decision Support

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An exploratory study was conducted to document and characterize medical laboratory practitioners' (MLP) consultations with other health providers regarding utilization of clinical laboratory (CL) information in a tertiary care academic medical center. An 11-week sample of electronic and face-to-face consultations (n=325) between medical laboratory professionals (MLP) and other healthcare providers (hospital-based users of laboratory information) was described and characterized by time initiated, medical service, urgency status, and type of provider initiating the consultation. Logistic regression was used to categorize independent variables, i.e., complexity, medical subject area, and testing cycle phase involved that predicted disposition of consultations. Statistically significant differences were found in the frequency and types of consultations among medical services and healthcare providers. Results indicate that MLP consultations vary in complexity when examined by phase of testing cycle addressed, medical subject area, and CL section. Complexity, including medical subject, predicts consultation disposition. These findings can be used to define MLP evidence-based practice (EBP) roles, establish priority for quality management, and increase patient safety and quality care. Findings support a significant MLP CL consultative practice which is largely uncharacterized in formal job

analyses/descriptions, in MLP formal and continuing education curricula, and in the healthcare literature.

Evaluation of a Centrifugal Ultrafiltration Device in the Measurement of Free Testosterone

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Measurement of circulating testosterone is important to evaluation of androgen disorders. In circulation the majority of testosterone binds to sex hormone binding globulin and albumin. However, clinical evidence indicates that the concentration of circulating free testosterone correlates better with biological activities than total testosterone. There are two ways to separate free testosterone from protein bound testosterone: equilibrium dialysis and ultrafiltration. The centrifugal ultrafiltration device by Vivaproducts filters based on molecular weight. In this study we aimed to evaluate the performance of the Vivaproducts filters with a molecular weight cutoff of 30,000 daltons in separating serum free testosterone in comparison with a Millipore ultrafiltration device. Thirty nine patient serum samples were split and filtered by the Millipore ultrafiltration device and the Vivaproducts ultrafiltration device. The filtrates were measured for testosterone by radioimmunoassay. The free testosterone levels ranged from 0.9 to 312.8 pg/ml. Deming regression showed a slope of 1.047, r of 0.9748, and an error of estimate of 15.16. The mean difference between the two filtration devices was 5.8%. In conclusion, the centrifugal ultrafiltration device by Vivaproducts was shown comparable to the Millipore ultrafiltration device in determination of free testosterone.

Comparison of the use of Hemoglobin A1c Methodologies

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The increasing prevalence of diabetes mellitus, particularly type 2, has been recognized as a public health concern. At least 29.1 million Americans have diabetes and an estimated 1.9 million new cases of diabetes are diagnosed every year in the United States.

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An additional 87.3 million Americans are estimated to be pre-diabetic. The cost of diabetes is rising, with healthcare expenses in the United States estimated to be as high as \$192 billion by the year 2020. Hemoglobin A1c (HbA1c) serves as a gold standard for the diagnosis of diabetes or pre-diabetes in individuals. HbA1c measurements are crucial for monitoring the health of diabetics and serves as a principle consideration when tailoring medication and treatment plans. The purpose of this study was to compare the use of differing methodologies based on National Glycohemoglobin Standardization Program Certified Methodologies for measurement of HbA1c in the monitoring of blood glucose management in patients with DM.

Additionally, we examined discrepancies among the various methodologies. We found that immunoassay was the most prevalent methodology used (n=88) in the 169 testing platforms reviewed. Several genetic factors and hemoglobin variants were found to interfere with automation commonly used in HbA1c determination. Furthermore, point-of-care instrumentation studies were analyzed to reveal several inconsistencies in the reporting of results. The comparison of different testing methods utilized to monitor glycemic control in diabetic patients culminated in an understanding of the distinguishing capabilities and shortcomings involved in HbA1c and additional methodologies employed in the clinical laboratory.