ASCLS Annual Meeting 2010: Official Abstracts of Submitted Papers, Case Studies and Posters

Anaheim, CA

The following abstracts have been accepted for presentation at the 2010 American Society for Clinical Laboratory Science (ASCLS) Annual Meeting and Clinical Laboratory Exposition to be held July 27 through July 31 in Anaheim, CA. Abstracts are reviewed by appropriate representatives of the ASCLS Abstract 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. Room assignments will be listed in the final program.

ORAL RESEARCH PRESENTATIONS
Thursday, July 29, 11:30am-12:30pm at the Anaheim Convention Center
Friday, July 30, 2:30-3:30pm at the Hyatt Regency

POSTER PRESENTATIONS
Tuesday and Wednesday, July 27 and 28, 10:00am-4:30pm; Thursday, July 29, 9:30am-Noon at the Anaheim Convention Center; *Authors will be present on Wednesday, July 28, 2010 from 10:30am to Noon to discuss their work and answer questions.*

Oral Research Abstracts

Antimicrobial Combination Therapy for *Pseudomonas aeruginosa:* Which is Best?
Nicholas M. Moore, MS, MT(ASCP), Christopher Crank, PharmD, Maribeth Flaws, PhD, SM(ASCP)SI, Jane Stevens, MS, MT(ASCP), SM(ASCP), Rush University Medical Center, Chicago, IL

Pseudomonas aeruginosa has been implicated in serious infections resulting in significant morbidity and mortality. Empiric therapy regimens are initiated before susceptibility testing of the suspected pathogen(s) is complete. There is much debate whether physicians should utilize an antibiotic combination when treating infections. The purpose of this retrospective, cross-sectional study was to examine antimicrobial susceptibilities of *P. aeruginosa* and to see if the combination of a beta-lactam with an aminoglycoside offered a higher probability of coverage against *P. aeruginosa* than a beta-lactam plus a quinolone or monotherapy with a beta-lactam alone. Five hundred and one clinical isolates identified as *P. aeruginosa* from February 2008 through January 2009 were examined from inpatients and outpatients at Rush University Medical Center, a tertiary care academic medical center in Chicago, Illinois. Overall, antibiotic susceptibilities for each drug tested decreased in 2008 as compared to 2007 with the exception of piperacillin/tazobactam (up to 90% susceptible from 68% in 2007). All combinations of beta-lactams tested with levofloxacin, gentamicin, tobramycin or amikacin were statistically significant ($p<0.05$) with the exception of piperacillin/tazobactam plus levofloxacin ($p=0.136$), indicating that *P. aeruginosa* would be susceptible to these antimicrobial combinations. Overall, empiric combination therapy that included a beta-lactam plus levofloxacin or an aminoglycoside increased the probability that *P. aeruginosa* would be susceptible to at least one of the antimicrobial agents.

Comparing phenotype and PLD1 activity during biofilm production in clinical isolates of *Candida albicans*
April L Harkins, PhD, MT(ASCP), Bridget Nelson, Rezvaneh Ghasemzadeh, Marquette University, Milwaukee, WI

Candida albicans biofilm formation has been shown to consist of multiple phenotypes (hyphae, pseudohyphae and yeast cell) and reports have shown the strain of *C. albicans* can influence the physical heterogeneity of the

biofilm. The purpose of this study was to evaluate the various biofilm architectures produced by *C. albicans* clinical isolates and to measure the phospholipase D1 (PLD1) activity during formation. PLD1 hydrolyzes phosphatidylcholine (PC) to phosphatidic acid (PA), a potent second messenger molecule in many cellular signaling processes, and a free choline. *C. albicans* PLD1 is involved in hyphal development *in vitro* and PLD1 deficient yeast are avirulent compared to wildtype in mouse models of candidiasis. Currently, there are no studies of PLD1 during biofilm formation. Studying lipid modifying enzymes (PLD1) in biofilms may elucidate signaling molecules as potential antifungal targets. This is especially important for medically-significant infections with high mortality rates. In this study, *C. albicans* biofilms were grown on silicone squares for 65 hours and cells were imaged and harvested for PLD1 activity assays. The PLD1 activity increased during biofilm formation compared to planktonic cells, with differences seen depending on the phenotype present in the biofilm. In conclusion, PLD1 activity plays a role in the development of *C. albicans* biofilms. In most cases, *C. albicans* isolates from "sterile" sites (i.e. blood or tissue) form more hyphae and more complex biofilms and had a higher amount of PLD1 activity than those isolates taken from "non-sterile" sites (i.e. sputum and genital tract).

Correlation of Student Assignment Submission with Examination Scores

Michelle S. Kanuth, PhD, MLS(ASCP)CMSBB, Jose H. Salazar, MS, MLS(ASCP)CM, University of Texas Medical Branch, Galveston, TX

Most students submit their assignments on or before the due date routinely. However, a few students get into the habit of frequently being late with assignments. These habitual procrastinators appear to be more likely to perform poorly on examinations. There are many studies on procrastination in studying and test scores, but nothing regarding other types of assignments. One class of thirty (30) Clinical Laboratory Science students was tracked through three (3) courses, CLLS 4417 Hematology and Coagulation II, CLLS 3326, Methodology Development and Assessment, and CLLS 4415 Immunology and Immunohematology. Submission of assignments for each student in each course was assessed to be either on time or late. On-time was defined as an average submission of the course assignments by 11:59 PM on the due date. Late submission was defined as an average submission of the course assignments at any time after 12:01 AM on the day after the due date. Each student's examination average in each course was also determined. Late submissions were correlated with the examination score average using Fisher's exact t-test. Preliminary results for CLLS 3326: 11 students averaged <75% on examinations. There were 12 students who met the criteria for late submissions; 9 of these averaged <75% on examinations. For these data, Fisher's p=0.0533, suggesting a positive association between these events.

Detection of P2Y12-receptor Blockade (Clopidogrel) in Patients with Cardiovascular Disease by Accumetrics VerifyNow® P2Y12 and INNOVANCE® PFA P2Y*

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This investigation compared results of clopidogrel-induced platelet inhibition (P2Y12-receptor blockade) as measured by INNOVANCE® PFA P2Y* (P2Y*), a novel test for the Platelet Functional Analyzer-100 (PFA-100 system), and an FDA-cleared device, the Accumetrics VerifyNow® P2Y12 (VNP). Patients (n = 101) undergoing cardiac catheterization were tested either following administration of 300-600 mg clopidogrel (6-24 hours) or 75 mg clopidogrel for at least 7 days. Blood samples were collected in 3.2% and 3.8% buffered sodium citrate for P2Y, while only in 3.2% buffered sodium citrate for VNP. Pre-defined cut-offs for clopidogrel-induced P2Y12 receptor blockade were: P2Y >106sec and VNP >20% inhibition. Detection rates of P2Y12-receptor blockade for each method are shown in the table below.

Method:	P2Y 3.2%	P2Y 3.8%	VNP
Sensitivity(%)	66	86	60

Sensitivity is determined by dividing the number of true positives (TP) by the TP plus the false positives (FP) x 100%. Concordance is the agreement between two

methods cut-offs usually expressed in percent. The concordance (%) between P2Y and VNP was 75% for results generated in 3.2% citrate. In conclusion, the INNOVANCE® PFA P2Y method agrees with the VerifyNow® P2Y12 method for detection of P2Y12-receptor blockade induced by clopidogrel therapy. Inhibition of platelet function by anti-platelet medications such as clopidogrel, a P2Y12 receptor blockade drug, is one of the main line of defenses in protecting patients against cardiovascular disease involvement.

Plant-Derived Antifungal Activities

Marcia Lee, DVM, CLS (NCA), Richard L. Bretz, PhD, Gloria A. Wada, MS, Alison M. Herrick, Christine R. Barrett, Miami University, Oxford, OH

This study's purpose was to characterize antifungal effects of the inner gel portion of tropical *Aloe* species' leaves upon virulence factors of the opportunistic fungal pathogen *Candida albicans*. Although *Aloe* species have an abundance of anecdotal medicinal and cosmetic applications, few studies have scientifically investigated their antifungal effects. Three species of *Aloe,* e.g, *A. barbadensis, A. cameronii* and *A. arborescens*, were acclimated for approximately 2 years in an environmentally controlled, fungicide-free greenhouse. Inner gel from these plants was extracted and homogenized. Next, antifungal activities of their 0.2 micron filtrates were determined using established germination frequency assays and microscopic detection of morphologic aberrations against 10 strains of *Candida albicans,* including clinical isolates and quality control strains obtained from the American Type Culture Collection (Manassas, VA USA). Both clinical isolates and quality control strains of *C. albicans* exposed to each *Aloe* species demonstrated significant (p<0.1) inhibition of germination. Aberrations in germ tube morphology were qualitatively and quantitatively assessed. Antifungal activities of gel components from *A. arborescens* obtained through ethanol extraction and C18 reversed phase chromatography were investigated. Thus, the conclusion was that the data strongly supported the hypothesis that plant-derived filtrates obtained from the inner gel of *Aloe* leaves inhibit germination of *Candida albicans.* This work was significant because it elucidated *Aloe*-derived antifungal activities that may have diverse beneficial applications against opportunistic fungi.

Poster Presentation Abstracts

Assessment of Exposure to PACs in Asphalt Workers: Measurement of Urinary PACs and their Metabolites with an ELISA Kit

Barbara A. MacKenzie, **Jerome P. Smith**, Raymond E. Biagini, Belinda C. Johnson, Larry D. Olsen, Shirley A. Robertson, Deborah L. Sammons, Cynthia A.F. Striley, Cynthia V. Walker and John E. Snawder, Centers for Disease Control and Prevention, National Institute for Occupational Safety and Health, Cincinnati, OH

Asphalt pavers are exposed to polycyclic aromatic compounds (PACs), several of which are carcinogens, by inhalation and dermal contact. However, data linking exposure to asphalt with specific health effects are limited. Measurement of PACs and PAC metabolites as biomarkers of exposure provide a useful tool in assessing health effects. An enzyme-linked immunosorbent assay (ELISA) kit developed for the determination of PACs in water was adapted for measuring PACs and their metabolites in urine and then applied to a pilot asphalt worker PAC exposure study. Currently, liquid-liquid extraction with gas chromatography/isotope dilution high-resolution mass spectrometry (GC/HRMS) is the preferred method to determine urinary PAC metabolites. Although sensitive and specific, GC/HRMS is time consuming and costly. PAC ELISA is promising as a more rapid and less costly routine method for determining worker exposure to PACs in asphalt emissions. The ELISA method measured from 14-720 ng/ml 1-hydroxypyrene equivalents with a lower limit of detection of 14 ng/ml urine. Measurements of PAC metabolite equivalents in urine from asphalt exposed and control concrete workers using the ELISA had a good correlation (R=0.89), using Excel, to the sum of select GC/HRMS PAC urine metabolites (naphthalene, fluoranthene, phenanthrene, and pyrene) and the PAC ELISA results were indicative of potential asphalt exposure.

The Effectiveness of Digital Microscopy as a Teaching Tool in the Clinical Laboratory Science Curriculum.

Demetra C. Castillo, MAdEd, MT (ASCP), Rush University, Chicago, IL

As the technology of medicine advances, the need for innovative techniques also grows. An essential component of the practice of Clinical Laboratory Science (CLS) is the microscope. While the microscope is the instrument of choice in many CLS related procedures, it is not without its drawbacks. The high cost of maintenance associated with the microscope has led to an increased demand for more cost effective methods. One such method is digital microscopy. It has converted all of the features of the traditional microscope to a computer driven software. It has been speculated that the implementation of digital microscopy facilitates understanding of morphology in the areas of pathology and histology. The aim of this study is to investigate the effectiveness of digital microscopy as a teaching tool in didactic coursework within the field of Clinical Laboratory Science. The implementation of digital microscopy will facilitate students' understanding of morphology and lead to improved assessment scores. Students enrolled in the hematology course reviewed known study slides using both traditional and digital microscopy methods and were assessed with unknown slides using both methods. A paired t-test was performed and it was determined that the implementation of digital microscopy produced statistically significant exam score improvement (Mean (digital) = 85.67% +/-6.84%); Mean (traditional) = 82.93% +/- 9.21%) ($p<0.05$). Exam score improvement was directly related to the ability of the student in accurately identifying key elements of the unknown peripheral blood smears. It appears that digital microscopy may be an effective learning tool.

Extracorporeal Membrane Oxygenation Support for Pediatric Acute Respiratory Distress Syndrome

Erin Meister, University of Massachusetts Dartmouth, N. Dartmouth, MA and Massachusetts General Hospital, Boston, MA; Eileen Carreiro-Lewandowski, MS, CLS, University of Massachusetts Dartmouth, N. Dartmouth, MA

Extracorporeal Membrane Oxygenation (ECMO) use in neonatal patients is well documented. Its use in adult and pediatric populations, however, is less clearly defined. In this case study, a 17 year-old male motor vehicle accident victim suffered severe trauma including bilateral lung contusions with pneumatoceles, several fractures to his femur, forearm, ribs, and thoracic vertebrae, plus splenic and liver lacerations. He was eventually transferred to Massachusetts General Hospital. The patient required intubation with mechanical ventilation and was diagnosed as having Pediatric Acute Respiratory Distress Syndrome (ARDS). Laboratory results revealed dropping oxygen (O2) saturation levels (66% at arrival-reference ranges 97-100%) despite 100% oxygen therapy. Due to increasing complications, the patient was placed on venous-arterial ECMO for cardio-pulmonary support and to avoid multi-system organ failure. Blood gas results pre-ECMO revealed a severe respiratory acidosis (pH 7.33, aPCO2 44 torr, HCO3 22-reference ranges 7.35-7.45, 35-42 torr, 24-28 mmol/L, respectively) and hypoxemia (aPO2 37 torr, O2 saturation 66 %-reference ranges 80-100 torr, 97-100%, respectively). Post-ECMO blood gas results showed significant improvement (pH 7.43,aPCO2 37 torr, aPO2 251 torr, O2 saturation 100%). The patient continually improved and was weaned to venous-venous ECMO and eventually discharged to a rehabilitation center for further recovery.

This case study illustrates the importance of laboratory data in effective use of ECMO in non-neonatal populations leading to expanding use of ECMO in treating critically ill patients.

Methylenetetrahydrofolate Reductase Enzyme Mutation and Anti-cardiolipin Antibodies in Association with 2nd Trimester Fetal Demise

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A 28-year-old woman was referred to a reproductive specialist for evaluation of infertility of three years duration. Through the aid of in-vitro fertilization, the patient was able to conceive. The pregnancy progressed without complication until the twenty-sixth week when the fetus died in-utero. An autopsy on the fetus revealed the presence of a large stricture in the umbilical cord.

Causes of umbilical cord strictures are unknown but it has been proposed that they may be related to antenatal thrombosis formation in either the cord or placenta. Subsequent laboratory testing on the mother revealed two abnormalities, a homozygous C677T mutation on the methylenetetrahydrofolate reductase enzyme (MTHFR) gene and elevated anti-cardiolipin antibodies. A defect in the MTHFR enzyme can lead to a deficiency of folate, B6 and B12 which in turn may cause hyper-homocysteinemia, a risk factor for thrombosis. Anti-cardiolipin antibodies have been linked to thrombotic tendencies due to their action against negatively charged phospholipids. Prenatal therapy for women with defects leading to thrombosis includes daily low dose aspirin and supplements of B6, B12 and folate. During pregnancy, administration of low molecular weight heparin is advised.

Saliva Samples Collected from Various Methods are Excellent Materials for Cellular Analysis

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The presence of cells in saliva has been described in literature. In fact, DNA / RNA from these cells are used to provide information for genomic studies. Little is known about their composition, except the presence of epithelial cells. The purpose of this study was to characterize cellular populations in saliva samples collected from various methods. Saliva samples were collected by the draining (DRA), draining after gum massage (GUM), or gargling (GAR) method. The absolute number of cells, flow cytometry analysis, and morphology study were used together to describe these samples. As noted previously, epithelial cells were present in all three collection methods. The presence of CD45 positive leukocytes was more pronounced in DRA and GUM methods. Among these leukocytes, mononuclear cells were the major population. The goal here is to provide information on types of cells in saliva. In addition, the potential differences in cellular composition will help us choose the proper collecting method for certain projects. For example, GAR method will be the method of choice for detecting malignancy in oral epithelium. DRA method will provide a population from minimally disturbed samples to study oral mucosal immunity. Cells collected from GUM method will give us a snapshot of cellular interactions within gum tissues. In summary, saliva samples, with significant amount of cells, are excellent materials for high content analysis. Through proper selection of collection methods and correlating morphology with functional implications, we are able to advance saliva diagnostics, and to gain insight on oral mucosal immunity.

Utah Medical Education Council Statewide Laboratory Sciences Workforce Survey

JoAnn Fenn, MS, MT(ASCP), University of Utah; Boyd Chappell, MBA, Utah Medical Education Council; Rebecca Christiansen, Utah Public Health Laboratories, Salt Lake City, UT

This survey was directed by the Utah Medical Education Council (UMEC) with consultation from a coalition of clinical laboratory professionals in Utah. It was a first attempt to gather data on the state's clinical laboratory workforce and provide information on age profiles, credentials, vacancies, length of time to fill vacancies, and rural vs. urban workforce issues. A voluntary survey was distributed to 6504 laboratorians, including Clinical Laboratory Scientists(CLS)and Clinical Laboratory Technicians(CLT) working in CLIA certified laboratories across the state. A total of 2548 persons responded, giving a 39.2% response rate. Data was analyzed using SPSS, Excel, and Microsoft Access. The 744 CLS respondents had a mean age of 43.8, with 39.2% $\geq$50 years in 2005-2006; in rural laboratories 47.2% of CLS were $\geq$50 years, and in several rural counties, 100% of CLS were $\geq$50 years. The 166 CLT respondents had a mean age of 37.7 years. Most CLS and CLT respondents worked in hospitals and reference laboratories. Certification was noted in 89.7% of CLS and 61.2% of CLT respondents. In 2008, a follow-up survey of the state's two major employers of CLS/CLTs showed vacancy rates of 39.7% for CLS and 21.5% for CLT. Average time to fill vacancies was 1.6 months and 1.4 months, respectively. This data suggests that rural areas in Utah will be in dire need to replace retiring CLSs over the next 10 years, and Utah training programs must remain viable to meet the needs of the major laboratory facilities in the state.