CANADIAN JOURNAL OF
INFECTIOUS DISEASES & MEDICAL MICROBIOLOGY
JOURNAL CANADIEN DES MALADIES INFECTIEUSES ET DE LA MICROBIOLOGIE MEDICALE

AMMI Canada – CACMID Annual Conference

April 4-6, 2013, Québec City, Québec

ORAL PRESENTATIONS

Thursday, April 4, 2013

Session A	A1-A5
Session B	B1-B5
Session C	C1-C5
Session D	D1-D5

Friday April 5, 2013

Session E	E1-E8
Session F	F1-F8
Session G	G1-G8
Session H	H1-H8

Saturday April 6, 2013

Session I	I1-I4
Session J	J1-J4
Session K	K1-K5

INNOVATION ACADEMY POSTER PRESENTATIONS

Thursday April 4 and Friday April 5, 2013	IA1-IA5

POSTER PRESENTATIONS

Thursday April 4 and Friday April 5, 2013	P1-P47

STUDENT POSTER PRESENTATIONS

Thursday April 4 and Friday April 5, 2013	SP1-SP45

AUTHOR INDEX

ORALS

THURSDAY APRIL 4
1600 – 1715
Room: 203

A1
ONCOLYTIC VIRUSES AS A POTENTIAL APPROACH TO ELIMINATE THE HIV RESERVOIR

C COSTINIUK[1,2], S COTE[2,3], F AL-GHAZAWI[2], L CARRASCO-MEDINA[3], C YOUNG[3], J ANGEL[1,2,3]
[1]The Ottawa Hospital; [2]University of Ottawa; [3]Ottawa Hospital Research Institute, Ottawa, ON

OBJECTIVE: Similar to cancer cells, HIV-infected cells differ from HIV-uninfected cells in that they have altered interferon signaling pathways, the apparent reason for the selectivity of certain oncolytic viruses (OVs). The objective of this study was to determine whether the OV recombinant Maraba virus (MG1) would have a greater propensity to target and kill HIV-infected cells compared to non-infected cells.

METHODS: U1, ACH-2, OM-10 and J1.1 cells, harbouring 1-2 copies of integrated proviral DNA per cell, were infected with varying multiplicities of infection (MOI) of green fluorescent protein (GFP)-encoded MG1. Controls included HIV-uninfected U937, A301, HL60 and Jurkat parent lines. CD4+CD25-HLADR- cells from 20 HIV-infected individuals on antiretroviral therapy were infected with MG1 and flow cytometry and MTT assay were performed to quantify MG1 infection and cell viability, respectively. PCR for total HIV DNA in cells, in addition to RT-PCR for total HIV RNA and ELISA for p24 antigen on cell-free supernatants, were performed after a 2-week stimulation period.

RESULTS: MG1 infected and killed a greater proportion of U1 than U937 cells at most MOIs tested but this was not observed in the other cell lines. With the initial experimental approach, MG1 did not appear to infect $CD4^+CD25^-HLADR^-$ cells and viability appeared preserved. Similarly, we were unable to detect any effect of MG1 on quantities of total HIV DNA in cells, or total HIV RNA or p24 antigen levels in supernatants.

CONCLUSION: MG1 infects and kills latently HIV-infected U1 cells to a greater degree than the HIV-uninfected parent U937 cells and may be a promising model to facilitate further studies of MG1 as a potential therapy for the eradication of latently HIV-infected cells. Further optimization of the experimental approach for primary cell experiments is required in order to determine the effect of MG1 on cells which constitute the HIV reservoir.

A2
A MAMMALIAN MODEL FOR INFLUENZA VIRUS AND *STAPHYLOCOCCUS AUREUS* CO-PATHOGENESIS

H WONG[1], F MIAN[1], S MUBAREKA[1,2]
[1]Sunnybrook Research Institute; [2]University of Toronto, Toronto, ON

OBJECTIVE: It is widely accepted that bacterial co-infection in patients with influenza contributes to poor outcome. *Staphylococcus aureus* has been implicated as an important co-pathogen. Our goal was to establish the murine model for influenza virus-*S. aureus* co-infection in our laboratory in order to study mechanisms of disease and ultimately use as a pre-clinical model.

METHODS: To establish sub-lethal doses of influenza virus and *S. aureus*, groups of BALB/c mice were inoculated intranasally with serial dilutions of recombinant A/Puerto Rico/8/1934(H1N1) (rPR8) or methicillin-resistant *S. aureus* (MRSA). Animal weights and mortality were recorded daily. We then determined whether animals co-infected with rPR8 and MRSA demonstrated a more severe disease phenotype compared to animals inoculated with a single pathogen. Thus, groups of BALB/c mice were inoculated with PBS, rPR8, MRSA, or rPR8 followed by MRSA 72h post-viral inoculation. Lungs were recovered and homogenates were plated for bacterial enumeration.

RESULTS: The sub-lethal range of rPR8 was 25-100 plaque-forming units (pfu). All mice inoculated with $\leq 1.33\times10^8$ cfu MRSA survived. In a co-infection experiment, survival was 100% in PBS and MRSA-inoculated groups, 40% in the rPR8-inoculated group and 0% in the co-infected group. Higher titers of MRSA were recovered from co-infected animals ($p=0.0015$).

CONCLUSIONS: A more severe disease phenotype was observed in a murine model of influenza virus- *S. aureus* (MRSA) co-infection compared to infection with either virus or bacteria alone. This model grants us the capacity to examine mechanisms of co-pathogenesis and evaluate therapeutic strategies.

A3
EVALUATION OF A COMMERICAL KIT (REALSTAR ADENOVIRUS PCR KIT 1.0) FOR QUANTIFICATION OF ADENOVIRUS IN PLASMA

S WONG[1], K PABBARAJU[1], D ZARRA[1], R TELLIER[1,2]
[1]Provincial Laboratory for Public Health (ProvLab); [2]Department of Microbiology, Immunology and Infectious Diseases, University of Calgary, Calgary, AB

OBJECTIVE: Adenoviruses (AdV) can cause serious disseminated infections in immunocompromised patients which can often be fatal. Cidofovir has been used in the treatment of disseminated infections; however, it has been associated with significant toxicity. Detection and quantification of AdVs using quantitative PCR can help to monitor response to therapy and improve the clinical management. In this study we have compared an in-house real-time PCR assay to the commercially available RealStar Adenovirus PCR kit (Altona), and to a previously evaluated Adenovirus R-Gene kit (Argene) for the quantification of AdV in plasma specimens.

METHODS: The commercial kits were evaluated by comparison to an in-house quantitative adenovirus PCR (Wong 2008) for sensitivity with different AdV serotypes including the newly discovered type 60. Reproducibility was performed using different viral loads of samples. Consecutive plasma samples collected from hematopoietic stem cell transplant (HSCT) recipients were used for the measurement of AdV viral load.

RESULTS: Both commercial kits detected all the adenovirus serotypes including representatives from the seven different species with a high sensitivity and reproducibility. Results were noticeably better than the in-house PCR for some serotypes including serotypes 14, 31 and 35.

CONCLUSION: The quantitative standards included in the kits were highly reproducible. The set-up of the standard curve and copy number calculation was easy to perform. These features are key advantages in a diagnostic laboratory for quantification assays in order to effectively and reproducibly monitor viral load in response to antiviral therapy.

A4
COMPARISON OF SIEMENS AND ABBOTT HIV AG/AB ENZYME IMMUNOASSAY (EIA) AND APPLICATION OF HIV NUCLEIC ACID TESTING (NAT) FOR ACUTE/RECENT HIV DIAGNOSIS

A MAK[1], N CHAHIL[1], A YU[3], M KRAJDEN[1,2]
[1]Public Health Microbiology and Reference Laboratory; [2]University of British Columbia; [3]BC Centre for Disease Control, Vancouver, BC

OBJECTIVE: To compare the Siemens and Abbott HIV Ag/Ab EIAs and to assess the utility of HIV RNA NAT to diagnose acute/recent HIV infection as part of a HIV pool NAT study.

METHODS: Between Sept 16 and Dec 6, 2012, 1910 samples were tested by Siemens Centaur 3rd, 4th gen (Ag/Ab) and Abbott Architect HIV Ag/Ab EIAs. Samples seronegative on all three assays underwent pooled HIV NAT. Samples that displayed weakly reactive signals on any assay underwent individual NAT whereas samples displaying strong signals underwent Western Blot (WB) testing.

RESULTS: 23 HIV reactive and 1864 HIV non-reactive samples were concordant by the Siemens 3rd and 4th gen and the Abbott HIV Ag/Ab EIAs. 18/23 concordantly reactive samples were WB reactive confirming established infection and did not undergo HIV NAT. 5/23 samples

displayed low level Ag/Ab signals and were HIV RNA NAT reactive. One EIA non-reactive sample was pooled HIV RNA NAT reactive confirming a pre-seroconversion HIV infection. All six samples represented acute/recent HIV infections. Of the 23 discordant HIV Ag/Ab samples after resolution, 6 were Centaur 4th gen non-reactive (NR) and Architect Equivocal; 4 Centaur reactive and Architect NR; 13 Centaur NR and Architect reactive. HIV RNA was not detected in all 23 discordant samples. The Centaur 4th gen EIA displays more frequent first time reactives that are NR on retesting.

CONCLUSIONS: HIV Ag/Ab EIA testing combined individual NAT of samples that display weakly reactive signals testing enables early discrimination of acute or recent HIV and can help rule out false positive Ag/Ab EIAs results.

A5

CLINICAL FACTORS THAT PREDICT NONCIRRHOTIC PORTAL HYPERTENSION (NCPH) IN HIV-INFECTED INDIVIDUALS: A PROPOSED DIAGNOSTIC ALGORITHM

N PARIKH[1], T KUSHNER[1], V MARTEL-LAFERRIERE[1], K CHILDS[2], D DROMANRAJU[1], ML VACHON[1], C TAYLOR[2], K AGARWAL[2], TD SCHIANO[1], M FIEL[1], DT DIETERICH[1]

[1]Mount Sinai School of Medicine, New York, New York, USA; [2]Kings College, London, United Kingdom

PURPOSE: NCPH is a clinical entity that has been described in HIV-infected patients and is associated with the use of certain antiretrovirals (ARV), especially didanosine (ddI). The aim of our study was to determine clinical factors that could be used to diagnose NCPH in HIV-infected patients.

METHODS: A retrospective case-control study was performed in 42 HIV-infected patients with NCPH and 76 without NCPH. Cirrhosis was excluded with the use of liver biopsy and/or transient elastography (TE).

RESULTS: Cases and controls were similar in age, race, and HIV duration. NCPH patients had a longer mean exposure to ARVs (15.2 vs. 7.5 years, $p < 0.01$) and a lower current CD4 count (344 vs. 661 mm^3, $p<0.01$), but had similar current CD4 % (27.0% vs. 34.2%, $p=0.547$). More cases than controls were exposed to ddI (92.9% vs 17.1%, $p<0.01$). Among the cases, 37 (88.1%) had esophageal varices (EV), 36 (85.7%) had splenomegaly, and 20 (47.6%) had ascites while only 1 patient (1.3%) had splenomegaly in the control group. The majority (52.4% [n=22]) presented with EV bleeding. Cases also had lower albumin (3.8 vs. 4.5 g/dl, $p<0.001$) and platelets (117 vs. $203\times10^3/\mu L$ $p<0.01$). Mean TE score was higher in the cases (10.6 vs. 5.3 kPa, $p<0.01$). Based on this analysis, we hypothesized a set of six criteria to diagnose NCPH in non cirrhotic HIV patients: history of EV, ascites, splenomegaly, thrombocytopenia, TE scores >8 kPa, history of ddI exposure. Forty-one cases met at least 2 out of 6 criteria (97.6%) vs 4 of the controls (5.3%).

CONCLUSION: In this cohort, meeting at least 2 of 6 proposed criteria resulted in a diagnostic sensitivity of 97.6% and specificity of 94.7% for NCPH in HIV patients. A larger, prospective trial is necessary to further validate these results.

THURSDAY APRIL 4
1600 – 1715
Room: 205A

B1

EVALUATION OF BRUKER MALDI-TOF MS AND SEPSITYPER KIT™ FOR THE DIRECT IDENTIFICATION OF ORGANISMS FROM STERILE BODY FLUIDS IN A CANADIAN PEDIATRIC HOSPITAL

M TADROS[1], A PETRICH[2]

[1]University of Toronto; [2]The Hospital for Sick Children, Toronto, ON

BACKGROUND: Blood stream and central nervous system infections are associated with high morbidity and mortality. Rapid identification of pathogens in sepsis and central nervous system infections is thus crucial for appropriate treatment decisions and has a potential impact on patients' outcomes.

OBJECTIVE: To evaluate Bruker SepsiTyper kit™ and Bruker MALDI-TOF MS for the rapid and correct identification of organisms from flagged positive blood culture broths by comparison to conventional identification methods.

METHODS: The Sepsityper™ kit is a commercially available kit (Bruker Daltonics) used for preparing cells directly from positive blood cultures broths. It comprises lysis and washing buffers that are used directly on 1 ml of positive blood culture broths followed by ethanol/formic acid extraction. Identification was done using matrix assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) and compared to standard phenotypic identification methods.

RESULTS: Seventy-three blood culture specimens and 7 CSF specimens were analysed. Identification to genus and species levels was obtained in 75/80 (93.8%) and 37/50 (74%) of blood culture broths respectively. Gram negative organisms were more correctly identified to the species level than gram positive organisms ($p=0.2$)

CONCLUSIONS: The MALDI-TOF MS and the Bruker SepsiTyper™ kit are promising tools for the direct identification of organisms from sterile body sites. Their good reliability and rapid turnaround time can likely improve patients' outcomes.

B2

REPRODUCIBILITY OF BIOTYPER (BT) AND VITEK MS (VTMS) MATRIX-ASSISTED LASER DESORPTION IONIZATION-TIME OF FLIGHT (MALDI) IDENTIFICATION (ID) OF BACTERIA TESTED UNDER DIFFERENT CONDITIONS

BM WILLEY[1], P LO[1], SM POUTANEN[1,2]

[1]Mount Sinai Hospital/University Health Network; [2]University of Toronto, Toronto, ON

OBJECTIVES: As many methods are used to isolate bacteria from patient samples, it is important to ensure ID reproducibility prior to using MALDI in a clinical setting. This study used common organisms plated to various agars to assess BT and VTMS ID accuracy when performed at various intervals during incubation.

METHODS: The following strains were included. 6 ATCC strains: *E. coli* (EC) 25922 *P. aeruginosa* (PA) 27853; *S. aureus* (SA) 29213 and 43300; and *E. faecalis* (Ent) 29212 and 51299 (*vanB*$^+$); and 4 clinical strains: EC (N10-505*ndm*+); PA; MRSA; *E. faecium* (*vanA*$^+$). 10uL of 0.5 McFarland suspensions were plated to 11 Oxoid agars as follows: EC to 5% sheep blood (SBA), chocolate (Choc), Brucella (Bruc), MacConkey w/out salt (MAC), MAC+CV/salt/cefpodoxime (McPod), Mueller-Hinton (MH), BrillianceCRE; PA to SBA, Choc, MAC, McPod; SA to SBA, Choc, Bruc, MAC, Colistin/Nalidixic Acid Blood (CNA), Denim Blue and *Staphylococcus* Chromogenic; Ent to SBA, Choc, CNA, Bruc, MAC, Brilliance VRE. BT and VTMS ID was done after 4-6h, 16-18h, 22-24h and 36h in O_2, CO_2 or anaerobic conditions at 35-37°C, as per each manufacturers' instructions.

RESULTS: Of 250 MALDI done per system, VTMS correctly identified 100% with high confidence [99.9% (all but 3); 99.7% (2 Ent); 98.7% (1 PA)]. BT correctly identified 87.6% with high scores (>1.9): 51/51 EC, 36/36 PA, 67/90 SA (74.4%) and 65/73 Ent (89%). BT had correct ID but low scores (1.7-1.8) for 18 SA and 4 Ent, and unreliable scores (<1.7) for 5 SA and 4 Ent (3 Ent on MAC had no peaks).

CONCLUSIONS: While both systems could ID strains grown on all agars under various conditions, VTMS was able to identify a significantly higher amount of organisms under more conditions with confidence (100% vs. 87.6%) ($P<0.0001$).

B3

PROSPECTIVE HEAD-TO-HEAD EVALUATION OF BIOTYPER (BT) AND VITEK MS (VTMS) SYSTEMS COMPARING MALDI-TOF (M) TO CONVENTIONAL (C) IDENTIFICATIONS (ID) OF BACTERIA AND YEASTS CARRIED OUT TO THE SPECIES (SPP.) LEVEL

BM WILLEY[1], G SMALL[1], I EDWARDS[1], K BERNARD[2], P LO[1], K WONG[1], DE LOW[1,3], SM POUTANEN[1,3]

[1]Mount Sinai Hospital/University Health Network, Toronto, ON; [2]National Microbiology Laboratory, Winnipeg, MB; [3]University of Toronto, Toronto, ON

OBJECTIVES: While M-ID will revolutionize patient care by improving speed and accuracy of organism ID, it is important that microbiology laboratories carefully assess which system is most compatible with their setting. This study evaluated the Bruker BT and bioMérieux VTMS (IVD) systems for accuracy and ease of use.

METHODS: From Dec 2011 to Jan 2012, 1111 unselected semi-consecutive isolates (bacteria and yeasts) from all specimen types were submitted on primary isolation media for M-ID. Remnants of colonies picked for C-ID were used for parallel BT and VTMS testing which was done following each manufacturer's instructions. 16S sequencing (NML) was used to arbitrate discrepancies as needed.

RESULTS: Among 1111 isolates tested, VTMS and BT ID agreed with C-ID (% [95% CI]) in 1079 (97 [96-98]) and 1035 (93 [92-94]) isolates, respectively, (P<0.0001). All 3 ID agreed in 994 (89 [87-91]) isolates comprising 41 genera/91 spp. 32 isolates had incorrect or no ID by VTMS: the ID was not in the IVD database for 21 (1.9%); 1 (0.09%) had an incorrect but 99.9% ID; 3 (0.3%) were correct to genus but uncertain at spp. level; 6 (0.5%) mucoid isolates produced insufficient peaks; 1 (0.09%) had an incorrect ID due to an error related to organism placement on the slide. 76 had incorrect or no ID by BT: 41(3.7%) had unreliable scores (>1 but <1.9); 16 (1.4%) had high scores (>1.9) and were correct to genus but the spp. was incorrect; 5 (0.5%) had high scores (>1.9) but had uncertain spp. ID; 5 (0.5%) were mucoid and had no peaks; and 9 (0.8%) with >1.9 scores had incorrect ID due to errors related to organism placement on the target.

CONCLUSIONS: Among this study dataset, VTMS IVD produced significantly more reliable ID than BT (97% vs. 93%; P<0.0001) with less technical errors (P=0.02).

B4

KIR GENE PROFILES IN TWO MANITOBAN ABORIGINAL COHORTS WITH HIGH RATES OF TUBERCULOSIS

M SHARMA[1,2], K BRAUN[1], C LOPEZ[1], S KIAZYK[1,2], L LARCOMBE[1], TB BALL[1,2], P ORR[1], J WOLFE[2]

[1]University of Manitoba; [2]Public Health Agency of Canada, Winnipeg, MB

OBJECTIVE: In Manitoba (MB), the incidence of tuberculosis (TB) in Canadian born Aboriginals is more than 24 times higher than that of non-Aboriginals. This, in combination with the increasing Aboriginal population in MB is likely contributing to the increasing TB incidence rates in this group. Profiling of killer immunoglobulin-like receptor (KIR) genes has been shown to correlate with ethnicity as well as disease states. The objective of this study was to compare KIR profiles of two MB Aboriginal cohorts with high rates of TB.

METHODS: Cohort A consisted of 93 Aboriginal samples (14 latent, 20 active, and 59 controls) collected in Northern MB communities. Cohort B consisted of 54 Aboriginal samples (5 latent, 35 active, and 14 controls) from a MB TB clinic. KIR profiles were determined using the Miltenyi Biotec KIR typing kit.

RESULTS: KIR frequencies in both cohorts were similar except for KIR1D, present in 38.71% of cohort A and 74.07% of cohort B ($P<0.0001$). When analysed by disease status, further distinction between these cohorts is seen: presence of KIR1D is higher in all TB disease status groups of cohort B, and present in 100% of latent TB cases; KIR2DS2/L2 frequencies are higher in cohort A; KIR2DS3 is absent from all latent samples in both cohorts. Twenty-two KIR profiles were identified in cohort A and 14 in cohort B, 11 of which overlap between the two groups.

CONCLUSION: This study shows the similarities and differences in KIR gene frequencies and profile distribution among two different cohorts of Aboriginals with high rates of TB. Further investigation of the immunological aspects of TB disease may lead to a better understanding of disease progression and novel treatment approaches.

B5

GENETIC ANALYSIS OF PRIMAQUINE TOLERANCE IN A PATIENT WITH RELAPSING VIVAX MALARIA

AT BRIGHT[1], T ALENAZI[2,3], S SHOKOPLES[4], J TARNING[5,6], G PAGANOTTI[7], N WHITE[5,6], S HOUSTON[2], E WINZELER[1], S YANOW[4,8]

[1]University of California, San Diego, California, USA; [2]University of Alberta Hospital, Edmonton, AB; [3]King Saud Bin Abdulaziz University for Health Sciences, Riyadh, Saudi Arabia; [4]Provincial Laboratory for Public Health, Edmonton, AB; [5]Mahidol University, Bangkok, Thailand; [6]Oxford University, Oxford, United Kingdom; [7]La Sapienza University, Rome, Italy; [8]University of Alberta, Edmonton, AB

OBJECTIVE: Patients with *Plasmodium vivax* malaria are treated with primaquine (PQ) to eliminate liver stage parasites and prevent relapse of the infection. PQ tolerance has been reported but not well characterized, and the genetic basis of treatment failure is unknown. We performed genetic analysis to identify parasite and host factors that may contribute to PQ tolerance in a patient with three consecutive *P. vivax* relapses that occurred without any possibility of re-infection.

METHODS: Parasite DNA isolated from blood samples collected during each infection [EAC01-03] was analyzed using whole genome capture followed by high-throughput sequencing. The patient's DNA was genotyped at four alleles from the cytochrome P450 (CYP) gene family: *CYP1A2*1C*, *CYP2B6*6*, *CYP3A4*1B* and *CYP2D6*4*. Plasma concentrations of PQ and its major metabolite, carboxy-primaquine (CPQ), were measured with a newly developed stereoselective bioanalytical LC-MS/MS method.

RESULTS: The three parasite isolates were found to be genetically related, but not identical, and are proposed to be meiotic siblings. All three contained nearly identical single nucleotide variants in genes homologous to known *P. falciparum* drug resistance genes including *pvdhps*, *pvmdr* and *pvmrp*. We also determined if the patient carried alleles in *CYP* genes that might also explain his treatment failure. The patient was homozygous for the wild-type allele in all four genes. Finally, we examined whether the patient metabolized PQ to carboxy-primaquine (CPQ), the main PQ metabolite found in plasma. The total PQ and CPQ concentrations were 90 ng/mL and 1042 ng/mL, respectively. These data demonstrate appropriate absorption of PQ and metabolism.

CONCLUSIONS: This case study demonstrates the feasibility of using molecular tools to better understand therapeutic responses to primaquine. The multiple relapses in the patient described here result from previously acquired hypnozoites that likely possessed a genetic profile rendering them tolerant to PQ, whereas host variation in drug metabolism appears an unlikely explanation for PQ failure.

THURSDAY APRIL 4
1600 – 1715
Room: 205B

C1
FIDAXOMICIN VERSUS VANCOMYCIN IN THE TREATMENT OF *CLOSTRIDIUM DIFFICILE* INFECTION: CANADIAN OUTCOMES

C LEE[1], T LOUIE[2], M GERSON[3], K WEISS[4], W ARNOTT[5], S GORBACH[6]

[1]St Josephs Healthcare, Hamilton, ON; [2]Foothills Medical Centre, Calgary, AB; [3]Humber River Hospital, Toronto, ON; [4]Maisonneuve-Rosemont Hospital, Montreal, QC; [5]Optimer Pharmaceuticals Toronto, ON; [6]Optimer Pharmaceuticals, San Diego, California, USA

OBJECTIVES: To compare the efficacy and safety of fidaxomicin (FDX) vs. vancomycin (VAN) in the treatment of *C. difficile* infection (CDI) based on post-hoc analysis of Canadian data from two randomized, double-blind international clinical trials.

METHODS: Patients with confirmed primary episode or first recurrence of CDI received FDX 200 mg twice daily or VAN 125 mg four times daily for 10 days. The modified intent-to-treat population consisted of 406 Canadian patients (from a total of 1105 patients). Patients were assessed daily for initial clinical cure and weekly for recurrence of infection within the 30 days after treatment completion. Additional analyses of specific patient subpopulations were performed and included concomitant antibiotics (AB), cancer (CAN), renal dysfunction (RD), *C. difficile* strain (BI vs. non-BI) and age (≥65 years). All subjects who received at least one dose of study medication and with at least one post dose safety assessment were evaluated for safety.

RESULTS: Cure rates were equivalent with FDX and VAN (90.1% vs. 92.2%, 95% CI: −7.7, 3.5) while recurrence rates were lower with FDX (14.4% vs. 28.0%, p=0.001). Lower rates of recurrence were observed with FDX in the following subpopulations: AB=16.2% FDX vs. 38.7% VAN (p=0.036); CAN=5.3% FDX vs. 29.6% VAN (p=0.061); RD=23.3% FDX vs. 37.5% VAN (p=0.142); non-BI=11.8% FDX vs. 28.3% VAN (p=0.004); and age ≥65=16.1% FDX vs. 30.9% VAN (p=0.026). Both treatment regimens had comparable overall safety profiles.

CONCLUSIONS: Consistent with results from the international trials, FDX was safe, efficacious and well-tolerated in treating CDI in Canadian patients. FDX was superior to VAN in reducing recurrence rates of a disease which is associated with high incidence of relapse and has high clinical and economic burden. Specific populations, such as those on AB or with other risk factors for recurrence, may benefit when initially managed with FDX.

C2
THE MOLECULAR EPIDEMIOLOGY OF *CLOSTRIDIUM DIFFICILE* IN CANADIAN HOSPITALS: 2007-2011

MR MULVEY[1], D BOYD[1], T DU[1], R HIZON[1], M MILLER[2], D GRAVEL[1], C FRENETTE[3,4], L HOLMES[5], K KATZ[6], P KIBSEY[7], A MCGEER[8], D MOORE[9], A SIMOR[10], K SUH[11], G TAYLOR[12], CANADIAN NOSOCOMIAL SURVEILLANCE PROGRAM (CNISP)

[1]Public Health Agency of Canada, Winnipeg, MB; [2]Jewish General Hospital; [3]Montreal General Hospital; [4]Royal Victoria Hospital, Montreal, QC; [5]BC Children's Hospital, Vancouver, BC; [6]North York General Hospital, North York, ON; [7]Victoria General Hospital, Victoria, BC; [8]Mount Sinai Hospital, Toronto, ON; [9]Montreal Children's Hospital, Montreal, QC; [10]Sunnybrook Health Sciences Centre, Toronto; [10]The Ottawa Hospital, Ottawa, ON; [12]University of Alberta Hospital, Edmonton, AB

OBJECTIVE: The emergence of hypervirulent strains causing serious *Clostridium difficile* infections (CDI) is a growing concern in Canada and *many other countries. This study examined the molecular epidemiology of C. difficile* over 5 years in Canada.

METHODS: Between 2007-11, all toxin positive stools identified in the months of April and May from participating CNISP sites were submitted to the NML. *C. difficile* was isolated using an alcohol shock procedure. PCR was used to detect *tcdA, tcdB, tcdC, cdtB,* and *tpi*. Isolates were typed by PFGE using *Sma*I. Etest was used to determine antimicrobial susceptibilities to metronidazole, vancomycin, clindamycin, rifampin, moxifloxacin, and tigecycline.

RESULTS: 2,119 *C. difficile* isolates were typed with 12 NAP types identified over the study period. There were 472 (22.3%) isolates that were not categorized as a NAP type. The most common strains identified were NAP1 (42.7%; n=904), NAP4 (12.2%; n=258), and NAP2 (8.4%; n=177). A significant decrease in NAP1 was observed from 2010 (n=169; 42.6%) to 2011 (n=137; 29.4%) (P<0.001). NAP2 dramatically decreased from 20.6% in 2007 (n=78) to 4.3% in 2011 (n=20) (P<0.001). NAP4 has significantly increased from 6.7% in 2007 (n=31) to 19.7% in 2011 (n=92) (P<0.001). No metronidazole, vancomycin, or tigecycline resistant isolates were identified.

CONCLUSIONS: The factors related to the trends in incidence and geographic distribution of NAP types in Canada remain a mystery. Further studies involving patient demographics and outcomes, infection control practices, and antimicrobial use practices are required to better understand these changes. Metronidazole and vancomycin remain appropriate for treatment of CDI.

C3
THE HYPERVIRULENT *CLOSTRIDIUM DIFFICILE* BI STRAIN IN CANADA: INCIDENCE, TYPE AND COMPARATIVE EFFICACY OF FIDAXOMICIN AND VANCOMYCIN

T LOUIE[1], K WEISS[2], L VALIQUETTE[3], W ARNOTT[4], S GORBACH[5]

[1]Foothills Medical Centre, Calgary, AB; [2]Maisonneuve-Rosemont Hospital, Montreal; [3]Université de Sherbrooke, Sherbrooke, QC; [4]Optimer Pharmaceuticals Canada, Toronto, ON; [5]Optimer Pharmaceuticals, San Diego, California, USA

OBJECTIVES: To determine the incidence and geographic distribution of the 'hypervirulent' *Clostridium difficile* BI strain in Canada and the relative efficacy of fidaxomicin (FDX) and vancomycin (VAN) in treating *C. difficile* infection (CDI). Post-hoc analysis of Canadian data from two randomized, double-blind international clinical trials was performed.

METHODS: Four hundred six Canadian patients with confirmed first episode or first recurrence of CDI received FDX 200 mg twice daily or VAN 125 mg four times daily for 10 days. Patients were assessed daily for clinical cure and weekly for recurrence of infection within the 30 days after treatment completion. Restriction endonuclease analysis (REA) was used to determine strain type.

RESULTS: 84.2% of patients had isolates available for REA typing. One hundred thirty-three patients were infected with the BI strain (38.9%), which was the most commonly detected strain. Other strains identified included non-specific (31.6%), Y (11.1%), G (9.4%), J (6.7%), K (1.2%), BK (0.3%), CF (0.3%), DH (0.3%), and L (0.3%). The incidence of the BI strain was highest in Ontario (58.9%), followed by Quebec (48.3%) and the Western Canadian provinces (British Columbia, Alberta, and Saskatchewan, 14.3%). No patients from other Canadian provinces or territories were enrolled. For the BI strain, cure rates were 83.3% for FDX and 88.1% for VAN (95% CI: -16.6, 7.4) while there was a 43% relative reduction in recurrence rates with FDX (16.4% [9/55] FDX vs. 28.8% [17/56] VAN, p=0.113).

CONCLUSIONS: 38.9% of Canadian patients who had isolates available for typing were infected with the BI strain. Most patients infected with the BI strain were from Ontario and Quebec. In BI infected patients, FDX was noninferior to VAN in the initial treatment of CDI while FDX-treated patients experienced a 43% reduction in CDI recurrence in this post hoc analysis.

C4

USEFULNESS OF TESTING MULTIPLE ISOLATES PER SPECIMEN FOR RELIABLE TYPING OF *CLOSTRIDIUM DIFFICILE* USING MMLVA TO ASSIST OUTBREAK INVESTIGATIONS

L MILLER[1], G BROUKHANSKI[1,2]

[1]Public Health Ontario Laboratories; [2]LMP, University of Toronto, Toronto, ON

BACKGROUND: Investigation of *Clostridium difficile* (CD) outbreaks requires reliable and sensitive method to establish transmission of outbreak strains. We evaluated use of MMLVA to improve typing reliability and turnaround time.

To utilize high resolution power of MMLVA and ability to detect multiple strains per specimens we evaluated effectiveness of typing 4 CD isolates per specimen.

METHODS: Seventy-five specimens from 10 medium-sized outbreaks (six to 10 specimens each) were selected for the analysis. Specimens were heat treated to select spores and plated on CDMN. DNA from 4 colonies was extracted by boiling in InstaGene matrix. To setup two PCR reactions per extract (10 µl) for MMLVA we used liquid handler (Biomek NXp) which was also used for setting fragment analysis on 3130xl Genetic Analyzer. Results were analyzed with BioNumerics software.

RESULTS: PFGE typing of specimens demonstrated that eight of 10 (80%) outbreaks had NAP1 as a predominant strain which complicates outbreak investigation. Out of 381 colonies tested with MMLVA 20 (5.2%) failed to amplify (likely because it was not CD) and in 51 (13.4%) one of two PCR reactions failed (likely due to issues with PCR setup). 310 (81.4%) were amplified successfully. Twenty-six specimens (8.4%) had multiple strains with MMLVA profiles which were completely different (n=3, 1%), different in two or three loci (2, 0.6%) or similar (21, 6.8%).

CONCLUSIONS: Our results show that testing 4 isolates per specimens with MMLVA has benefit of generating sufficient data for outbreak analysis in situations of occasional failure of PCR amplification and also when multiple CD strains are present per specimen. Automation of the procedure and decreasing volumes of PCR reactions allows testing without significant increase in the cost.

C5

IDENTIFICATION OF PEPTIDES ANTAGONISTS TARGETING TOXINS IN *CLOSTRIDIUM DIFFICILE*

J LEBLANC[1,2]

[1]QEII Health Sciences Centre; [2]Dalhousie University, Halifax, NS

INTRODUCTION: Pathogenesis of *C. difficile* is attributed to toxins TcdA and TcdB. This study aimed to identify peptides able to prevent activity of these toxins.

METHODS: Overlapping peptides were synthesized to mimic the sequence composition of in the TcdA or TcdB receptor binding domains (RBD) from historical and hypervirulent *C. difficile* (strains 630 and Nap1, respectively). Each peptide was evaluated using cell culture cytotoxicity neutralization assays (CCCNA). Briefly, TcdA and/or TcdB were purified from broth culture supernatants of 630 and Nap1 wild-type (WT) strains or their toxin-deficient mutants ($TcdA^-B^+$ or $TcdA^+B^-$). Double mutants ($TcdA^-B^-$) were used as controls. Normalized toxins amounts were added to FSK or HT29 cells in presence or absence of each peptide. Cytopathic effect (CPE) was expressed as a percentage ± SD from triplicate values (n=1000) obtained in three independent experiments.

RESULTS: Two peptides prevented TcdB-induced CPE in a dose-dependent manner. The peptides had identical sequences derived from libraries mimicking TcdB RBDs from strains 630 and Nap1. On FSK cells, complete peptide-mediated inhibition was observed for toxins derived from the WT strains and the $TcdA^-$ mutants. On TcdA-sensitive HT29 cells, inhibition of CPE was only observed with TcdB derived from the $TcdA^-$ mutants. Partial inhibition of CPE was observed for WT-derived toxins. The residual activity was attributed to TcdA since it was blocked with anti-TcdA antibody.

CONCLUSIONS: This study identified a peptide capable of inhibiting *C. difficile* TcdB *in vitro*. Inhibition of toxins using 'peptide antagonists' is a novel avenue of research which could lead to new adjunct therapies against *C. difficile* infection.

THURSDAY APRIL 4
1600 – 1715
Room: 205C

D1

ANTIMICROBIAL SUSCEPTIBILITY PROFILE OF *NEISSERIA GONORRHOEAE* ISOLATES IN THE PROVINCE OF QUÉBEC: 2010–2012

B LEFEBVRE[1], A LABBÉ[1], S VENNE[1,2], G LAMBERT[1], C FORTIN[1], A BOURGAULT[1], C TREMBLAY[1]

[1]Institut national de santé publique du Québec; [2]Ministère de la santé et des services sociaux du Québec, Québec, QC

BACKGROUND: Monitoring antimicrobial resistance of *N. gonorrhoeae* is necessary to detect trends in antimicrobial resistance, increases in minimum inhibitory concentrations (MICs) and to assist in the development of treatment guidelines. The emergence of antibiotic resistance, especially cephalosporins is a major public health challenge.

METHODS: *N. gonorrhoeae* strains isolated in clinical laboratories throughout the province were submitted to the Laboratoire de santé publique du Québec where their susceptibility profile to azithromycin, cefixime, ceftriaxone, ciprofloxacin and spectinomycin was determined by the agar dilution method as recommended by CLSI.

RESULTS: A total of 2212 strains (902 in 2010, 797 in 2011 and 513 in 2012 [partial data]) were analyzed (women 632 [28.5%], men 1572 [71.1%], unknown 8 [0.4%]). All strains were susceptible to cefixime, ceftriaxone and spectinomycin, 816 (36.9%) strains were resistant to ciprofloxacin and 26 (1.2%) resistant to azithromycin (MIC ≥2 mg/L). Distribution of azithromycin MICs were 2 mg/L (n=3), 4 mg/L (n=2), 8 mg/L (n=6) and 16 mg/L (n=15). Among the strains resistant to azithromycin, 5 were also resistant to ciprofloxacin. Reduced susceptibility (RS) to cefixime (MIC = 0.125 mg/L or 0.25 mg/L) was identified in 63 strains (7.0%) in 2010 (MIC = 0.125 mg/L [n=61] and 0.25 mg/L [n=2]), 78 (9.8%) in 2011 (MIC = 0.125 mg/L [n=72] and 0.25 mg/L [n=6]) and 24 (4.7%) in 2012 (MIC = 0.125 mg/L [n=20] and 0.25 mg/L [n=4]). These strains were sensitive to azithromycin, spectinomycin and ceftriaxone, but resistant (97.6%) to ciprofloxacin. Five strains with RS to ceftriaxone (also RS to cefixime) were identified (MIC = 0.125 mg/L); one in 2010, one in 2011 and three in 2012. The proportion of RS to cefixime strains is not higher among men (7.1%) than women (8.4%). For 2012, more complete data will be presented including MICs for tigecycline, ertapenem and gentamicin.

CONCLUSIONS: Reduced susceptibility to cefixime has emerged in Québec although it remains at very low level. Furthermore, resistance to another treatment option, azithromycin, is also emerging. This highlights the need to continue our resistance monitoring program to support public health interventions.

D2

MOLECULAR CHARACTERIZATION OF METHICILLIN-RESISTANT *STAPHYLOCOCCUS AUREUS* (MRSA) ISOLATES FROM PATIENTS WITH BACTEREMIA IDENTIFIED BY THE CANADIAN NOSOCOMIAL INFECTION SURVEILLANCE PROGRAM (CNISP), 2008-2010

GR GOLDING[1], AE SIMOR[2], A MOUNCHILI[3], J SHURGOLD[3], MR MULVEY[1], D GRAVEL[3], K KATZ[4], M LOEB[5], A MCGEER[6], P KIBSEY[7], K WEISS[8], J EMBIL[9], S SMITH[10], C FRENETTE[11], CANADIAN NOSOCOMIAL INFECTION SURVEILLANCE PROGRAM (CNISP)

[1]National Microbiology Laboratory, Winnipeg, MB; [2]Sunnybrook Health Sciences Centre, Toronto; [3]Public Health Agency of Canada, Ottawa; [4]North York General Hospital, Toronto; [5]McMaster University, Hamilton; [6]Mount Sinai Hospital, Toronto, ON; [7]Royal Jubilee Hospital, Victoria, BC; [8]Maisonneuve-Rosemont Hospital, Montreal, QC; [9]Health Sciences Centre, Winnipeg, MB; [10]University of Alberta, Edmonton, AB; [11]McGill University Health Centre, Montreal, QC

OBJECTIVE: Bloodstream infections (BSI) caused by MRSA are associated with substantial morbidity and mortality. It was the goal of this study to characterize MRSA recovered from blood cultures of patients identified through CNISP.

METHODS: Surveillance for MRSA BSIs was performed in 54 Canadian hospitals. The first blood culture isolate from each affected patient was characterized by *spa* typing, antimicrobial susceptibility, SCC*mec* typing, and PCR for PVL.

RESULTS: From 791 available isolates, the majority of MRSA BSI isolates were CMRSA2 (USA100/800; ST5) (57.0%) and CMRSA10 (USA300; ST8) (26.8%). Among MRSA BSI meeting the definition of a healthcare-associated infection, 69.7% were CMRSA2 and 16.7% were CMRSA10, while among those meeting community-associated infection criteria 18.1% were CMRSA2 and 58.8% were CMRSA10. The prevalence of these two strain types also differed from western to eastern provinces, with CMRSA10 predominating in the west (41.7% of all western MRSA BSI isolates) and CMRSA2 in the east (86.0%). Among all MRSA BSI isolates, 28.3% harboured the PVL encoding gene. The two predominant SCC*mec* types were SCC*mec*II (59.1%) and SCC*mec*IV (36.1%). Overall, high rates of resistance to ciprofloxacin (88.2%) and erythromycin (87.7%) were observed and all isolates were susceptible to vancomycin, daptomycin, and linezolid.

CONCLUSIONS: CMRSA2 and CMRSA10 were the two predominant strains identified. Significant differences in the acquisition of these two strain types from healthcare/community settings and different regions of Canada were observed. Strain and patient variables associated with morbidity and mortality of MRSA BSIs remains to be elucidated.

D3

ANTIFUNGAL SUSCEPTIBILITY OF INVASIVE *CANDIDA* ISOLATES FROM CANADIAN HOSPITALS: RESULTS OF THE CANWARD 2012 STUDY

J FULLER[1], S JIWA[1], S SHOKOPLES[1], L TURNBULL[1], R RENNIE[1], H ADAM[2], M BAXTER[2], D HOBAN[2], GG ZHANEL[2]

[1]Provlab Alberta, University of Alberta, Edmonton, AB; [2]Medical Microbiology and Infectious Diseases, University of Manitoba, Winnipeg, MB

BACKGROUND: CANWARD is an ongoing national surveillance study that assesses pathogens causing infections in patients attending Canadian hospitals, as well as determines the prevalence of antimicrobial resistance in these isolates. Here we present the antifungal susceptibility data for candidemia isolates collected in 2012.

METHODS: *Candida* species isolated from bloodstream infections were collected from 11 participating medical centres during the 2012 study period. Antifungal susceptibility testing and interpretation was performed as per CLSI M27-S4 broth microdilution method and the recently modified breakpoints for fluconazole (FLUC), voriconazole (VORI), caspofungin (CASP), and micafungin (MICA).

RESULTS: Of the 290 *Candida* spp. collected, *C. albicans* (CA) was predominant (55.0%), followed by *C. glabrata* (CG, 16.5%) and *C. parapsilosis* (CP, 13.8%). MIC_{90} and susceptibility (S) values are below. Azole resistance was detected in three *C. albicans* and three *C. parapsilosis*, while echinocandin resistance was not.

		MIC_{90} (%S)					
	No.	AMB	ITRA	FLUC	VORI	CASP	MICA
CA	159	1.0 (-)	0.12 (98)	0.25 (98)	≤0.015 (99.5)	0.12 (100)	≤0.007 (100)
CG	48	1.0 (-)	1.0 (-)	8.0 (100*)	0.25 (-)	-	0.03 (100)
CP	40	1.0 (-)	0.12 (-)	1.0 (92.5)	0.03 (97.5)	1.0 (100)	1.0 (100)

*AMB, amphotericin B; ITRA, itraconazole; *, susceptible dose-dependent; -, data not available*

CONCLUSION: Canadian surveillance of antifungal resistance in invasive *Candida* infections is an important long-term objective of the CANWARD program. The species prevalence and MIC distributions of CANWARD candidemia isolates has remained unchanged for several years with no evidence of emerging resistance.

D4

EMERGENCE OF MULTIDRUG RESISTANT *SALMONELLA ENTERICA* SEROTYPE 4,[5],12:I:- HUMAN INFECTIONS IN CANADA

MR MULVEY[1], R FINLEY[2], V ALLEN[3], L ANG[4], S BEKAL[5], S EL BAILEY[6], D HALDANE[7], L HOANG[8], G HORSMAN[9], M LOUIE[10], L ROBBERTS[11], J WYLIE[12], M MCCRACKEN[1], S LANGNER[1], R AHMED[1], H TABOR[1], M GILMOUR[1]

[1]National Microbiology Laboratory, Winnipeg, MB; [2]Centre for Food-borne, Environmental and Zoonotic Infectious Diseases, Guelph; [3]Ontario Agency for Health Protection and Promotion, Toronto, ON; [4]Queen Elizabeth Hospital, Charlottetown, PEI; [5]Institut national de santé publique du Québec, Montreal, QC; [6]Saint John Regional Hospital, Saint John, NB; [7]Provincial Public Health Laboratory Network of Nova Scotia, Halifax, NS; [8]BC Centre for Disease Control, Vancouver, BC; [9]Saskatchewan Disease Control Laboratory, Regina, SK; [10]Alberta Provincial Laboratory for Public Health, Calgary, AB; [11]Newfoundland Public Health Laboratory, St John's, NL; [12]Cadham Provincial Laboratory, Winnipeg, MB

OBJECTIVE: Over the last decade a marked increase in *Salmonella enterica* serotype 4,[5],12:i:- with a core resistance to ampicillin, streptomycin, sulphonamide, and tetracycline (ASSuT) has been observed in Europe. This study describes the emergence and characterization of isolates of multidrug resistant *S.* 4,[5],12:i:- in Canada.

METHODS: Human clinical isolates of *S.* 4,[5],12:i:- were identified by provincial laboratories from 2003 to 2010 and submitted to the NML for further testing. Serotyping and phage typing were performed by standardized methods. MIC values were determined by the broth microdilution using the Sensititre. PCR was used to determine the presence and location of resistance genes. MLST was performed on a selected number of isolates.

RESULTS: A total of 26,251 *Salmonella* were submitted to the NML as part of the CIPARS. *S.* 4,[5],12:i:- accounted for a total of 766 isolates (2.9%), and the numbers increased from 42 (1.4%) in 2003 to 164 (4.8%) in 2010. The majority of isolates remained susceptible to all antimicrobials tested (n=456; 59.5%). The $ASSuT^+$ phenotype was observed in 11.9% (n=91) of *S.* 4,[5],12:i:- and it increased from two isolates in 2003 to 35 isolates in 2010. Two sequence types were observed. ST34 were associated with the ASSuT isolates (n=24; 37%) which contained bla_{TEM}, *strA-strB*, *tet*(B), and *sul2*. ST19 were associated with the ACSSuT phenotype and contained *strA-strB*, *sul2*, and *tet*(A). ESBL genes were found in four isolates and included $bla_{CTX\text{-}M\text{-}55}$, $bla_{CTX\text{-}M\text{-}57}$, and two isolates with $bla_{CMY\text{-}2}$.

CONCLUSIONS: S. 4,[5],12:i:- has significantly increased from 2003 to 2010 and is now the fifth most common serotype reported in Canada causing human disease. Similar antimicrobial resistance patterns, phage types, and sequence types have been observed in Europe.

D5

SURVEILLANCE OF CARBAPENEM-RESISTANT ENTEROBACTERIACEAE AND *ACINETOBACTER BAUMANNII* IN CANADA: RESULTS OF THE CNISP 2011-2012

LF MATASEJE[1], E BRYCE[2], D ROSCOE[2], DA BOYD[1], J EMBREE[3], D GRAVEL[4], K KATZ[5], P KIBSEY[6], M KUHN[7], A MOUNCHILI[4], A SIMOR[8], G TAYLOR[9], E THOMAS[10], N TURGEON[11], MR MULVEY[1]

[1]Public Health Agency of Canada, Winnipeg, MB; [2]Vancouver General Hospital, Vancouver, BC; [3]Universisty of Manitoba, Winnipeg, MB; [4]Public Health Agency of Canada, Ottawa; [5]North York General Hospital, Toronto, ON; [6]Victoria General Hospital, Victoria, BC; [7]Moncton Hospital, Moncton, NB; [8]Sunnybrook Health Sciences Center, Toronto, ON; [9]University of Alberta Hospital, Edmonton, AB; [10]Children's and Women's Health Centre, Vancouver, BC; [11]Hotel-Dieu do Quebec du CHUQ, Quebec City, QC

OBJECTIVE: Carbapenem-resistance complicates treatment of Gram-negatives and is a growing concern in Canada. This report describes the results of the Canadian Nosocomial Infection Surveillance Program (CNISP) for carbapenem resistant isolates.

METHODS: From Sept. 2011 to Aug. 31. 2012, all carbapenem reduced susceptible Enterobacteriaceae (ENT) and resistant *A. baumannii* (AB) were submitted to the National Microbiology Laboratory from participating CNISP hospitals. PCR identified carbapenemase-producing isolates. Antimicrobial susceptibility was conducted using Vitek2. PFGE was conducted to determine strain relatedness.

RESULTS: A total of 196 isolates (186 ENT and 10 AB) were identified from 14 hospital sites. 53% were from male patients, 58.5% were from patients >65. Most isolates were collected from medical wards (49%) or outpatient clinics (20.4%) and isolated from urine (38.8%) or rectal screens (28.6%). 76 carbapenemases (76/196, 38.8%) were identified from 74 isolates: 59 KPC, 3 NDM, 3 OXA-48, 3 SME and 8 others. Most KPCs were detected in *K. pneumoniae* (KP) (n=32) and *E. cloacae* (EC) (n=15). One AB isolate harbored 3 carbapenemases (IMP, OXA-23 and OXA-58). By PFGE analysis 2 KPC EC clusters were identified from central Canada. Fifty-eight percent of all KPC KP clustered to the ST258 pattern type. All but 2 isolates from this cluster were from a single site. Isolates harboring carbapenemases were resistant to meropenem (93.2%), ertapenem (92.8%), aminoglycosides (62.2%), ciprofloxacin (58.1%), tigecycline (27%) and colistin (2.7%).

CONCLUSION: The number of carbapenemase producing isolates identified is of concern especially in Enterobacteriaceae. Pan-resistance was observed in a *K. pneumoniae* harbouring KPC.

FRIDAY APRIL 5
1600 – 1800
Room: 203

E1

EVALUATION OF THE β LACTA™ TEST FOR THE RAPID DETECTION OF EXTENDED SPECTRUM BETA LACTAMASES IN *ENTEROBACTERIACEAE*

M TADROS[1], W SMALL[2], B TAM[2], R DEVLIN[2], Z HUSSAIN[2]

[1]University of Toronto; [2]St Michael's Hospital, Toronto, ON

BACKGROUND: Extended spectrum beta lactamases (ESBL) producing *Enterobacteriaceae* is a worldwide problem. The rapid detection of resistance to third generation cephalosporins from clinical specimens is crucial in early choice of the right therapeutic option. The ß LACTA™ is a simple and rapid test based on the cleavage of a chromogenic cephalosporin.

OBJECTIVES: To evaluate the performance of the ß LACTA™ test in the rapid detection of class A, AmpC and carbapenemases from *Enterobacteriaceae* by comparing it to the standard methods used to detect these enzymes.

METHODS: Ninety-six isolates from clinical and screening specimens were used for the evaluation. Fifty-seven isolates were producers of Class A ESBL according to conventional laboratory methods, 24 isolates were positive for AmpC, five isolates were carbapenemase producers and 10 isolates were negative for all the aforementioned enzymes.

RESULTS: For class A ESBL producing isolates, the ß LACTA™ test was positive in 52/57 isolates (91%), 4 isolates gave indeterminate results (7%) and 1 isolate was negative (2%). The test was able to detect AmpC production in only one of the positive isolates tested (4%). The ten negative isolates were all negative by the ß LACTA™ test. The overall sensitivity of the test was 65% and the specificity was 100%. The sensitivity of the ß LACTA™ test for detecting Class A ESBL and carbapenemase production was 89%.

CONCLUSIONS: Our preliminary evaluation showed that the ß LACTA™ test is a simple and rapid test that has a potential role for the detection of class A ESBL, and carbapenemase but not AmpC producing isolates of *Enterobacteriaceae*.

E2

EVALUATION OF THE CARBA-NP TEST FOR DETECTION OF *ENTEROBACTERIACEAE* AND *PSEUDOMONAS AERUGINOSA* PRODUCING CARBAPENEMASES

D BOYD, MR MULVEY

Public Health Agency of Canada, Winnipeg, MB

OBJECTIVE: The Carba-NP test is a phenotypic test designed to detect the hydrolysis of the beta-lactam ring of imipenem by an organism producing a carbapenemase. Briefly, a crude bacterial extract is incubated with a phenol red-imipenem solution and if hydrolysis occurs the solution will transition from red to orange to yellow over time due to a drop in the pH. The test as published is said to be 100% specific and sensitive. The objective of this study was to validate the test with a panel of organisms harbouring various classes of carbapenemases and non-carbapenemase producers with reduced susceptibility to carbapenems.

METHODS: The Carba-NP test was performed as published except the volumes of all solutions were halved. The test was carried out on *E. coli*, *Klebsiella* spp., *Enterobacter* spp., *Citrobacter* spp., *Morganella* spp., and *Pseudomonas aeruginosa* harbouring the class A carbapenemase KPC (n=24); class B carbapenemases NDM (n=17), VIM (n=5), and IMP (n=2); and class D carbapenemases OXA-48 and OXA-181 (n=15). In addition 6 *Serratia marcescens* harbouring SME (class A) and 5 *E. cloacae* harbouring NMC (class A) were tested. Thirty-five non-carbapenemase producers, most harbouring ESBLs and with reduced susceptibility to carbapenems, were also included.

RESULTS: All non-carbapenemase producers were negative. Amongst carbapenemase producers a positive results was obtained for 22/24 KPC producers, 12/17 NDM producers, 3/5 VIM producers, 0/2 IMP producers, 3/15 OXA-48/181 producers, 1/6 SME producers, and 1/5 NMC producers. Blinded calls by up to 3 personnel were highly variable due to the subtlety of the colour change in many cases.

CONCLUSION: We have obtained conflicting results as compared to the originally published data for Carba-NP. The high number of false negatives amongst carbapenemase producers in our limited sample size suggests further studies are required before the Carba-NP test can be adopted as a routine confirmatory test for the identification of carbapenemase producing strains. The test may have value where KPC and/or NDM producers are endemic. Optimisation of the test by using more concentrated extract and/or different carbapenems eg, meropenem, ertapenem should be explored.

E3

VALIDATION OF KIRBY-BAUER TESTING FOR CEFAZOLIN SUSCEPTIBILITY IN ENTEROBACTERIACEAE USING A BLOOD AGAR PURITY PLATE FROM A COMMERCIAL ID/AST SYSTEM

S EDAGIZ[1], P LAGACÉ-WIENS[1,2], J KARLOWSKY[1,2], A WALKTY[1,2], M ALFA[1,2]
[1]University of Manitoba; [2]Diagnostic Services of Manitoba; Winnipeg, MB

OBJECTIVE: The Clinical and Laboratory Standards Institute breakpoints for cefazolin against Enterobacteriaceae that were published in have been revised by the Subcommittee on Antimicrobial susceptibility testing. The new breakpoints, published in January 2011 are: susceptible, ≤2 µg/mL; intermediate, 4 µg/mL; and resistant, ≥8 µg/mL. This created a problem for clinical microbiology laboratories that use commercial instruments to perform routine antimicrobial susceptibility testing (AST), because the performance of the instrument at the lowest concentration ranges has not been established and for some instruments the dilution range does not span the new breakpoint. The objective of our study was to validate the use of disc diffusion for cefazolin using the purity plate from the commercial ID/AST system rather than performing disc diffusion on a second plate using conventional methodology, in order to streamline workflow and reduce cost.

METHODS: Fifty *Enterobacteriaceae* isolates including AmpC ß-lactamase-producing organisms were tested. Routine identification and susceptibility testing was done using the Vitek 2 (Biomérieux). A 30 µg cefazolin disc was placed on the first quadrant of the purity plate in parallel with reference Kirby-Bauer disk diffusion testing. We compared the antimicrobial susceptibility testing results generated by the two methods.

RESULT: The categorical agreement was 88%. Six (12%) minor errors occurred. There were no major or very major errors.

CONCLUSION: Cefazolin disk diffusion on a purity plate resulted in >10% total errors and thus does not provide a reliable alternative to the standardized CLSI Kirby Bauer disk diffusion test.

E4

RE-EVALUATION OF THE CRITICAL CONCENTRATION FOR ETHAMBUTOL DRUG SENSITIVITY TESTING OF *MYCOBACTERIUM TUBERCULOSIS* ON THE BACTEC MGIT 960™

S CHRISTIANSON[1], D VOTH[2], M SHARMA[1,2], J WOLFE[1]
[1]Public Health Agency of Canada; [2]University of Manitoba, Winnipeg, MB

BACKGROUND: Upon the transition from the Bactec 460™ (B460) to the Bactec MGIT 960™ (M960) method for rapid broth based drug sensitivity testing for *M. tuberculosis*, critical concentrations (CC) were assessed to ensure correlation between results from the B460 and M960. Despite reproducibility issues, a CC of 5.0 µg/mL was established for ethambutol (EMB) on the M960. The aim of this study was to correlate resistance on the B460 with the M960 and mutations in the *emb*CAB operon, which are often associated with EMB resistance.

METHODS: We selected 32 B460 EMB resistant and 10 B460 EMB sensitive isolates and compared the data to M960 MIC data and *emb*CAB sequencing.

RESULTS: B460 EMB sensitive strains were sensitive on the M960 (MICs<=2.0µg/mL). There were 16 strains resistant at the high B460 CC of 7.5 µg/mL (12: M960 MIC ≥8.0 µg/mL, 3: M960 MIC = 4.0 µg/mL and 1: M960 MIC = 5.0 µg/mL). 14/16 isolates resistant at the B460 CC of 2.5 µg/mL but sensitive at 7.5 µg/mL had M960 MICs of 4.0 and 5.0 µg/mL. There were 18 isolates with M960 MICs = 4 or 5 µg/mL. Many had inconsistent results at the M960 CC of 5.0 µg/mL, but all were resistant at 4.0µg/mL. 13/18 had *emb*B mutations, which are highly correlated with EMB resistance. All 18 were identified as resistant using the B460 CC of 2.5 µg/mL.

CONCLUSION: This data shows that an M960 CC of 4.0 µg/mL more accurately identifies strains with a lower level of EMB resistance, and more closely emulates B460 results. Sequence data confirms that many of the strains that fall into the MIC range of 4.0-5.0 µg/mL have mutations in the *emb*CAB operon, specifically in the *emb*B gene, the most common site of mutations in EMB resistant strains.

E5

COMPARISON OF CHICAGO SKY BLUE STAIN WITH ROUTINE POTASSIUM HYDROXIDE FOR THE RAPID DETECTION OF FUNGAL ELEMENTS IN SUPERFICIAL SPECIMENS

J MUSA, D BRAMBERGER, C BECHARD, S NIGH, L NOORPOUR, B GANDHI, A BELHAJ, H ALMOHRI
LifeLabs Medical Laboratory Services, Toronto, ON

OBJECTIVE: To compare Chicago Sky Blue (CSB) stain with routine potassium hydroxide (KOH) wet preparations in detection of fungal elements from superficial specimens, and to determine the best alternative to the calcofluor stain currently used in our laboratory.

METHOD: A total of 150 clinical samples (known 50 positives and 100 negatives) were examined using both methods and results compared to the reference method by the calcofluor stain method routinely used in the laboratory. Slides were initially read under regular light microscopy with KOH. The preparations were then stained with CSB by adding 1 drop of 1% CSB solution, and read again at 30 minutes. Comparison was done in reading time required and in sensitivity, specificity, and reproducibility. For CSB stained slides; 25 negative specimens were randomly selected to be read after 24 hours to check for late positive results. 25 positive specimens were randomly selected to be read after 48, 72, and 96 hours to check for viability of the CSB Stain.

RESULTS: CSB stain had a sensitivity, specificity and reproducibility of 100%. KOH showed a specificity and reproducibility of 100% but sensitivity of 96%. CSB stain use enabled a faster reading technique than the KOH method. Less time was required to read the preparations, providing an increased productivity of 46%. Of 25 CSB stained negative specimens that were read after 24 hours to check for late positives, no positives were detected. All the positive preparations examined after 48, 72, and 96 hours had fungal elements that remained detectable.

CONCLUSION: Chicago Sky Blue stain is an acceptable, reliable and inexpensive staining technique that can be used as an alternate method to calcofluor stain for the rapid detection of fungal elements. It is superior to KOH wet preparation.

E6

COMPARISON OF SPECTROPHOTOMETRIC AND VISUAL ENDPOINT DETERMINATION FOR ANTIFUNGAL SUSCEPTIBILITY TESTING OF *CANDIDA* ISOLATES USING BROTH MICRODILUTION

S JIWA, S SHOKOPLES, B JANSEN, J FULLER
Provlab, University of Alberta, Edmonton, AB

BACKGROUND: The Clinical and Laboratory Standards Institute (CLSI) recommends visual endpoint determination in the broth microdilution (BMD) standards guidelines for yeast susceptibility testing. European standards using a similar methodology promote spectrophotometric endpoint measurement. In this study, we evaluated the performance of spectrophotometric analysis of *Candida* BMD tests.

METHODS: BMD susceptibility testing, as per CLSI M27-S4, was performed on a challenge set of *Candida* isolates. Growth endpoints were determined at 24 h of incubation visually and spectrophotometrically on a microplate reader for fluconazole (FLUC), voriconazole (VORI), caspofungin (CASP), and micafungin (MICA). Essential agreement (EA) of spectrophotometry with the reference visual reading was defined within ± 1 doubling dilution.

RESULTS: 135 *Candida* isolates were tested, including 77 *C. albicans* (Ca), 24 *C. parapsilosis* (Cp), and 16 *C. glabrata* (Cg). Greater than 90% of each species were in EA for FLUC, VORI, and MICA. For CASP, the EA was 100% and 92% for Cg and Cp, respectively, but was substantially lower for Ca at 66%. Ca growth morphologies significantly changed (diffused) in microplate wells at higher CASP concentrations, while Cp and Cg morphologies with CASP remained constant.

CONCLUSION: Spectrophotometric measurement of antifungal susceptibility has the potential to improve upon the labours of BMD interpretation and showed a high degree of agreement in this study with the reference method for the majority of species:drug pairs. It is likely that morphological growth changes lead to the disparity observed with CASP and Ca and further analyses will be required before the utility of spectrophotometry in CLSI BMD methods can be determined.

E7

COMPARISON OF TWO COMMERCIAL URINE TRANSPORT PRESERVATIVE SYSTEMS

P DALEY, Y GILL, W MIDODZI
Memorial University, St John's, NL

OBJECTIVE: Urine collected for bacterial culture may overgrow during transportation.
METHODS: Consecutive urine samples received in an academic hospital laboratory were cultured at baseline and inoculated into Copan UriSwab™ and BD VacutainerTM at 22°C. Sterile container at 22°C and 4°C were used as controls. Quantitative culture of uropathogens was performed on Blood and MacConkey and interpreted according to laboratory policy at 24, 48 and 72 hours. Per cent with significant growth were compared to baseline using logistic regression.
RESULTS: 507 urine samples were included. 95 (19.0%) urine samples had significant growth on receipt. Odds ratio (95% C.I.) for significant growth of uropathogens after 72 hours of storage was 17.58 (13.92-22.19) for sterile container at 22°C, 6.49 (3.91-10.76) for BD at 22°C, 5.08 (3.06-8.41) for Copan at 22°C and 4.36 (2.63-7.21) for sterile container at 4°C.
CONCLUSION: Urine preservation systems provided similar performance to refrigeration for up to 72 hours. Copan was slightly superior to BD, but not significantly different. The use of a urine preservative may allow unrefrigerated transport from remote locations without significantly greater overgrowth as compared to refrigerated transport.

E8

COMPARISON OF TWO BACTERIAL STORAGE SYSTEMS: THE PRO-LAB DIAGNOSTICS MICROBANK™ CRYOVIAL SYSTEM COMPARED TO GLYCEROL CITRATE FREEZER VIALS (GCFV) AT −20^{0}C AND −70°C OVER AN EXTENDED PERIOD OF TIME

A GERULL-UPTON, A CAREY, J DANIEL, S MILBURN
London Health Sciences Centre, London, ON

OBJECTIVE: The purpose of the evaluation was to compare the ability of the Pro-Lab Diagnostics Microbank™ Cryovial System (MB) and Glycerol Citrate Freezer Vials (GCFV) to maintain the viability of fastidious bacteria at −20°C and −70°C.
METHODS: A total of 119 isolates comprised of 35 *Haemophilus* spp, 27 *S. pneumoniae*, 14 *Neisseria* spp, 13 *Campylobacter* spp, 12 *Moraxella* spp, 6 *Pasteurella* spp, 4 *Eikenella* spp, 4 *Kingella* spp, 2 *Capnocytophaga* spp and 2 *Cardiobacterium* spp. Each of the isolates was inoculated into 4 vials (2 MB and 2 GCFV) as per the manufacturer's instructions. One set was frozen at −20°C. The other in −70°C. Each vial was then sub cultured every 3 months and the growth tracked and recorded as 3+ ,2+ , 1+ and No Growth (organism death).
RESULTS: At −20°C neither the MB nor GCFV was able to maintain viability, with the majority of the species, past 24 months. The remaining organisms surviving to 30 months or greater had varying results: *Campylobacter* spp and *Haemophilus* spp each have one surviving isolate. Of the surviving *Neisseria* spp, 36% were MB vials, 7% GCFV. *Moraxella* showed the largest variation with 100% of the MB isolates as compared to 25% GCFV.
The results for the −70°C vials showed that both MB and GCFV had a good total survival rate to 30 months: percent viability varied. *Haemophilus* spp at 3+ growth recovery was 51% for MB, 9% GCFV. *S. pneumoniae* was 41% MB, 7% CGFV. *Neisseria* 36% MB, 21% CGFV. *Moraxella* spp was 100% MB , 92% CGFV. *Pasturella* spp 67% MB, 50% CGFV. *Eikenella* spp 100% , 25% CGFV. *Campylobacter* spp generally showed poorer viability with those surviving to 2+ growth at 38% MB , 15% GCFV. *Capnocytophaga* spp and *Cardiobacterium* spp were both 100% in both MB and CGFV to 2+ viability. *Kingella* spp had the poorest results with both MB and CGFV having 1+ recovery at 30 months.
CONCLUSION: Our results show that in general is not good practice to freeze the fastidious organisms at −20°C in either system but there is significant difference in viability when the MB was used versus the GCFV when stored at −70°C.

FRIDAY APRIL 5
1600 – 1800
Room: 205A

F1

WHOLE GENOME ANALYSES OF *LEGIONELLA PNEUMOPHILA* SEROGROUP 1 OUTBREAK IN QUEBEC CITY IN 2012

P PLANTE[1], S LÉVESQUE[2], N MENDIS[3], H CHAREST[2], F RAYMOND[1], L ROBITAILLE[1], I GOUPIL-SORMANY[4], F DESBIENS[4], S FAUCHER[3], C TREMBLAY[2], J CORBEIL[1]
[1]Université Laval, Québec; [2]LSPQ/INSPQ, Sainte-Anne-de-Bellevue, QC [3]McGill University - Macdonald Campus, Sainte-Anne-de-Bellevue; [4]DRSP de la Capitale-Nationale, Québec, QC

OBJECTIVE: During the major *L. pneumophila* serogroup 1 (LP1) outbreak that occurred in Quebec City in the summer of 2012, an environmental strain recovered from a cooling tower was matched to the human strains using pulsed-field gel electrophoresis (PFGE). It is considered to be the epidemic strain, most isolates recovered from the environment displaying different PFGE profiles. Whole genome sequencing (WGS) was performed to identify putative virulence markers that could have contributed to this major outbreak.
METHODS: A total of 40 LP1 isolates were analyzed by WGS including 23 obtained from patients, 9 from environmental sources geographically related to the outbreak, 3 from patients with LP1 pneumonia unrelated to the outbreak and 5 isolates from a former outbreak in the same area in 1996. WGS was performed on an Illumina MiSeq platform using the Nextera XT kit for library preparation. Sequence assembly was achieved using the Ray assembler. Genomes were also compared to 5 fully sequenced LP1 reference strains available in public databases.
RESULTS: The epidemic strain is the only one harboring two secretion systems, lacking LPG1057 (a diguanylate cyclase implicated in biofilm formation) and possessing 2 plasmids. Strains lacking Lpg1057 were confirmed to produce less biofilm than other strains. Type II secretion system were present in all strains. Type IV secretion system were found in all strains except for unrelated patient strains and 1996 strains.
CONCLUSION: Differences in genomic composition were observed between the epidemic strains and unrelated strains recovered from the environment during the outbreak. Deficit in biofilm formation, which could promote aerosol distribution, could in part explain the spread of the epidemic strain during the outbreak. These data suggest that molecular assays targeting genetic markers associated with virulence in *Legionella* are valuable tools for laboratory investigations of outbreaks. These highlights the importance of thorough and regular cooling tower disinfection measures to avoid dissemination of strains that may harbor a higher aerosol potential.

F2

WHOLE GENOME SEQUENCE ANALYSIS OF *SALMONELLA* ENTERITIDIS OUTBREAK ISOLATES IN BRITISH COLUMBIA

KA MACDONALD[1,2], A PACCAGNELLA[1], MN MCCABE[1], D EISLER[1], M IMPERIAL[1,3], J PRADINUK[1], B AUK[1], J FUNG[1], JL ISAAC-RENTON[1,3], A MCNABB[1], P TANG[1,3], L HOANG[1,3], N PRYSTAJECKY[1,3], W HSIAO[1,3]

[1]BC Public Health Microbiology and Reference Laboratory, Provincial Health Services Authority, Vancouver, BC; [2]National Microbiology Laboratory, Public Health Agency of Canada, Winnipeg, MB; [3]Department of Pathology and Laboratory Medicine, University of British Columbia, Vancouver, BC

OBJECTIVE: Outbreaks of salmonellosis occur regularly in BC, and *Salmonella enterica* serovar Enteritidis (*S. enteritidis*) is the most commonly isolated organism. *S. enteritidis* is highly clonal with a relatively conserved core genome and minor variations across lineages. Pulsed Field Gel Electrophoresis (PFGE), therefore, has limited discriminatory power for certain *S. enteritidis* lineages, and the results are not useful for epidemiological investigations. Whole Genome Sequencing (WGS) can provide detailed information on genomic alterations needed for surveillance and source tracking initiatives. This work assessed strain diversity of outbreak isolates of S. Enteritidis in BC from 2003 to 2011 via WGS.

METHODS: WGS was performed on an Illumina MiSeq platform. All isolates were sequenced to a minimum of 30x genome coverage, sufficient for high quality mapping analysis. 21 isolates of S. Enteritidis, from 6 outbreaks were sequenced to gain an initial understanding of strain diversity within and between outbreak strains. Reads were mapped to the S. Enteritidis reference genome P125109.

RESULTS: Preliminary Single nucleotide polymorphism (SNP) analysis demonstrated that we could separate isolates with the same PFGE patterns or phagetypes into clusters corresponding to distinct outbreaks. SNP analysis and whole genome annotation also provided the resolution necessary to disambiguate an outbreak that is likely to have multiple sources of contamination.

CONCLUSIONS: WGS analysis was a useful tool for assessing and understanding sequence diversity of a highly clonal organism, such as S. Enteritidis. Further improvements in time and cost of WGS analysis will result in great improvements in understanding strain evolution, outbreak investigation, and source tracking.

F3

THE USE OF WHOLE GENOME SEQUENCING TO DIFFERENTIATE CANADIAN NAP1 *CLOSTRIDIUM DIFFICILE* ISOLATES CURRENTLY INDISTINGUISHABLE BY PULSE FIELD GEL ELECTROPHORESIS AND RIBOTYPING

T LYNCH[1], N KNOX[1], P MABON[1], A PETKAU[1], T DU[1], M MILLER[2], D GRAVEL[3], C FRENETTE[4], L HOLMES[5], K KATZ[6], P KIBSEY[7], A MCGEER[8], D MOORE[9], A SIMOR[10], K SUH[11], G TAYLOR[12], G VAN DOMSELAAR[1], M GRAHAM[1], MR MULVEY[1]

[1]National Microbiology Laboratory, Public Health Agency of Canada, Winnipeg, MB; [2]SMBD-Jewish General Hospital, Montreal, QC; [3]Centre for Communicable Diseases and Infection Control, Public Health Agency of Canada, Ottawa, ON; [4]McGill University Health Centre, Montreal, QC; [5]Children and Women's Health Center of British Columbia, Victoria, BC; [6]North York General Hospital, Toronto, ON; [7]Victoria General Hospital, Victoria, BC; [8]Mount Sinai Hospital, Toronto, ON; [9]Montreal Children's Hospital, Montreal, QC; [10]Sunnybrook Health Sciences Centre, Toronto; [11]The Ottawa Hospital, Ottawa, ON; [12]University of Alberta Hospital, Edmonton, AB

OBJECTIVE: *Clostridium difficile* infection (CDI) is the leading cause of infectious healthcare-associated diarrhea. Global emergence of the epidemic NAP1/027 strain has led to numerous outbreaks in healthcare facilities globally and is associated with increased morbidity and mortality. Standard molecular characterization in North America includes pulse field gel electrophoresis (PFGE) and PCR ribotyping. We hypothesize that whole genome sequencing (WGS) will help to resolve differences within the NAP1 strain that the current molecular methods cannot.

METHODS: WGS was performed on 45 NAP1/027 isolates selected from 23 hospital sites across Canada in 2009 (under CNISP). PFGE and PCR ribotyping were used to initially type the isolates. WGS was conducted using the Illumina MiSeq. Reads from the 45 genomes were mapped against a NAP1/027 genome that was previously sequenced in-house (CD26A54_S) using SMALT. The core single nucleotide variants (SNVs) from non-repetitive and non-horizontally acquired regions were identified and used to create a phylogenomic tree using PhyML.

RESULTS: All NAP1 isolates were indistinguishable by both PFGE (type 001) and PCR ribotyping (027). Core SNVs ranged from 2-74. At the city level, isolates from Edmonton appeared to be more closely related to those from British Columbia than Calgary possibly suggesting interprovincial transmission events. Our results also resolved differences at the hospital level within Ontario.

CONCLUSIONS: The use of WGS provided enhanced discriminatory power compared to current molecular typing methods and could become a valuable tool for tracking the spread of CDI among institutions and within the community.

F4

PROTEOMIC ANALYSIS OF A METRONIDAZOLE-RESISTANT STRAIN OF *CLOSTRIDIUM DIFFICILE*

P CHONG[1], T LYNCH[1], S MCCORRISTER[1], P KIBSEY[2], M MILLER[3], D GRAVEL[4], G WESTMACOTT[1], MR MULVEY[1]

[1]National Microbiology Laboratory - Public Health Agency of Canada, Winnipeg, MB [2]Victoria General Hospital, Victoria, BC [3]Jewish General Hospital, Montreal, QC [4]Centre for Communicable Diseases and Infection Control, Public Health Agency of Canada, Ottawa, ON

OBJECTIVE: Metronidazole (MTZ) is a frontline treatment for *Clostridium difficile* infection (CDI), but recent reports have described isolates of *C. difficile* with reduced susceptibility to MTZ. A shotgun proteomics approach was used to study changes to the proteome of a MTZ-resistant *C. difficile* isolate.

METHODS: CD26A54_R (MTZ-resistant), CD26A54_S (reduced susceptibility), and VLOO13 (MTZ-sensitive) were grown to mid-log phase, and spiked with MTZ at concentrations 2 doubling dilutions below the MIC. The cultures were incubated for an additional 30 minutes prior to cell harvest, followed by protein extraction for trypsin digestion, iTRAQ-labeling, and 2D-LC-MS/MS analysis.

RESULTS: After combining the results of 4 independent 2D-LC-MS/MS experiments, 1942 proteins were identified, with a protein false discovery rate of 0.4%. Differential expression analysis was performed on all strains, with or without MTZ treatment. In the absence of MTZ, higher expression was observed of some proteins in CD26A54_S and/or CD26A54_R that may be involved with reduced susceptibility or resistance to MTZ, including: various DNA repair proteins, such as the Uvr excinuclease; proteins involved in electron transport; and the ferric uptake regulator (Fur), a multifunctional regulator involved in iron homeostasis. Previous genomic analysis indicated a point mutation in the *fur* gene of CD26A54_R, which displays growth deficiencies in the absence of iron in basal medium. The expression of the DNA repair protein RecA was increased more highly in CD26A54_R after MTZ-treatment, compared to CD26A54_S or VLOO13, suggesting that a greater rate of repair of MTZ-damaged DNA may account for the higher level of MTZ resistance observed with CD26A54_R.

CONCLUSION: The proteomic data suggests that a multi-factorial response may be associated with MTZ-resistance in *C. difficile*, including possible roles of altered iron metabolism and/or DNA repair. To the best of our knowledge, this work represents the most comprehensive analysis on MTZ-resistant *C. difficile* to date.

F5

MOLECULAR DETECTION OF *STREPTOCOCCUS PNEUMONIAE* IN NASOPHARYNGEAL SWABS AND DEDUCTION OF "SERO" TYPES BY POLYMERASE CHAIN REACTION (PCR)

A LANG[1,2,3], M EL-SHERIF[3,4], T HATCHETTE[1,2,3,4], S MCNEIL[1,2,3,4], J LEBLANC[1,2,3,4]

[1]Dalhousie University; [2]Canadian Center for Vaccinology (CCfV), IWK Health Centre; [3]PCIRN SOS Network Investigators; [4]Capital District Health Authority (CDHA), Halifax, NS

OBJECTIVE: For *S. pneumonia*, molecular approaches offer many advantages over traditional serological typing methods (e.g. Quellung reaction). The goals of this study were to evaluate typing using a multiplex PCR (MP-PCR) developed by the Centers for Disease Control and Prevention (CDC) and optimize a real-time PCR for the detection of *S. pneumoniae* from nasopharyngeal (NP) swabs.

METHODS: Performance of the CDC multiplex PCR was verified using a panel of previously characterized isolates (n=45). To ensure specificity, DNA from each isolate was tested against each of the 8 PCR multiplex reactions. This DNA was also subjected to 2 real-time PCR assays targeting *lytA* and *cpsA*. The limit of detection (LoD) for NP swabs was determined using 10-fold dilutions of quantified organism and the clinical utility was assessed using a panel of clinical specimens (n=374) with or without invasive pneumococcal disease (IPD). Threshold cycle (Ct) values were calculated by the real-time PCR instrument software.

RESULTS: The MP-PCR was specific and the LoD was ~17,771 CFU/ml. This value was ~100-fold less sensitive than *lytA* and *cpsA* real-time PCR, which displayed values of ~172 CFU/ml (Ct ~38) and 823 CFU/ml (Ct ~37), respectively. Real-time *lytA* PCR detected *S. pneumoniae* in 46/374 clinical specimens (NP swabs); however, only 15 were typeable by MP PCR. LytA-positive specimens with Ct values >34 fell below the LoD of the MP-PCR and therefore could be typed.

CONCLUSIONS: Screening of NP swabs for *S. pneumoniae* using real-time PCR and subsequent typing using MP-PCR provides a valuable framework for epidemiologic studies. Serotype distribution could thus be monitored pre- and post-pneumococcal vaccine programs without the use of traditional culture.

F6

COMPARISON OF 3 AMPLIFICATION ASSAYS FOR THE DETECTION OF SHIGA TOXIN-PRODUCING *E COLI*

L CHUI[1,2], R ALLEN[2]

[1]Provincial Laboratory for Public Health; [2]University of Alberta, Department of Laboratory Medicine and Pathology, Edmonton, AB

OBJECTIVE: Shiga toxin-producing *E. coli* (STEC) is a worldwide problem because it has great impact on human health and meat/processing industries. The objective of this study is to compare conventional PCR, real-time (RT) PCR and loop-mediated isothermal amplification (LAMP) assays for detecting STEC.

METHODS: DNA was extracted from overnight cultures of bacteria or stool samples and used as template for the different amplification assays. Specificity, sensitivity, and limit of detection (LOD) were determined. Cost consumables/reagents and turn around time (TAT) from each of the tests were also evaluated. A blind panel of positive and negative clinical samples were included in the study.

RESULTS: The LAMP assay has a LOD of 1 CFU/reaction for both stx_1 and stx_2; a TAT of approximately 1.5h when SYBR Green I is added post amplification. However with the Genie II instrument, the assay can be completed within 30 min. Conventional PCR has a similar LOD with a TAT of 4h. The LOD of RT- PCR is at 1 CFU for stx_1 and 10 CFU for stx_2 and the TAT is 65 min. All three assays show 100% specificity and sensitivity and similar cost of less than $5.00 per test.

CONCLUSIONS: All three assays are comparable in terms of performance and cost. LAMP assay is easy to perform and requires only a heat block but artifacts were observed when agarose gel electrophoresis was used as the detection method. Addition of SYBR Green I to the amplicons shortened the TAT but caused false positivity. Performing LAMP on the Genie II shortened the TAT and also allows the confirmation of the amplification by melting curve analysis. Based on the budget and set up of the laboratories, a choice could be made by using the different amplification assays to accomplish the goal of detecting STEC.

F7

EVALUATION OF TWO COMMERCIAL KITS FOR DETECTING SHIGA TOXIN-PRODUCING *E COLI* (STEC)

L CHUI[1,2], J CHIAO-LING KUO[2]

[1]Provincial Laboratory for Public Health; [2]University of Alberta, Department of Laboratory Medicine and Pathology, Edmonton, AB

OBJECTIVE: STEC are emerging pathogens responsible for diarrheal disease causing major outbreaks worldwide. Most microbiology laboratories use differential media which fail to detect non-O157 STEC. Our study evaluated the *SHIGA TOXIN CHEK* and the *SHIGA TOXIN QUIK CHEK* and the objectives were: to determine the prevalence of STEC in the Lethbridge region; and to compare the performance of two immunoassays to an in-house real time-PCR (RT-PCR) for stx_1 and stx_2.

METHODS: *SHIGA TOXIN CHEK* is an enzyme-linked immunoassay and *SHIGA TOXIN QUIK CHEK* is a membrane enzyme immunoassay which can differentiate the 2 toxins. 784 stool samples were submitted in the summer of 2012. An aliquot of each sample was tested directly, or after MAC broth enrichment, by both assays. Panels of bacteria were included to determine the sensitivity and specificity.

RESULTS: Of the 784 diarrheal samples, 20 were STEC positive, including 6 O157:H7 STEC and 14 non-O157 STEC. The sensitivity of both immunological assays for clinical samples is between 65-80% and is 95% for RT-PCR. The specificity of all 3 assays are >98%. There was no cross reactivity with the panel of bacteria cells when tested by all 3 assays. The limit of detection was at 7×10^5 CFU/reaction with the immunological assays and 1 bacterial cells/reaction for stx_1 and 10 bacterial cells/reaction for stx_2 for RT-PCR.

CONCLUSIONS: The prevalence of STEC is 2.6% in which 70% were non-O157 and not reported. Although both immunoassays can be used to detect Shiga toxins from direct fecal samples and broth cultures, false negative results could occur with low bacterial load. *SHIGA TOXIN QUIK CHEK* might be an alternative for frontline microbiology laboratories if amplification test is unavailable.

F8

VALIDATION OF A REAL-TIME PCR FOR *TREPONEMA PALLIDUM* DETECTION

S MAN[1], M LEE[1], K FERNANDO[1], M MORSHED[1,2]

[1]BCCDC Public Health Microbiology and Reference Laboratory, Provincial Health Services Authority; [2]University of British Columbia, Vancouver, BC

OBJECTIVE: To reduce the turnaround time and increase the sensitivity of syphilis detection, we evaluated and validated a real-time PCR method for the detection of *Treponema pallidum*. To assess sample quality and adequacy, we multiplexed this assay with a human β-globin gene detection assay.

METHODS: DNA from samples was extracted using Qiagen DNA Extraction Kit and real-time PCR was performed on the ABI Taqman 7500. The screening test, which targeted tp47 kDa membrane protein of *Treponema pallidum*, was multiplexed with a reaction that targeted the human β-globin gene. Positive samples were then subjected to a confirmatory test which targeted the *T. pallidum* polA gene. Analytical validation was based on a comparison between this method and an in-house established traditional PCR that targeted polA gene (n=50). The clinical validation was evaluated with routine samples (n=20).

RESULTS: The sensitivity and specificity of the real-time PCR assay was greater than that of the conventional PCR assay. Monitoring the human β-globin DNA ensured that sampling and storage were adequate and the DNA amount was sufficient for testing. The elimination of gel electrophoresis by using real-time PCR significantly shortened the turn-around-time and uncertainty in results when gel bands were weak.

CONCLUSION: This validation study showed the Taqman assay was rapid and reproducible. The implementation of an assay to detect for sample quality and adequacy helped confirm the validity of a negative result.

FRIDAY APRIL 5
1600 – 1800
Room: 205B

G1
MEASUREMENT OF PATIENT HAND HYGIENE IN AN ACUTE CARE HOSPITAL

JA SRIGLEY[1,2], C FURNESS[2,3], M GARDAM[1,2]
[1]University Health Network; [2]University of Toronto; [3]Infonaut Inc, Toronto, ON

OBJECTIVES: Healthcare worker hand hygiene (HH) is one of the most important ways to prevent healthcare-associated infections (HAIs), but there has been comparatively little emphasis on patient HH despite the fact that nosocomial pathogens may be acquired by patients via their unclean hands. There are few data on how often hospital patients wash their hands or when HH is indicated for patients. There are four moments when patient HH may be indicated in order to reduce the risk of HAIs: before eating, after using the bathroom, and on entering and leaving their room.

METHODS: A real-time locating system was installed on a multi-organ transplant unit. Continuous real-time HH data was generated via ultrasound technology in the hospital environment and worn by patients and staff. Patient use of alcohol-based hand rub and soap dispensers was recorded during visits to bathrooms and pantries.

RESULTS: During a 30-day period, there were 176 patient room stays. These patients made 5561 visits to the bathroom, of which 1927 (34.7%) were associated with use of the soap dispenser in the bathroom. Soap use ranged from 0% of visits in 44 patients (25.0%) to 100% in 10 (5.7%). In a 55-day period, patients visited two pantries 666 times. Of those pantry visits, 42 (6.3%) were associated with use of alcohol-based hand rub before entering or soap while inside.

CONCLUSIONS: Patients perform HH infrequently after bathroom visits and when visiting the pantry. This may contribute to transmission of pathogens from the hospital environment via the fecal-oral route. Future research will assess patient HH on entering and leaving their rooms, as well as whether HH at any of the four moments are associated with increased risk of HAIs.

G2
DISINFECTANTS INTERFERE WITH ATP METHODS USED FOR RAPID MONITORING OF ENVIRONMENTAL CLEANING COMPLIANCE

M ALFA[1,2,3], N OLSON[3], P DEGAGNE[1]
[1]Diagnostic Services of Manitoba; [2]University of Manitoba; [3]St Boniface Research Centre, Winnipeg, MB

OBJECTIVE: To ensure that housekeeping staff are compliant with cleaning of high-touch surfaces in the healthcare environment, rapid monitoring tools have been recommended. Rapid test kits that detect ATP from bioburden as well as organic residuals are commercially available for cleaning monitoring. The objective of this study was to determine the effect of disinfectants and cleaners on detection of ATP.

METHODS: The 3M™ Clean-Trace ATP Surface test was used to detect ATP (as measured by relative light units (RLUs). Sterile RO water, Virox Accel Intervention™ RTU (INT), 5000ppm Bleach (BL) and PerDiem (PD) were the agents tested. ATS-T (organic test soil) alone or containing ~10^8 cfu/mL *E. faecalis*, *P. aeruginosa* and *C. albicans* (ATS-Tbugs) and pure ATP were inoculated on toilet seats (0.1 mL/2cm^2) and dried overnight. Inoculated and uninoculated areas on the toilet seats were spritzed with the test agents (all tests done in triplicate). After 1 and 3 minutes contact the rapid ATP surface test was used to evaluate the inoculated surfaces.

RESULTS: The average RLUs from the ATS-T, ATS-Tbugs, and ATP alone were 31,633, 351,824, and 528,837, respectively. Water and PD had no impact on ATP in ATS-T, ATS-Tbugs but INT and BL resulted in 70% to 99% reduction in RLUs after 1 min in ATS-T, ATS-Tbugs and ATP alone. Bleach on the uninoculated surface produced up to 600 RLUs compared to all other agents that had background levels of RLU (<60 RLUs).

CONCLUSIONS: Bleach was the only test agent that produced false RLUs. INT and BL both significantly reduced the ATP levels after only 1 minute contact time with ATS-T and ATS-Tbugs but PD did not.

G3
VANCOMYCIN-RESISTANT ENTEROCOCCUS (VRE) TRANSMISSION AND RISK FACTORS FOR VRE ACQUISITION

R KAKI[1], Y YU[1], C O'NEILL[2], D MERTZ[1,2]
[1]McMaster University; [2]Hamilton Health Sciences, Hamilton, ON

OBJECTIVES: We analysed transmission rates and potential risk factors for VRE acquisition for patients sharing the room with a newly diagnosed VRE carrier.

METHODS: Retrospective chart review at two adult acute care sites of Hamilton Health Sciences, Hamilton, ON, from 2010-2011. Contacts sharing a room with a newly detected index patient were isolated and screened. In addition, point prevalence screening was performed. An outbreak was defined when 3 or more new VRE cases in non-roommates occurred within 7 or 5 cases within 30 days.

RESULTS: A total of 254 index patients were detected including 15 patients that were contacts of previously detected index patients. Of these, 163 (64.2%) were sharing a room prior to detection with one of 368 contact patients. Forty (10.9%) contacts were VRE positive, with the first screening being positive in 17 (42.5%) of cases. Transmission rates differed between hospitals (Table). Exposure to vancomycin (OR 4.24; 95% CI 2.16-8.34), fluoroquinolones (OR 2.33; 95% CI 1.19-4.55), and antibiotics with anti-anaerobic activity (OR 2.78; 95% CI 1.42-4.43) were associated with VRE acquisition. There was no association with diarrhea in the index patient or age and co-morbidities in contacts. In multivariate analysis, exposure to vancomycin (OR 2.98; 95% CI 1.45-6.14) and anti-anerobic antibiotics (OR 2.34; 95% CI 1.14-4.81) as well as being a patient in hospital #2 as compared to #1 (OR 2.26; 95% CI 1.03-4.96) were found to be independent risk factors.

CONCLUSIONS: Exposure to vancomycin and antibiotics with anaerobic activity were independent risk factors for VRE acquisition. Transmission rates were not significantly higher during VRE outbreaks. Sensitivity of the first screening was less than 50% corroborating the need for multiple screening samples.

G4
DETECTION OF CARBAPENEMASE-PRODUCING *ENTEROBACTERIACAE* (CRE) FROM RECTAL SWABS: WHICH METHOD IS MOST SPECIFIC?

BM WILLEY[1], A MAZZULLI[1], A TSANG[2], T FUNG[1], M LAROCQUE[1], DA BOYD[3], L MATASEJE[3], C HUM[1], M NG[1], T PIPKIN[1], P LO[1], MR MULVEY[3], AJ MCGEER[1,2], DE LOW[1,2], SM POUTANEN[1,2]
[1]Mount Sinai Hospital/University Health Network; [2]University of Toronto, Toronto, ON; [3]National Microbiology Laboratory, Winnipeg, MB

OBJECTIVES: To determine which CRE detection method is most specific (Sp), 2011 CRE detection study results were compared to new 2012 evaluations of meropenem (M) based methods using the same predominantly multi-drug resistant (R) strain collection. Methods included: a) selection on Oxoid ESBL agar (ESBL) then to carbapenem (carb) disc test (DD), b) ESBL then to carb spot agar, or c) selection on a carb agar. Methods with <100% CRE recovery were excluded from analyses.

METHODS: 254 distinct *Enterobacteriaceae* (ENT) comprising 109 PCR-confirmed CRE (89 KPC, 15 NDM, 3 OXA48, 2 SME) and 145 non-CRE (including *ompC/F*, *ompK35/36*, derepressed-*ampC* mutants, etc, with carb reduced-susceptibility) were grown from −80°C to MacConkey (Mc) and ESBL with ertapenem (Etp) discs for selective pressure. 10 uL

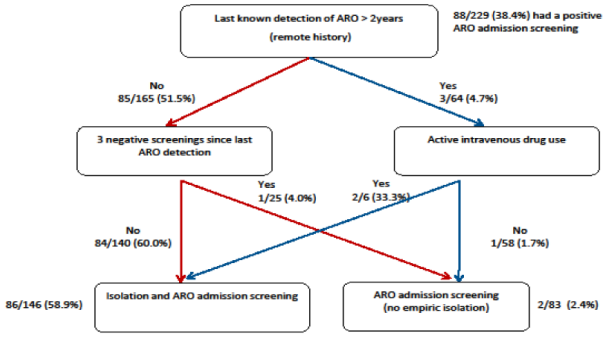
from 0.5 McFarland suspensions were streaked to Oxoid Mc with M (Mc-M) at 0.125, 0.25, and 0.5 mg/L and spotted to Oxoid Mueller-Hinton agar with M (MH-H) at 0.125, 0.25 and 0.5 mg/L. Screen/spot agars were read after 18h at 37/35°C, respectively. These data were compared to 2011 results.
RESULTS: Methods with 100% CRE detection (95% CI: 95.9-100) were: Mc-M0.125 and Mc-M0.25 mg/L screens and ESBL to MH-M0.25 mg/L spot in 2012; ESBL to MH-Etp1 mg/L spot, ESBL to CLSI Etp-DD or ESBL to M-DD using a modified <25mm breakpoint in 2011. Methods growing the lowest % non-CRE (95% CI) were: ESBL to M-DD [15 (11-22)], ESBL to MH-Etp1mg/L spot [29 (22-37)], ESBL to Etp-DD [33 (26-41], Mc-M0.125mg/L screen [30 (23-38)], ESBL to MH-M0.25mg/L spot [34 (27-42)], and ESBL to MH-M0.125mg/L spot [38 (31-47)].
CONCLUSIONS: Of all methods with 100% CRE detection, ESBL agar followed by M-DD testing using a modified <25 mm breakpoint was the most specific.

G5
ALGORITHM FOR INFECTION CONTROL MANAGEMENT OF PATIENTS WITH AN ANTIBIOTIC RESISTANT ORGANISM (ARO) HISTORY

C O'NEILL, K NURI, C BERTRAND, E DESOUZA, P PERRY, P PELTSCH, G FISHER, J CRAIG, C GITTENS WEBBER, S DALGLEISH, J FULLERTON, D MERTZ
Hamilton Health Sciences, Hamilton, ON

OBJECTIVES: Hospitalized patients with a history of an antibiotic resistant organism (ARO) are empirically isolated until admission ARO screening results are available. Hospitals are struggling with the growing demands for isolating these patients. We aimed to identify risk factors for persisting ARO colonization and to create a simple algorithm to determine appropriate isolation requirements while screening results are pending.
METHODS: An algorithm tool was created based on 245 consecutive patients admitted with an ARO history to the three tertiary acute care teaching hospitals of Hamilton Health Sciences, ON, Canada, from February 3 to May 2 2012. The algorithm was validated on a second population of 273 consecutive patients admitted from May 3 to July 19.
RESULTS: ARO admission screening results were available for 229/245 (93.5%) admissions in 207 patients in the test population. The majority of patients had a previous history of methicillin resistant Staphylococcus aureus (MRSA; 144/229, 62.9%) or vancomycin resistant enterococci (VRE; 71/229, 31.0%). After admission, 88/229 (38.4%) had a positive screening result. A history of < 2 years since last ARO detection was the most significant risk factor (OR 21.6, 95% CI 6.5-71.6). The final algorithm (Figure 1) reduced the number of unnecessary isolations from 141 to 60 (specificity of 57.4%). Only 2 patients with positive admission screening results were not isolated (sensitivity 97.7%). The algorithm was similarly accurate for MRSA, VRE and other ARO. A total of 249 patients with a history of an ARO were available in the validation population with comparable findings (sensitivity 93.6%, specificity 56.8%, overall accuracy 70.7%).

Figure 1: Algorithm- Results for Test Population



CONCLUSIONS: The algorithm proved to be a simple, effective and highly sensitive decision making tool by decreasing the number of unnecessary isolations by almost 60%, and can therefore improve patient flow and utilization of single rooms.

G6
TRENDS IN ANTIVIRAL TREATMENT FOLLOWING THE 2009 INFLUENZA PANDEMIC IN CANADA

R MITCHELL[1], G TAYLOR[2], A MCGEER[3], C FRENETTE[4], K SUH[5], A WONG[6], K KATZ[7], K WILKINSON[1], B AMIHOD[8], D GRAVEL[1]
[1]Public Health Agency of Canada, Ottawa, ON; [2]University of Alberta Hospital, Edmonton, AB; [3]Mount Sinai Hospital, Toronto, ON; [4]McGill University Health Centre, Montreal, QC; [5]The Ottawa Hospital, Ottawa, ON; [6]Royal University Hospital, Saskatoon, SK; [7]North York General Hospital, Toronto, ON; [8]SMBD-Jewish General Hospital, Montreal, QC

BACKGROUND: Antiviral treatment is associated with reduced mortality, length of stay and improved clinical outcomes among patients hospitalized with influenza.
METHODS: Using Canadian Nosocomial Infection Surveillance Program data, antiviral treatment among adults hospitalized with laboratory-confirmed influenza during the 2009-2010 influenza pandemic season was compared to those in the post-pandemic seasons (2010-2011 and 2011-2012).
RESULTS: A higher proportion of adults hospitalized with laboratory-confirmed influenza received antiviral treatment during the 2009 influenza pandemic season (997/1,113; 89.6%) compared with the 2010-2011 (873/1,092; 80.0%, p<0.001) and 2011-12 (391/600; 65.2%, p<0.001) influenza seasons. The decrease in antiviral use between the 2009 pandemic season and the 2010-11 season was statistically significant in all age groups except for inpatients ≥65 years. A decrease in antiviral use among inpatients ≥65 years was not observed until the 2011-2012 season. A median of 3 days between symptom onset and antiviral treatment was reported for all three influenza seasons. Among high-risk groups, adults with underlying medical conditions were significantly more likely to receive antiviral treatment during the pandemic than during the post-pandemic seasons (89.7% vs. 75.7%, p<0.001). A higher proportion of adults admitted to the ICU during the 2009 pandemic (94.2%) received antiviral treatment compared with the 2010-11 and 2011-12 seasons (84.6%, p<0.001). There was no difference in antiviral treatment among inpatients who died within 30 days of admission during the pandemic (84.3%) than during the post-pandemic seasons (78.9%, p=0.370).
CONCLUSION: Antiviral treatment of adults hospitalized with laboratory-confirmed influenza significantly fell in the two seasons following the 2009 influenza pandemic. In order to guide strategies aimed at minimizing the impact of influenza among hospitalized adults, reasons for the decline in antiviral treatment need to be further explored.

G7
COMPLICATED *CLOSTRIDIUM DIFFICILE* INFECTION IS UNCOMMON IN CHILDREN

K SCHWARTZ[1,2], I DARWISH[2,3], N THAMPI[1,4,5], MR MULVEY[5], S RICHARDSON[1,2]
[1]Hospital for Sick Children; [2]University of Toronto; [3]University Health Network; [4]Mount Sinai Hospital, Toronto, ON; [5]National Microbiology Laboratory, Winnipeg, MB

OBJECTIVES: *Clostridium difficile* infection (CDI) is the most common cause of health care–associated diarrhea in children. Increasing rates of severe CDI have been reported in adults and severe CDI has been reported in individual cases in children; however, a widespread increase in disease severity has not been reported from pediatric cohorts. The purpose of this study was to examine the features of CDI in a pediatric population, with special attention to the occurrence of CDI-related severe outcomes.
METHODS: A retrospective chart review was conducted for patients positive for *C. difficile* by cytotoxin assay between August 1, 2008 and July 31, 2012. Basic demographics, nosocomial versus community acquisition, clinical severity, treatment, and outcome data were assessed. Pulsed field gel electrophoresis typing and PCR detection of toxin A (*tcdA*), toxin B (*tcdB*), binary toxin and *tcdC* genes were performed on isolates from nosocomial cases by the National Microbiology Laboratory.
RESULTS: Of the 308 children with CDI, 90% experienced resolution of symptoms by 30 days after disease onset. Five children required transfer to the ICU and seven children received surgical consultation for severe CDI symptoms. During the study period, no colectomies were performed and one death occurred where CDI was felt to contribute. Various combinations of clinical and

laboratory features were not predictive of severe outcomes. Hospital-acquired infections comprised 74% of cases. The NAP-4 strain was most frequent among the typeable strains (41%), followed by NAP-1 (19%). There was no association between NAP-1 strains, or *tcdC* deletion, and severe disease.
CONCLUSIONS: Complicated CDI in children is uncommon compared to adults. Further prospective pediatric studies on CDI in both the community and hospital settings are required to better understand risk factors and optimal treatment.

G8

CHARACTERIZATION OF *LEGIONELLA PNEUMOPHILA* SEROGROUP 1 STRAINS INVOLVED IN THE 2012 OUTBREAK IN QUEBEC CITY: CONVENTIONAL MOLECULAR METHODS AND WHOLE GENOME SEQUENCING

S LÉVESQUE[1], H CHAREST[1], P PLANTE[2], P CANTIN[3], G MARCHAND[4], C HUOT[5], J VILLENEUVE[5], R DION[1], I GOUPIL-SORMANY[5], F DESBIENS[5], J CORBEIL[2], C TREMBLAY[1]
[1]LSPQ/INSPQ, Sainte-Anne-de-Bellevue; [2]Université Laval; [3]CEAEQ/MDDEFP, Québec; [4]IRSST, Montréal; [5]DRSP de la Capitale-Nationale, Québec, QC

BACKGROUND: In 2012, a major *L. pneumophila* serogroup 1 (LP1) outbreak occurred in Quebec city, causing 181 reported cases including 13 fatalities. LP is a ubiquitous Gram-negative bacteria within freshwater and man-made aquatic environments such as cooling tower systems. Infection in humans occurs through inhalation of droplets carrying the bacteria. Strains genotyping is key to identify the source of an outbreak. Pulsed-field gel electrophoresis (PFGE) is the reference method for laboratory investigations of outbreaks.
OBJECTIVE: To evaluate whether other typing methods could add to the molecular investigation of this outbreak, we compared profiles obtained with PFGE to Sequence-based typing (SBT) and to whole genome sequencing (WGS).
METHODS: LP1 isolates recovered from patients were analyzed using PFGE and SBT. Water samples from cooling towers located near outbreak cases were cultured according to AFNOR guidelines isolates were typed by both methods. WGS was performed using the Nextera XT kit for library preparation from all patient isolates, a subset of environmental isolates and isolates from a former outbreak in the same area in 1996. Sequence assembly was performed using the Ray assembler.
RESULTS: 23 LP1 isolates obtained from human cases were characterized. Among these, 22 displayed a unique PFGE pattern. 131 cooling tower samples from 70 buildings were cultured. 146 LP1 isolates were obtained from 33 samples and 6 different PFGE patterns were identified. Only one cooling tower harbored strains with the same PFGE pattern as patients and was identified as the source of the outbreak. SBT was performed on 53 isolates (23 human and 30 environment isolates) with a minimum of one strain typed for each PFGE pattern. This method was less discriminatory than PFGE since more than one PFGE pattern harbored the same SBT type. Comparison of SBT types showed that all strains had already been described in North America and Europe. Phylogenetic analysis of sequences obtained with WGS showed the clustering of patient strains with the matching strain from the cooling tower, in agreement with PFGE and SBT results. It also revealed that this cluster was unrelated to strains from the 1996 outbreak.
CONCLUSION: PFGE has proven utility in outbreak investigations. However, SBT is useful for strain comparison to international databases. WGS confirmed the findings of the two typing methods. The use of WGS could add additional information about virulence factors.

FRIDAY APRIL 5
1600 – 1800
Room: 205C

H1

A POINT PREVALENCE STUDY TO EVALUATE CLINICAL COMPLIANCE WITH A VENTILATOR ASSOCIATED PNEUMONIA ALGORITHM IN THE INTENSIVE CARE UNIT

Q MOHIUDDIN[1], S NELSON[1], M STEINBERG[1], T JIVRAJ[1], C HARRIS[1], L BURRY[1,2], L VARGA[1], S LAPINSKY[1,2], C BELL[1,2], A MORRIS[1,2,3]
[1]Mount Sinai Hospital; [2]University of Toronto; [3]University Health Network, Toronto, ON,

BACKGROUND: Ventilator associated pneumonia (VAP) is a common occurrence in mechanically ventilated patients, and is difficult to diagnose accurately and reliably. In an effort to standardize the approach to diagnosis and management of VAP, a working group was formed at Mount Sinai Hospital (MSH) and the University Health Network (UHN) to develop a VAP algorithm. This algorithm was piloted in the intensive care unit (ICU) at MSH in November of 2011.
METHODS: We conducted a point prevalence study to evaluate clinical compliance with the VAP algorithm in the MSH ICU. A retrospective chart review was carried out and data was collected from all patients who had sputum and/or bronchoalveolar lavage (BAL) samples sent to the microbiology lab for suspicion of VAP during a pre-determined 6-week period pre-implementation of the VAP algorithm, and again post-implementation.
RESULTS: In the pre-algorithm patient group (n=23) there were 41 sputum samples and 11 BAL samples sent to the microbiology lab, of which 9 samples were culture positive. In the post-algorithm patient group (n=12) there were 19 sputum samples and 5 BALs, of which 5 samples were culture positive. Of the 41 sputum samples obtained from the pre-algorithm group, 33 were done in patients who had calculated CPIS scores of ≤6 and 8 in patients who had CPIS scores >6; of the 11 BAL samples, 9 were done in patients who had CPIS scores ≤6 and two were performed in patients who had CPIS scores >6.
CONCLUSIONS: Our data demonstrates excellent uptake of the VAP algorithm by healthcare providers at MSH. In addition to demonstrating a decrease in the number of sputum and BAL samples sent to the lab, the duration of empiric and tailored antimicrobial therapy was decreased. Patient outcomes after the algorithm's implementation were comparatively better than prior to the algorithm's implementation. Further analysis following implementation of the VAP algorithm at additional sites will enhance the validity and reliability of the data.

H2

HOSPITAL WIDE ROLL-OUT OF ANTIMICROBIAL STEWARDSHIP: A STEPPED WEDGE RANDOMIZED CONTROLLED TRIAL

L PALMAY, M ELLIGSEN, S WALKER, A RACHLIS, S MUBAREKA, V ALLEN, R PINTO, N DANEMAN
Sunnybrook Health Sciences Centre, Toronto, ON

Inappropriate antimicrobial use, increasing antibiotic resistance, and lack of development of new antimicrobial agents have provided the impetus for worldwide initiatives in antimicrobial stewardship. However, there is paucity of good quality evidence to evaluate the impact of such initiatives. As a result of the positive impact our institution's Antimicrobial Stewardship Program (ASP) had in our critical care units, we rolled out our program to seven patient care services using a stepped-wedge design to provide a more rigorous evaluation of our intervention. The objective of this study was to determine the impact of antimicrobial stewardship on antimicrobial use , *C. difficile* infection rates, bacterial resistance rates, mortality and length of stay in seven non-intensive care medical and surgical services. A formal review of all patients on their 3rd or 10th day of broad-spectrum antibiotic therapy was conducted at our institution using a stepped–wedge randomized design over a 24 month period. The primary outcome of the preliminary analysis was a comparison of days of therapy (DOTs) of broad-spectrum

antibiotics/1000 patient days during the intervention period compared to the control period. Secondary outcomes included nosocomial *C.difficile* infection rates, bacterial resistance, length of stay and mortality. Over the two-year period, the ASP reviewed a total of 2,733 orders with an overall suggestion and acceptance rate of 47% and 80%, respectively. Overall, our program reduced broad-spectrum antimicrobial use by 10% ,with no significant changes in *C. difficile* rates, antimicrobial resistance, length of stay or mortality. Our institution's ASP hospital-wide roll-out was, therefore, successful in reducing broad-spectrum antimicrobial use without negatively compromising patient safety.

H3

A SURVEY TO EVALUATE CRITICAL CARE TRAINEES' PERCEPTIONS OF ANTIMICROBIAL STEWARDSHIP PROGRAMS IN INTENSIVE CARE UNITS

L DRESSER[1,2], M STEINBERG[1], M SO[1], C BELL[1,2], D SCALES[2,3], A MORRIS[1,2,4]

[1]University Health Network; [2]University of Toronto; [3]Sunnybrook Health Sciences Centre; [4]Mount Sinai Hospital; Toronto, ON

BACKGROUND: Antimicrobial stewardship is a multi-disciplinary programmatic initiative aimed at optimizing antimicrobial therapy. Accreditation Canada has mandated that Antimicrobial Stewardship Programs (ASPs) will be a Required Organizational Practice for all Canadian acute care hospitals in the next accreditation cycle. Critical Care Trainees (CCTs), while rotating through different hospitals, have a varied experience in working in ICUs where there may or may not be ASPs in place. The purpose of the survey was to determine CCTs' perceptions and attitudes towards Antimicrobial Stewardship Programs in Intensive Care Units.

METHODS: We distributed an on-line, anonymous survey to all CCTs who have rotated through GTA ICUs between July 2010 and June 2012. Survey items assessed ASP knowledge and experiences. Attitudes towards ASPs were assessed on a 5-point Likert scale.

RESULTS: Response rate to the survey was 32% (n=18/57). Only 44% of the respondents were familiar with the concept of an ASP prior to their training as a Critical Care Fellow. At some point during their Critical Care Fellowship, 22% (4/18) of respondents rotated through an ICU that did not have an ASP in place, and 94% (17/18) rotated through at least one ICU that had an ASP in place during their fellowship.

The majority of respondents (89%) felt that the ASP increased their knowledge of appropriate antimicrobial use in the ICU. Most (83%) felt that time spent with the ASP team was an efficient use of their time. Only 17% reported that they felt the ASP affected their autonomy in a negative way. Importantly, a large majority (89%) of respondents felt that the patients in the ICU benefitted from having an ASP in place.

CONCLUSION: In these academic centres in the GTA, CCTs are strongly supportive of Antimicrobial Stewardship in the ICU and feel that ASPs provide a valuable service to both patients and clinicians.

H4

IMPACT OF AN ANTIMICROBIAL STEWARDSHIP PROSPECTIVE AUDIT AND FEEDBACK PROGRAM ON CANDIDEMIA IN THE INTENSIVE CARE UNIT

L DRESSER[1,2], T JIVRAJ[3], M STEINBERG[3], K DUPLISEA[1], S NELSON[3], S POUTANEN[2,3], J SINGH[1,2], N LAZAR[1,2], S LAPINSKY[2,3], A MORRIS[1,2,3]

[1]University Health Network; [2]University of Toronto; [3]Mount Sinai Hospital, Toronto, ON

BACKGROUND: An Antimicrobial Stewardship Program (ASP) that utilizes Prospective Audit and Feedback (PAF) 3-5 days/week has been operational in 3 MSICUs of 2 large tertiary academic centers for over three years. The purpose of this study was to evaluate the rates of candidemia pre and post-ASP implementation.

METHODS: The microbiology Laboratory Information System (LIS) was queried to identify total number of blood cultures processed and the number positive for yeast over defined study periods pre- ASP implementation, and post. ASP with PAF was introduced sequentially from ICU 1 to 3. Antibacterial and antifungal consumption was reported as Defined Daily Doses (DDD's) or costs in dollars. Statistical analysis using t-test for continuous and Chi-square with Yates' correction for non-parametric data were used.

RESULTS: In ICU's 1 and 3 the rate of blood cultures positive for yeast decreased post-ASP compared to the pre-study period; in ICU 2 the rate remained the same. Similarly the unique episodes of candidemia decreased in ICU's 1 and 3 but remained static in ICU 2. Systemic antibacterial consumption was statistically significantly lower in each ICU comparing up to 2 fiscal years before and after ASP-PAF introduction. The total number of patients treated in any of the ICUs did not change significantly over the study period.

CONCLUSIONS: Since the implementation of ASP with PAF in these 3 ICUs, there has been a decrease in antibacterial consumption in each unit. Candidemias in the 2 ICU's with the highest pre-ASP incidence have decreased. Factors contributing to the change in candidemias at ICU 1 and 3 may be explained in part by impact of ASP activities in addition to ongoing infection prevention and control initiatives. The lack of change in candidemias in ICU 2 may reflect the baseline low rates of fungemia and less broad spectrum antimicrobial consumption compared to ICU 1 and 3, reflective of the different patient mix.

H5

CRITICAL REVIEW OF ANTIMICROBIAL RESISTANCE SURVEILLANCE IN CANADIAN HUMAN AND VETERINARY MEDICINE: PRELIMINARY RESULTS AND IDENTIFICATION OF UNMET NEEDS

J GRANT[1], L SAXINGER[2], P KEEN[1], D KAO[1], D PATRICK[1]

[1]University of British Columbia, Vancouver, BC; [2]University of Alberta, Edmonton, AB

BACKGROUND: Protection of human, animal, and ecosystem health depends on judicious, optimized use of available antibiotics. Surveillance of antimicrobial resistance in key pathogens of importance in human and veterinary medicine is required to understand and control resistance emergence. As part of a larger project, we examined current Canadian surveillance programs for antimicrobial resistance in humans and in animals.

METHODS: Information on antimicrobial resistance surveillance programs in Canada and world-wide was collected by a formal literature search. Seventeen databases were reviewed, and 7639 papers were reviewed for inclusion, with data and/or directly relevant information extracted from 128 papers.

RESULTS: 17 surveillance programs met criteria for evaluation. Three of these were defunct or not yet reporting, 7 were provincial or sub provincial in scope, leaving 4 that were national with a narrow AMR focus (enteric pathogens, human salmonellosis, gonorrhea, and group A streptococci), and 3 that are national and broader in scope. These three and the federal antimicrobial utilization-resistance surveillance in the agrifood sector are presented in the table.

Major Canadian Antimicrobial Resistance Monitoring Programs: All Funding Sources, National Scope

	CNISP	CanWARD	Canadian Bacterial Surveillance Network	CIPARS
Description	Active surveillance of nosocomial infections and antimicrobial resistant organisms (AROs), sentinel sites	Voluntary prospective sampling, 10-15 hospitals - 500 bacterial clinical isolates yearly (blood, resp, urine, wound)	Voluntary prospective sampling from hospital laboratories (10 provinces represented, last listed 2008)	Surveillance of isolates from abbatoirs, retail meat, and human salmonellosis, across Canada
Human AMR relevant pathogens surveilled (year of last available report)	MRSA (2009) VRE (2005) C. difficile (2007) Others on project basis	Broad pathogen representation, 500 isolates per site, reflects approx 5-10% of antibiogram isolates yearly (2011)	*S. pneumoniae* (2009) *Group A Strep, H.influenzae, E. coli, Klebsiella spp., S.aureus, VGS, M. catharalis* (2005)	Human *Salmonella* isolates from provincial public health and reference laboratories (2010, limited, 2008 full report)

Domain	Nosocomial infections	Hospital specimens only (including ED)	Hospital specimens only (including ED)	Vet- Agrifood and limited human
Funding/Support	Federal	Pharma-Academic	Pharma-Academic	Federal

CONCLUSION: AMR is a recognized public health crisis. The majority of extant Canadian AMR surveillance programs are focal with respect to pathogen focus and/or geographic reach, and with only one national scope, non pharmaceutical funded program which is able to address emerging resistance, focused on hospital- nosocomial AMR. There are no national, federally supported community AMR surveillance programs in Canada.

H6

INADEQUATE ANTIBIOTIC THERAPY IS ASSOCIATED WITH DECREASED SURVIVAL IN PATIENTS WITH EXTENDED-SPECTRUM BETA-LACTAMASE (ESBL)-PRODUCING *ENTEROBACTERIACEAE* (ENT) BLOODSTREAM INFECTIONS (BSIS)

N DEWHURST[1,2], SM POUTANEN[2,3,4], B HAMANDI[2,4], L DRESSER[2,4], M PITRE[4], A LIN[4], D LOWE[4]

[1]St. Michael's Hospital; [2]University of Toronto; [3]Mount Sinai Hospital; [4]University Health Network, Toronto, ON

OBJECTIVE: ESBL-producing ENT BSIs have been associated with significant mortality among hospitalized patients. This study was designed to determine the impact of inadequate therapy on mortality associated with ESBL-producing ENT BSIs.

METHODS: We conducted a retrospective cohort study of all inpatients with ESBL-producing ENT BSIs (1 episode per patient) at 3 tertiary care hospitals from Nov 21, 2005 to Apr 1, 2011 to examine 7 day and 30 day mortality rates in patients receiving adequate and inadequate antibiotic therapy. Patients were identified through the laboratory and their charts were reviewed. Adequate therapy was defined as receipt of an antibiotic for at least 7 days (or until death or discharge) to which the organism was susceptible. Cox regression analyses were conducted to determine associations between survival and potential risk factors or confounders.

RESULTS: We identified 112 patients with ESBL BSIs during the 5.5 year period, with a total follow-up of 2698 patient-days. Mortality rates at 7 days were 7/12 (58.3%) and 9/99 (9.1%) for patients who were inadequately and adequately treated, respectively, (OR 14.0, 95% CI: 3.7–53.3, p=0.001), while 30 day mortality rates were 7/11 (63.6%) and 22/97 (22.7%) for inadequately and adequately treated patients, respectively (OR, 6.0, 95% CI: 1.6–22.3, p=0.004). A proportional hazards model revealed that inadequate antibiotic therapy (hazard ratio [HR], 3.7, 95% CI: 1.3–10.8) and increasing PITT bacteremia score (HR, 1.5, 95% CI: 1.3–1.7) were independently associated with decreased survival.

CONCLUSIONS: Inadequate antibiotic therapy in patients with ESBL-producing ENT BSIs is associated with a 3.7 times decreased chance of survival.

H7

PIPERACILLIN/TAZOBACTAM (PTZ) IS NOT ASSOCIATED WITH REDUCED SURVIVAL IN EXTENDED-SPECTRUM BETA-LACTAMASE (ESBL)-PRODUCING *ENTEROBACTERIACEAE* (ENT) BLOODSTREAM INFECTIONS

N DEWHURST[1,2], SM POUTANEN[2,3,4], B HAMANDI[2,4], L DRESSER[2,4], M PITRE[4], A LIN[4], D LOWE[4]

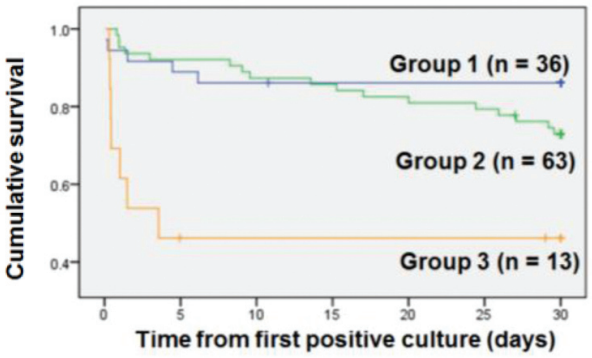
[1]St Michael's Hospital; [2]University of Toronto; [3]Mount Sinai Hospital; [4]University Health Network, Toronto, ON

OBJECTIVE: To determine if the use of PTZ to treat patients with ESBL-producing ENT bloodstream infections (BSIs) is associated with reduced survival.

METHODS: We conducted a retrospective cohort study of all inpatients with ESBL-producing ENT BSIs (1 episode per patient) at three tertiary care hospitals from Nov 21, 2005 to Apr 1, 2011 to examine the 30-day survival in those treated with PTZ as part of their therapy. Patients were identified through the laboratory and their charts were reviewed. Adequate therapy was defined as receipt of an antibiotic for ≥7 days (or until death or discharge) to which the organism was susceptible. Three treatment groups were analyzed: 1) adequate (received PTZ); 2) adequate (did not receive PTZ); 3) inadequate. Cumulative survival rates of the three groups were compared using a Kaplan-Meier survival graph and analyzed by log rank tests.

RESULTS: 112 patients with ESBL BSIs were identified. Survival rates for groups 1 and 2 were similar, but significantly higher than group 3 (Figure).

CONCLUSION: PTZ use for the treatment of ESBL-producing ENT BSIs did not negatively impact survival.



Group 1 vs. 2 – Log rank P = 0.184
Group 1 vs. 3 – Log rank P = 0.002
Group 2 vs. 3 – Log rank P = 0.004

H8

INAPPROPRIATE USE OF ANTIBIOTICS AND *CLOSTRIDIUM DIFFICILE* INFECTION

JA SRIGLEY[1,2,3], A BROOKS[4], M SUNG[4], D YAMAMURA[1,4,5], S HAIDER[1,4], D MERTZ[1,4]

[1]McMaster University, Hamilton; [2]University Health Network; [3]University of Toronto, Toronto; [4]Hamilton Health Sciences; [5]Hamilton Regional Laboratory Medicine Program, Hamilton, ON

OBJECTIVES: Inappropriate use of antibiotics may put patients at unnecessary risk for *Clostridium difficile* infection (CDI), but there are limited data on whether preceding antimicrobial therapy given to patients with CDI had been appropriate.

METHODS: Adult patients from two tertiary acute care hospitals with hospital-associated CDI were enrolled from June 2011 to May 2012. Two reviewers independently assessed appropriateness of antibiotics received in hospital during the preceding eight weeks. An intervention was implemented October 1, with review occurring on the day of CDI diagnosis or the next working day to allow direct feedback to prescribers.

RESULTS: There were 126 CDI episodes in 124 patients. In 121 episodes (96.0%), patients had received preceding antibiotics in hospital, and 93 (73.8%) had received at least one inappropriate course. A total of 456 courses of antibiotics were reviewed, with 206 (45.2%) deemed inappropriate due to incorrect diagnosis in 116 (56.3%), unsuitable spectrum of activity in 56 (27.2%), and prolonged duration in 34 (16.5%). Initial treatment for CDI was inappropriate in 25 patients (19.8%). The intervention phase included 74 patients; 17 (23.0%) were on inappropriate antibiotics on the day of CDI diagnosis. All but one of 18 recommendations were accepted by the admitting team.

CONCLUSIONS: Inappropriate use of antibiotics prior to diagnosis of hospital-associated CDI is common. Reviewing CDI patients was an effective approach to identify opportunities for improving antibiotic utilization. Providing direct feedback made it possible to educate clinicians, modify antibiotic use, and potentially improve patient outcomes.

SATURDAY APRIL 6
1115 – 1230
Room: 203

I1
NOROVIRUS IN BRITISH COLUMBIA 2012: WATCHING STRAIN REPLACEMENT IN REAL-TIME

N PRYSTAJECKY[1,2], B AUK[1], A LI[1], L LI[1], J FUNG[1], B WONG[1], C KONG[1], J ISAAC-RENTON[1,2]
[1]BC Public Health Microbiology and Reference Laboratory (BCPHMRL), Provincial Health Services Authority; [2]University of British Columbia, Department of Pathology and Laboratory Medicine, Vancouver, BC

OBJECTIVE: Norovirus (NoV) is the leading cause of gastroenteritis worldwide. Like influenza, NoV has an annual recurring seasonal pattern. Each season in North America, the number of NoV outbreaks increases rapidly in Oct and Nov, with activity peaking between Jan and Mar. Also, like the Influenza A virus, NoV evolves rapidly, producing within-group variants with new phenotypes and varying population impacts. When there are significant NoV variant changes, transmission dynamics may also change. Here, we detail the unique patterns of NoV in British Columbia in 2012, including the observation of a new dominant strain emergence in close to real-time.

METHODS: Outbreak samples are submitted according to British Columbia (BC) Public Health Microbiology and Reference Laboratory (PHRML) testing guidelines. Stool and vomitus specimens are tested by qRT-PCR and representative amplicons from each outbreak are sequenced by Sanger sequencing. Outbreak metadata is also captured.

RESULTS: In 2012, we observed a shift in the dominant GII.4 NoV strain from GII.4K New Orleans (2009) to GII.4 Sydney (2012). Accompanying this shift was a dramatically increased number of gastroenteritis outbreaks in the months of October and November to levels not seen since 2006. Furthermore, in 2012, we saw an increased incidence in NoV genogroup I and greater volume of samples from in day care and acute care hospital facilities.

CONCLUSIONS: Overall, 2012 has proven to be an unusual year for NoV in BC, with changes in transmission dynamics arising from the emergence of a new strain in the population. This work highlights the importance of NoV testing in outbreak management as well as the contributions of genotyping data and the importance of sharing this data with our public health partners on a global scale, to better understand dynamics of spread and how better to intervene. Studies to understand the evolution of spread of this new NoV variant are underway.

I2
EMERGENCE OF NOROVIRUS GII.4 VARIANTS AND CHANGES IN THE BIANNUAL PATTERN OF NOROVIRUS GASTROENTERITIS OUTBREAKS IN ALBERTA

XL PANG[1,2], E HASING[2], R TELLIER[1], JK PREIKSAITIS[2], BE LEE[2]
[1]Provincial Laboratory for Public Health (ProvLab); [2]University of Alberta, Edmonton, AB

OBJECTIVE: A unique biannual pattern of epidemic and quiescent norovirus gastroenteritis outbreak (NGO) was reported in Alberta from July 2000 to June 2008. An increased burden of NGO was associated with the emergence of new GII.4 variants every two years. In this study, 4.5 years of follow-up data of NGO and circulating norovirus was analyzed to see if the biannual pattern has continued.

METHODS: Stool specimens from suspected NGO were tested for norovirus genogroup I (GI) and II (GII) using multiplex real time RT-PCR. Sequence analysis within the capsid gene was done for one isolate of each NGO. NGO activity was analyzed using an annual observational period from July 1 to June 30 of the following year. Norovirus genotypes and the GII.4 variants were assigned using reference strains in GeneBank or The Norovirus Genotyping Tool.

RESULTS: Norovirus was detected in 68% (677/996) of suspected outbreaks between July 2008 and December 2012 with GI in 10.2%, GII in 88.8% and mixed GI and GII in 1.0%. Of the 569 NGO with sequence data, 80.1% (456) were identified as GII.4. GII.4 variant 2006b remained as the predominant strain for three winter seasons until GII.4 variant 2010 emerged in 2009. A new variant (GII4.2012) emerged in September 2011 and became predominant in Oct 2012. The biannual pattern of NGO has changed and comparatively high numbers of NGO were found in consecutive observation periods, 2010/11 and 2011/12, and were on the rise in 2012/2013.

CONCLUSIONS: A novel GII.4 variant 2012 became the predominant strain causing NGO in Alberta this winter. Changes in the unique biannual pattern of NGO in Alberta needs further study. Possible contributing factors include changing strain, virulence and antigenic drift among GII.4 norovirus variants, strain-related host susceptibility, waning herd immunity, and education campaigns on hand hygiene and infection control practices for pandemic influenza 2009.

I3
GENETIC DIVERSITY OF HUMAN METAPNEUMOVIRUS SURFACE GLYCOPROTEINS, QUEBEC, CANADA, 2001-2010

J PAPENBURG[1], J CARBONNEAU[2], S ISABEL[2], MG BERGERON[2], JV WILLIAMS[3], G DE SERRES[4], ME HAMELIN[2], G BOIVIN[2]
[1]Montreal Children's Hospital; [2]Centre de recherche en infectiologie de l'Université Laval, Quebec City, QC; [3]Vanderbilt University School of Medicine, Nashville, Tennessee, USA; [4]Institut National de Santé Publique du Québec, Quebec City, QC

OBJECTIVES: Human metapneumovirus (hMPV) is a paramyxovirus that is a major cause of respiratory tract infections (RTI) worldwide. We aim to describe the genetic diversity of the hMPV F (fusion) and G (attachment) surface glycoproteins.

METHODS: Nasopharyngeal aspirates were collected in hospitalised and outpatient children <3 years old with acute RTI in Quebec City between 2001 and 2010. Specimens were tested by multiplex PCR/DNA hybridization assay. hMPV-positive samples (n=163) underwent hMPV F and G gene sequencing. Furthermore, complete and unique hMPV-F (n=124) and G (n=217) sequences were obtained from GenBank and authors of other studies. Phylogenetic analyses were computed.

RESULTS: Sequences clustered into two groups (hMPV-A and -B) and five subgroups (A1, A2a, A2b, B1 and B2). Overall, 56% of sequences belonged to hMPV group A. Multiple subgroups circulated each year in Quebec City. With the exception of B1, each of the five subgroups was the predominant clade during ≥1 season. The A1 clade has not been detected since 2002/2003 in our cohort. There was no evidence of inter- or intragenic recombination. hMPV-F was highly conserved (mean a.a. identity: 97.4%), whereas hMPV-G exhibited greater diversity (mean a.a. identity: 48.6%). Both genes showed overall purifying evolution ($P<0.0001$). For hMPV-F, groups A and B each had 1 positively selected, but 110 and 71 negatively selected a.a. sites, respectively. For hMPV-G, we detected 12 and 10 sites under positive selection in groups A and B, respectively, as well as 26 and 20 negatively selected sites.

CONCLUSIONS: Predominant circulating hMPV lineages vary by year. hMPV-F is highly constrained and undergoes significant purifying selection. Despite high genetic diversity, hMPV-G exhibits relatively few positively selected a.a. sites.

I4
DISTRIBUTION OF RHINOVIRUSES IN HOSPITALIZED AND NONHOSPITALIZED CHILDREN WITH RESPIRATORY TRACT INFECTIONS

JF ROUSSY, J CARBONNEAU, ME HAMELIN, G BOIVIN
Research Center in Infectious Diseases, CHUL and Laval University, Quebec, QC

OBJECTIVE: Acute respiratory tract infection (RTI) is the second leading cause of death in children aged <5 years worldwide. Human rhinoviruses (HRVs) are among the most prevalent pathogens causing RTI. Studies have reported that HRVs cause upper respiratory tract infections but also bronchiolitis and pneumonia. Also, a new HRV species (HRV-C) was associated with more severe diseases than the other genotypes (HRV-A

and HRV-B) in some studies. The objective of this work was to evaluate the epidemiology and clinical characteristics caused by different HRV genotypes in a pediatric setting.

METHODS: RESPIVIR is a prospective cohort study that enrolled children aged 0 to 35 months presenting as outpatients or hospitalized with acute RTI in Quebec City during four consecutive winter seasons (2006 to 2010). A multiplex RT-PCR/DNA microarray hybridization assay was used to detect HRVs and 20 other respiratory viruses (Infiniti RVP) in nasopharyngeal aspirates (NPA). HRV genotyping was performed with a specific RT-PCR and amplicon sequencing.

RESULTS: Of the 1039 RTI episodes studied, at least one respiratory virus was identified in 86.1% of hospital and 90.1% of clinic samples. 85(8.2%) of the positive samples were HRV$^+$ with the Infiniti RVP. Of those 85 HRV, 83 were confirmed with the specific RT-PCR (67.5% in hospital and 32.5% in clinic). There were 50 HRV-A (60.2%), 1 HRV-B (1.2%) and 32 HRV-C (38.6%). Co-infection with >1 virus was more frequent in HRV$^+$ cases (39.6% vs 12% in HRV$^-$ cases, p<0.0001). RSV was the most frequently co-detected virus in HRV$^+$ cases (45.5%). Comorbidities were more frequent in HRV$^+$ cases vs HRV$^-$ cases (25.3% vs 13.1%, p=0.0045). There was no difference between HRV+ and HRV$^-$ cases or between all HRV$^+$ and HRV-C$^+$ cases for sex, age, diagnosis and hospital duration. Hospitalization >5 days was comparable for HRV$^+$ and RSV$^+$ cases (16.1% vs 16.3%).

CONCLUSION: HRVs were the third most frequently detected respiratory viruses in our pediatric cohort. HRVs were also frequently associated with lower RTI (48% of HRV$^+$ cases had bronchiolitis and 28.6% had pneumonia) although the genotype C was not linked to more severe disease in our study.

SATURDAY APRIL 6
1115 – 1230
Room: 205A

J1
ONLINE TUBERCULOSIS TRAINING FOR CANADIAN PHYSICIANS AND NURSES: A SUCCESS STORY

J NEVALA[1], A COADY[2], J COURTEMANCHE[2], V GALLANT[1], T WONG[2]
[1]Public Health Agency of Canada; [2]Health Canada, Ottawa, ON

OBJECTIVE: In Canada, between 2001 and 2010, the annual incidence rate of tuberculosis (TB) has remained relatively stable with an average of 1644 new and relapsed cases being reported annually. In 2010, foreign-born and Aboriginal Peoples accounted for 87% of all reported cases (66% and 21%, respectively). In 2010/2011, healthcare provider (HCP) needs assessments conducted by Health Canada (HC) and the Public Health Agency of Canada (the Agency) identified a need for TB training. A TB outbreak in Northern Canada also resulted in a request to develop HCP training. As a result, complementary, self-directed learning modules for HCPs serving populations at highest risk for TB in Canada were developed. The modules address specific prevention and control issues amongst First Nations and Inuit communities, foreign-born populations and those with co-morbid conditions.

METHODS: Survey results were used to develop key objectives while expert planning committees supported by the Agency and HC guided the content development. The physician modules met the accreditation criteria of the College of Family Physicians of Canada. The self-learning modules were made available in 2011 on a medical education website.

RESULTS: A cumulative total of 1690 physicians and 1080 other health care providers have completed the modules. Evaluations indicated: >90% of participants found the module's learning objectives were realistic and met and that the teaching methods used were effective. Participants had modified their approach to TB screening. Several key themes emerged including increased vigilance in screening, changes in practice, recognizing stigma around TB, more attention to issues related to vulnerable populations and to include in differential diagnosis.

CONCLUSION: The Agency and HC are building on this success and currently working on a second, collaborative initiative to develop similar TB training tools for unregulated community health workers working with First Nations, Inuit and Métis populations in Canada.

J2
ANALYSIS OF *BORDETELLA PERTUSSIS* ACTIVITY IN ALBERTA, CANADA USING A WEB-BASED PLATFORM: DATA INTEGRATION FOR ALBERTA LABORATORIES, 2004–2012

S FATHIMA[1], C FERRATO[1], BE LEE[2], L YAN[1], SN MUKHI[3], V LI[1], L CHUI[1,2], SJ DREWS[1,4]
[1]Provincial Laboratory for Public Health (ProvLab), Edmonton and Calgary; [2]University of Alberta, Edmonton, AB; [3]Canadian Network for Public Health Intelligence, Winnipeg, MB; [4]University of Calgary, Calgary, AB

OBJECTIVE: ProvLab undertakes all *B pertussis* PCR testing in Alberta (AB). Data Integration for Alberta Laboratories (DIAL) is a secure web-based platform, which extracts, interprets and collates ProvLab specimen data, information on laboratory testing and outbreak investigations. DIAL was used to analyze *B pertussis* activity in AB from July 1, 2004 to June 30, 2012.

METHODS: Repeat specimens from an individual >90 days apart were considered as a new case. Based on testing history, cases were classified as sporadic, from community-based outbreaks (COB) or institutional outbreaks (IOB). Statistical significance was analyzed using X^2 test or binary logistic regression (BLR) +/− Bonferroni correction (BC) (IBM SPSS version 20).

RESULTS: Of 20,469 suspect sporadic cases, 1349 (6.6%) were confirmed. Of 1537 suspect COB cases, 187 (12.2%) were confirmed. Of 123 suspect IOB cases, 38 (30.9%) were confirmed. There were 21 suspected COB and 23 suspected IOB investigated. Overall, 1,574 (7.1%) cases were pertussis-positive and eight sporadic cases were pertussis-indeterminate. Compared to 2005, lower positive rates were observed for sporadic cases in 2006, 2007, 2010 and 2011 (p<0.01, BLR-BC), and 2008 had a higher positive rate for COB but a lower rate for IOB (p<0.05 and p<0.01, respectively, BLR-BC). Using a reference age group of ≤8 weeks, significantly higher positive rates were identified in 5 to <10 years of age and 10 to <15 years of age only for sporadic cases (p<0.01, BLR-BC). For sporadic cases, the positivity rate was highest in northern AB (13.0%) (p<0.001, BLR).

CONCLUSION: Using DIAL, *B pertussis* positivity was different in three different settings with annual variation identified in all three settings. In the sporadic cases there was also significant difference by patient age and regions.

J3
EVALUATION OF AMPLIFICATION TARGETS FOR THE SPECIFIC DETECTION OF *BORDETELLA PERTUSSIS* BY REAL-TIME PCR

M HASAN[1,2], R TAN[1,2], G AL-RAWAHI[1,2], E THOMAS[1,2], P TILLEY[1,2]
[1]Department of Pathology, Children's and Women's Health Centre of BC; [2]Department of Pathology and Laboratory Medicine, University of British Columbia, Vancouver, BC

OBJECTIVE: Pertussis continues to be a major public health challenge in Canada. *Bordetella pertussis* PCR assays are typically based on the multi-copy insertion sequence IS481, which offers high sensitivity, but lacks specificity. In this study, we evaluated the performance of several real-time PCR assays that target different genes or genomic regions, specific to *B. pertussis*.

METHODS: Four real-time PCR assays targeting IS481 element, pertussis toxin (*ptx*)-promoter, pertactin gene and a unique genomic region of *B. pertussis*, designated as BP283, were selected from the literature. In addition, two new sets of primers and Taqman probes were designed, targeting the *ptx*-promoter and porin gene. All assays were tested using a panel of ATCC strains and culture confirmed patient isolates of *B. pertussis*, *B. parapertussis*, *B. holmesii* and *B. bronchiseptica*. The newly designed, porin assay and the previously published pertactin assay were then used to test 105 nasopharyngeal wash (NPW) specimens. ***Results:*** The test panel, comprised of different *Bordetella* species, were all detected by IS481 assay,

confirming the non-specific nature of this target. The previously published *ptx*-promoter assay detected *B. bronchiseptica* along with *B. pertussis* strains, while the *ptx*-promoter assay, designed in this study, lacked sensitivity and failed to detect all *B. pertussis* strains. The BP283 assay was specific to *B. pertussis* but it also failed to detect all *B. pertussis* strains tested. The pertactin assay detected all *B. pertussis* strains but it weakly detected *B. bronchiseptica* at a concentration of $5.4X10^5$ CFU/ml. In this panel, the best performance was observed with the newly designed porin assay, which was highly specific to *B. pertussis*. Among the 105 NPWs, 18 samples (17%) were positive by the new porin assay and 17 samples (16%) were positive by the pertactin assay, which was used as the reference assay. The new assay had a sensitivity and specificity of 100% and 98.8%, respectively, and the agreement between the two assays is 99%. The 95% detection limit of the new porin assay was 4 CFU/reaction, as determined by using an ATCC strain of *B. pertussis*.

CONCLUSION: The new real-time PCR assay, which targets the porin gene of *B. pertussis*, is highly specific and sensitive compared to other previously described *B. pertussis* PCR assays.

J4

CHARACTERIZATION OF THREE STRAINS OF THE RECENTLY-DESCRIBED GENUS *AURITIDIBACTER* REFERRED TO THE NATIONAL MICROBIOLOGY LABORATORY

K BERNARD[1], AL PACHECO[1], T BURDZ[1], D WIEBE[1], B NG[1], P VAN CAESEELE[2], R GALL[3], L HOANG[4]

[1]National Microbiology Laboratory-PHAC; [2]Cadham Provincial Laboratories; [3]University of Manitoba Department Surgery, Winnipeg, MB; [4]BCCDC, Vancouver, BC

OBJECTIVE: The genus and species novum *Auritidibacter ignavus* in the family *Micrococcaceae* was first described in 2011 derived from the ear of a man with fulminant otitis externa. This Gram positive coccobacillus is (relatively) biochemically inert but motile. We describe features for three strains from three patients, referred to the NML over the past two years, which were closest to *A. ignavus* by 16S.

METHODS: Strains studied were: two strains from BC (100628, ear sample from a 57-year-old male; 120636, left mastoid biopsy, 51-year-old female) and one from MB (120779, from the ear of a 76-year-old male with chronic otitis media and otorrhea, repeatedly treated with topical ciprofloxacin). Standard methods were used for nearly full 16SrRNA gene sequencing/phylogenetic and cellular fatty acid (CFA) composition analyses. Biochemical testing was done using tube sugars and Biomerieux or Biolog panels.

RESULTS: All strains were strict aerobes. By 16S, isolates had 99.% to 99.9% identity with each other but 97.5% to 97.7% identity to closest relative *Auritidibacter ignavus*, suggestive of a new species in that genus. CFAs were similar to those described for this genus (majority of CFAs were branch-chained types i16:0 and a17:0). Strains were catalase +, oxidase −, hydrolysed tyrosine but were biochemically inert towards a wide variety of substrates. Production of gas from nitrite and PYRa were variable. Using the Biolog system, strains were weakly reactive towards some amino or carboxyl acids but not with sugars or alcohols.

DISCUSSION: Clinicians must be aware that unusual or rare taxa recovered from clinical materials can be poorly identified by commercial panels, and so molecular identification methods will be required to assign samples correctly to genus. This may represent a species novum in the genus *Auritidibacter*.

SATURDAY APRIL 6
1115 – 1230
Room: 205B

K1

INVASIVE ASPERGILLOSIS FOLLOWING REMISSION-INDUCTION CHEMOTHERAPY FOR ACUTE LEUKAEMIA: A TERTIARY CARE CENTER INCIDENCE

S BARKATI[1], S BÉLANGER[2], B VADNAIS[2], M LAVERDIÈRE[1,2]

[1]Université de Montréal; [2]Hôpital Maisonneuve-Rosemont, Montréal, QC

OBJECTIVE: Patients undergoing remission-induction (R-I) chemotherapy for acute leukaemia (AL) are at increased risk for invasive aspergillosis (IA). Anti-mould prophylaxis is an important component in IA prevention strategies and should be guided by IA incidence. The objective of this study was to evaluate our incidence of IA in patients following R-I chemotherapy for AL.

METHODS: From the central pharmacy registries, we retrieved all patients who had R-I chemotherapy prescriptions for AL between 2008 and 2010. Patients who received posaconazole prophylaxis during their first course of R-I chemotherapy were excluded. Clinical, microbiological, pathological and radiological parameters were retrospectively recorded from patients' medical charts up to 180 days post resolution of aplasia. Diagnostic criteria of IA were based on the 2008 EORTC.

RESULTS: A total of 123 patients with AL (76% AML) were included. Twenty-two patients did not receive their chemotherapy. More than one R-I chemotherapy course was administered in 26 of the 101 treated patients. A total of 136 courses of chemotherapy were analysed. Nine patients (8.9%) developed IA (three proven and six probable) at a median of 19 days (range 11-35 days) after the beginning of chemotherapy. In seven of the nine patients (78%) IA occurred during their first R-I chemotherapy course. Three patients died within the first year post IA diagnosis.

CONCLUSION: From central pharmacy registries, we have been able to evaluate our two-year incidence of IA following R-I chemotherapy for AL. Our high incidence of 8.9% triggered consideration for targeted anti-mould prophylaxis for AL patients undergoing R-I chemotherapy. Such consideration should be based on individual center own IA incidence.

K2

POPULATION BURDEN OF LATE-STAGE CHRONIC HEPATITIS C VIRUS (HCV) SEQUELAE ATTRIBUTABLE TO HEAVY ALCOHOL USE IN BRITISH COLUMBIA

M KRAJDEN[1,2], J MCAULEY[1], D COOK[1], G BUTT[1,2]

[1]BC Centre for Disease Control; [2]University of British Columbia, Vancouver, BC

OBJECTIVE: The United States Center for Disease Control reported that an intervention to reduce alcohol intake among adults with chronic HCV infection may reduce morbidity and mortality. We estimated the direct health care costs in British Columbia (BC), from the payer perspective, attributable to heavy alcohol use for prevalent late-stage chronic HCV sequelae and death.

METHODS: Cirrhosis, decompensated cirrhosis (DCC), hepatocellular cancer (HCC), liver transplant (LT) and death were the HCV-related health outcomes analyzed. The estimated 2007 Canadian prevalence of these sequelae and their annual direct health care costs were used to estimate the BC population-attributable fraction (PAF) of sequelae attributable to heavy alcohol use (2008 $CDN).

RESULTS: The estimated prevalence of chronic HCV sequelae in BC was 2227 cirrhosis; 917 DCC; 74 HCC; 261 LT and 72 deaths. Mean annual costs per sequela were $1914 for cirrhosis; $16,283 for DCC; $23,636 for HCC; $132,820 (first year) and $19,782 (subsequent years) for LT and $8197 for death. The PAF for heavy alcohol use was estimated to be 36% for cirrhosis and DCC, and 25% for HCC, LT and death. Total annual direct health care costs for late-stage HCV sequelae were estimated to be $28.8 million, of which $9.3 million may be attributable to heavy alcohol use.

CONCLUSION: In BC, the proportion of total annual health care costs for prevalent cases of late-stage chronic HCV sequelae and death attributable to alcohol use could range from 25% to 36%. Annual alcohol-related health care costs of $9.3 million could be partially averted through innovative programming to reduce alcohol intake, which would be expected to improve health outcomes even without successful HCV antiviral treatment.

K3

COST IMPLICATIONS OF ONE-TIME HEPATITIS C VIRUS (HCV) SCREENING OF THE 1945–1965 BIRTH COHORT IN BRITISH COLUMBIA

M KRAJDEN[1,2], D COOK[1], T BULLER-TAYLOR[1], G BUTT[1,2], A YU[1]
[1]BC Centre for Disease Control; [2]University of British Columbia, Vancouver, BC

OBJECTIVE: The 1945–1965 birth cohort accounts for about 75% of prevalent HCV infections in the US, and the CDC recommends one-time HCV screening to identify those at risk of HCV-related morbidity and mortality. We estimated the cost of screening and confirmation of HCV infection for the BC 1945–1965 birth cohort.

METHODS: BC data and published estimates were used to calculate the costs of screening, HCV RNA confirmation and genotyping, and delivering results and counseling to those diagnosed.

RESULTS: The BC 1945–1965 birth cohort represents 1.36 million individuals (total population 4.4 million). 357,426 individuals have already been tested and 40,345 are anti-HCV positive, representing 64% of all individuals diagnosed with HCV to date. Based on an estimated overall cohort HCV prevalence of 1.5% to 3%, one-time screening of those still untested would be expected to identify an additional 15,018 to 30,035 individuals (total 55,363 to 70,380 HCV infections in the birth cohort). Screening, HCV RNA confirmation of chronic infection and genotyping, and delivery of results and counseling to the newly identified individuals would cost $15.6 million to $18.5 million.

CONCLUSION: Screening of the untested 1945–1965 BC birth cohort is expected to identify a substantial number of previously unidentified HCV infections. Many of these individuals would benefit from treatment to reduce HCV-related morbidity and mortality, but even if treatment were not offered to all those with chronic HCV infection, appropriate counseling and associated behaviour change (eg, reduction of alcohol intake; linkage to care and assessment) would be expected to improve health outcomes for these individuals.

K4

RISK GROUP 3 (RG3) MYCOSES IN QUÉBEC: A RETROSPECTIVE ANALYSIS OF DOCUMENTED HUMAN CASES (1988 TO 2012)

P DUFRESNE[1], S DUFRESNE[1,2], C TREMBLAY[1,3]
[1]Institut national de santé publique du Québec, Laboratoire de Santé Publique du Québec, Ste-Anne-de-Bellevue; [2]Centre Hospitalier de l'Université de Montréal, Department of Microbiology – Infectious Diseases, Hôpital Maisonneuve-Rosemont; [3]Centre Hospitalier de l'Université de Montréal, Department of Microbiology and Immunology, Hôtel-Dieu, Montréal, QC

OBJECTIVE: Risk Group 3 (RG3) pathogens are defined as those which usually cause serious disease, but do not ordinarily spread by casual contact from one individual to another. In Canada, six fungal species are classified as RG3: *Blastomyces dermatitidis*, *Cladophialophora bantiana*, *Coccidioides immitis/posadasii*, *Histoplasma capsulatum*, *Paracoccidioides brasiliensis* and *Penicillium marneffei*. The present study aimed to provide a descriptive analysis of RG3 fungal specimens submitted to the Québec provincial reference microbiology laboratory (Laboratoire de santé publique du Québec; LSPQ) in the past 24 years.

METHODOLOGY: All isolates of RG3 fungal pathogens submitted to LSPQ during the study period were compiled. Basic demographic (age, sex, sociosanitary region [SSR], date of sampling) and microbiological data (sampling site) were analyzed.

RESULTS: From 1988 to 2012, we identified 340 cases of RG3 mycoses: blastomycosis, n=173; histoplasmosis n=142; coccidioidomycosis, n=23; and *Cladiophilophora bantiana* n=2. No isolate of *Penicillium marneffei* nor *Paracoccidioides brasiliensis* was reported during that same period. *B dermatitidis* and *H capsulatum* represented 93% of RG3 fungal infections identified. Blastomycosis and histoplasmosis infections predominantly affects adult men (>80% of cases; average age >45). In the last five years, *B dermatitidis* has increasingly been reported (five year average: 11.3 vs 24 year: 6.9) whereas the numbers for *H capsulatum* have remained stable (4.6 vs 5.64).

CONCLUSION: Our results indicate that the vast majority of reported RG3 infections in Québec are caused by *B dermatitidis* and *H capsulatum*, both of which are endemic to the south-western part of Québec.

K5

DISTRIBUTION OF INVASIVE PNEUMOCOCCAL SEROTYPES IN CANADA: 2010–2012

W DEMCZUK[1], I MARTIN[1], A GRIFFITH[1], L SHERRARD[2], S DESAI[2], B LEFEBVRE[3], M GILMOUR[1]
[1]National Microbiology Laboratory, Winnipeg, MB; [2]Centre for Immunization and Respiratory Infectious Diseases, Ottawa, ON; [3]Laboratoire de santé publique du Québec, Ste-Anne-de-Bellevue, QC

OBJECTIVE: The 13-valent pneumococcal conjugate vaccine (PCV13) was introduced in most regions of Canada during 2010. This study presents shifts in the serotype distribution of invasive *Streptococcus pneumoniae* (IPD) in Canada over a three-year period from 2010 to 2012.

METHODS: A total of 1566 isolates of IPD that were submitted to the National Microbiology Laboratory between April 2010 and December 2012 were serotyped using established methods.

RESULTS: During the first year after introduction of PCV13 in Canada, a decrease in the proportion of PCV13 associated serotypes was observed only in isolates from children under two years of age from 54% (60/112) to 35% (41/116) (p=0.003). Two years after PCV13 introduction, despite no further reductions in the <2 year old age group, reduced proportions of PCV13 serotypes are now seen in the 2 to 4 year old age group from 66% (54/82) to 38% (24/63) (p<0.001); in 15 to 49 year olds from 54% (204/375) to 46% (149/325) (p=0.021); in 50 to 64 year olds from 48% (212/441) to 42% (146/348) (P=0.054); and in the ≥65 age group from 39% (227/577) to 34% (166/492) (p=0.052). An increase of the non-PCV13 serotype 22F from 2010 to 2012 has been observed in the <2 year old age group from 4% (4/112) to 13% (8/61) (p=0.030); in the 2 to 4 year old age group from 4% (2/57) to 11% (7/63) (p=0.109); and in the ≥65 age group from 10% (47/497) to 15% (73/492) (p=0.007).

CONCLUSIONS: The proportion of PCV13 serotypes has decreased over the three years since 2010. Continued monitoring of non-PCV13 serotypes, with particular attention to 22F, is important to identify possible emergent replacement serotypes.

INNOVATION ACADEMY POSTERS

Room 200B: Poster Viewing, Thursday April 4, 1100 – 1430 and 1835 – 2000 Friday April 5, 1100 – 1430

IA1

FRONT LINE OWNERSHIP APPROACH TO IMPROVE HAND HYGIENE COMPLIANCE AND REDUCE HEALTHCARE-ASSOCIATED INFECTIONS IN A LARGE ACUTE CARE ORGANIZATION

L GITTERMAN, P REASON, M GARDAM
University Health Network, Toronto, Ontario

OBJECTIVE: The overall goals of the initiative were to improve hand hygiene compliance and to reduce rates of hospital-acquired infections. A novel approach using the principles of Positive Deviance and Liberating

Structures methodologies was developed to engage staff across the organization.

METHODS: The novel approach termed Front-Line Ownership (FLO) maintains foundational elements (e.g. best practices) integral to hand hygiene but invites and encourages staff to come up with their own solutions in order to remove barriers and improve compliance. The FLO approach is based on principles of Positive Deviance and draws on complexity science. It uses Liberating Structures to engage staff and help healthcare workers interact in new ways to develop a more resilient culture that fosters individual and group responsibility for hand hygiene practice. Organizational leadership set specific hand hygienecompliance targets;however,with the exception of auditing, one standard program was not developed for the organization.Rather, it was expected that each unit develop their own ways to reach the required compliance that fit with their local context and culture. Acknowledging the complexity of hand hygiene and recognizing that each area in the hospital is unique from the next allowed staff to implement solutions that worked for them, but that would not necessarily work for another department or unit.

OUTCOMES: Over the 4 years that this approach has been implemented, University Health Network (UHN) has witnessed an over two-fold increase in hand hygiene compliance organization-wide(41% in 2008/2009, 88% in the first quarter of 2012/2013) that has surpassed all targets. Additionally, over a similar time period rates of Clostridium difficile-associated disease (CDAD) have decreased (0.58 cases per 1,000 patient days in 2007/2008, 0.46 cases per 1,000 patient days in the first quarter of 2012/2013). Similarly, rates of Methicillin Resistant Staphylococcus aureus (MRSA) have also decreased across the organization (0.41 cases per 1,000 patient days in 2007/2008, 0.33 cases per 1,000 patient days in 2011/2012). From a qualitative perspective, we have found that a culturalshift has taken place at UHN regarding hand hygiene:front line staff from all disciplines now exhibit a greater sense of ownership with respect to hand hygiene,which has the potential to lead to sustainable change. The FLO approach required multiple local actionsand with the clear support of leadership allowed for front line empowerment.This approach is novel in healthcare settings, but we feel it has great potential not only for improvements in infection control, but also for quality improvement initiatives and patient safety more broadly.

IA2

ANTIMICROBIAL PRESCRIPTION SURVEILLANCE SYSTEM (APSS): DELIVERING MAXIMUMRESULTSWITH LIMITED RESOURCES

V NAULT, M BEAUDOIN, J PERRON, L VALIQUETTE
Department of Microbiology-Infectious Diseases, Faculté de Médecine et des Sciences de la Santé, Université de Sherbrooke, Sherbrooke, QC

OBJECTIVE: AntimicrobialStewardship Programshave been identified as a key factor to reduce avoidable adverse effects, prevent antimicrobial (ATM)resistance, and reduce costs.Hospital-wide surveillance requires the revision of an overwhelming amount of clinical data. We have developed andimplemented a computerized ATM Prescription Surveillance System (APSS) to facilitate this process in our centre. APSS asynchronously identifies mismatches between dosages/intervals and published guidelines for creatinine clearance adjustments and weight. Length of treatment and route of administration are also evaluated. APSS identifies every potentially inadequate dose and alerts a clinical pharmacist with a documented message. The clinical pharmacist contacts the prescribing physician and suggests an alternate posology or treatment discontinuation. The objective of this study was to evaluate the impact of APSS use on ATM consumption and costs.

METHODS: We used data extracted from the clinical data warehouse of our712-bed academic centre. The study included all hospitalized adults receiving ATM from August23rd 2009 to August22nd 2012. APSSwas deployed on August 23rd2010 and was used 15 hours a week by a clinical pharmacist until October the 20th 2011. After this first evaluation period, APSS was fully integrated to our centre's health care plan and was used by dedicated clinical pharmacists 35 hours a week.

RESULTS: Preliminary analyses of thefirst 30 weeks of our program includes 133 082 patients-days of hospitalisation and 21 633 ATM-days, for an average of 163 ATMs-days per 1000 hospitalisations-days (RAU, ratio of ATM utilization).APSS use significantly reduced ATM utilization in comparison to the preceding 30 weeks (174 RAU, p=0,001). Interrupted Time series analyses with one degree of differentiation have shown an association with our interventionand the reduction of RAU.Since August 2010, 2718 recommendations were accepted (90% acceptance rate).

CONCLUSION: Consideringsteadily increasing costsof ATM in our centre(5% per year), our intervention reduced ATM expenses by 688 000$ in this 104 weeks period.Pharmacists, physicians, and decision makers consider our ASP to be a great success.

IA3

AN INNOVATIVE BROAD BASED SMART PHONE APPLICATION FOR ANTIMICROBIAL STEWARDSHIP

E PARFITT[1], P CAMPSALL[2], B MISSAGHI[1], V CHAUBEY[1,2], B DALTON[1], T ROSS[3], D GREGSON[1,3,4], J CONLY[1,4]
[1]Division of Infectious Diseases; [2]Department of Critical Care Medicine, University of Calgary; [3]Calgary Laboratory Services; [4]Snyder Institute for Chronic Diseases, University of Calgary Calgary, Alberta

BACKGROUND: The modification of current prescribing behavior is a critical intervention in a developing crisis in health care, with the emergence of broadly resistant pathogens. Antimicrobial stewardship aims to optimize patient care while minimizing selection of resistance and drug toxicity through appropriate use.

HYPOTHESIS: The development and distribution of an innovative smartphone "app" based on antimicrobial stewardship principles and Calgary specific antibiogram data will lead to improved antimicrobial prescribing.

METHODS: We are developing a smartphone application in Calgary as a clinical resource and knowledge translation tool based on antimicrobial stewardship principles endorsed by IDSA and SHEA. The app will contain Calgary specific antibiogram data in a user-friendly format for point-of-care use, and incorporate local clinical pathways for infectious syndromes to help clinicians select targeted therapy that minimizes toxicity and collateral damage. The app will provide general advice on prescribing as well as antimicrobial data including actual cost, bioavailability, tissue penetration, therapeutic drug monitoring tips, major drug interactions, and potential for antimicrobial resistance and *Clostridium difficile* infection. The pilot version will be trialed in the ICU. All app content is being developed within the multidisciplinary University of Calgary Antimicrobial Stewardship Working Group. Feedback on the user interface by potential consumers will shape the final product, and will be sought through incentivized surveys and focus groups. The product will be advertised through medical rounds and a social marketing campaign.

OUTCOMES: Clinical utility of the product to end-users will be measured through surveys and interviews. Outcomes of the intervention including defined daily doses, costs of therapy, resistance levels, and appropriateness of therapy for an index condition (Ventilator Associated Pneumonia) will be studied using an uncontrolled before-and-after design.

FUTURE APPLICATION: With successful launch of the ICU focused antimicrobial stewardship app, we plan to broaden the product for applicability to other settings including adult ambulatory care and paediatrics, and offer it as a template for development with other centres.

IA4
DEVELOPMENT AND IMPLEMENTATION OF SPIRIT (STEWARDSHIP PROGRAM INTEGRATING RESOURCE INFORMATION TECHNOLOGY)
M ELLIGSEN, S WALKER, L PALMAY, N DANEMAN
Sunnybrook Health Sciences Centre, Toronto, Ontario

IA5
A WEB-BASED SYSTEM FOR TRACKING OUTBREAK INVESTIGATIONS IN ALBERTA
L CHUI[1,2], B PAN[1], L YAN[1], V LI[1], B LEE[2], S MUKHI[1,2]
[1]Provincial Laboratory for Public Health; [2]University of Alberta, Edmonton, AB

OBJECTIVE: The Provincial Laboratory for Public Health (ProvLab) provides laboratory testing and support to public health officials for all outbreak investigations in Alberta. ProvLablaunched a new Exposure Investigation (EI) system within the Portal platform that provides a secure web-based tool for tracking EIs in Alberta.This study describes the implementation of the new system and data collected using this web-based tool from Nov 2011 to Dec 2012.

METHODS: The new EI system was launched on Nov2, 2011 and has been used by ProvLab, Alberta Health Services, Alberta Health, and First Nations and Inuit Health staff. The web-based EI system is secured with specific user ID and password. It has pull-down and multi-select features for users to input detailed outbreak information. Test results related to specific EIs can be viewed online by users and the outbreak data can be searched and summarized using the system. User actions are recorded for quality initiatives.

RESULTS: During the study period, 722 EIs were investigated and tracked: 521 gastrointestinal; 192 respiratory; 3 rash; 1 blood-borne; 2 nosocomial; 1 sexually transmitted; and 2 other types of outbreak. Over 550 users have been registered across Alberta and>11,100 actions including views and edits were recorded.

CONCLUSION: The new EI system includes the functionality of the previous EI application with the convenience of a web-based system, an efficient and improved interface, integrated targeted notification and access management system. It is a comprehensive information hub for outbreak investigations with enhancements and features collected over time. Client feedback has been extremely positive and the system will continue to evolve with changing needs and technology.

POSTERS

Room 200B: Poster Viewing: Thursday April 4, 1100 – 1430 and 1835 – 2000 Friday April 5, 1100 – 1430

P01
QUANTITATIVE ESTIMATION OF INTERLEUKIN-17 IN PATIENTS WITH CHRONIC LIVER DISORDERS
S MAHMOUD AHMED, M MORSI, HS OKASHA, EH ABDELMAKSOUD, NH GHAZY
Faculty Of Medicine, Alexandria University, Alexandria, Egypt

More than 20 years after the discovery of the HCV, it is now well established that HCV is of global importance affecting all countries, leading to a major global health problem that requires widespread active interventions for its prevention and control. Chronic hepatitis C was linked to the development of cirrhosis and hepatocellular carcinoma (HCC) in many areas of the world. T cells that produce IL-17 have recently been identified as a third distinct subset of effector T cells, and emerging data implicate Th17 cells as important in the pathogenesis of chronic hepatitis C infection by regulating innate and adaptive immunity, including autoimmunity. The present study was conducted to determine the role of IL-17, with its potent pro-inflammatory properties, among chronic hepatitis C cases with or without cirrhosis and HCC aimed at future immunetherapy. The study was conducted on 60 subjects with chronic hepatitis C infection before starting antiviral therapy; 20 chronic hepatitis C, 20 cirrhotic patients and 20 HCC HCV positive as well as 10 healthy subjects negative for HCV, HBV and HIV served as controls. IL-17 was quantitated after mitogen stimulated whole peripheral venous blood by commercial enzyme linked immunosorbant assay (ELISA). Our results demonstrated a significant increase in serum levels of IL-17 among cirrhotic and HCC patients infected with HCV, while in chronic hepatitis C virus cases, elevated IL-17 values were nonsignificant compared to controls. We can conclude that IL-17 plays an important role in HCV immunopathogenesis. It might be used as an indicator for cirrhosis and HCC as it promotes tumour growth by facilitating angiogenesis in the tumor microenvironment. Also, its therapeutic application needs to be furtherly evaluated by in vivo studies in experimental animals aiming at future immunotherapy.

P02
A NOVEL AEROTOLERANT GRAM-VARIABLE *LEPTOTRICHIA SPP.* CAUSING SEVERE PNEUMONIA IN A LYMPHOMA PATIENT RECENTLY TREATED WITH RITUXIMAB
G GERMAN[1], D HALDANE[2], B NG[3], K BERNARD[3]
[1]Health PEI – Queen Elizabeth Hospital, Charlottetown, PEI; [2]Division of Infectious Diseases, Department of Pathology and Laboratory Medicine, Dalhousie University, Halifax, NS; [3]Special Bacteriology Section, National Microbiology Laboratory, Public Health Agency of Canada, Winnipeg, MB

HISTORY: An elderly female nonsmoker presented from home with four days of increasing shortness of breath, sore throat, and a nonproductive cough unresponsive to moxifloxacin. Two weeks before presentation she had finished a month long course of Rituximab for treatment of her MALT lymphoma. There were no constitutional symptoms, recent travel, or exposure to animals.

ON EXAM: she was afebrile but in moderate distress, with an oxygen saturation of 87% on room air. Respiratory rate was 20, pulse 103, and blood pressure 93 mmHg/57 mmHg. The rest of her respiratory and cardiac exam was essentially unremarkable.

INVESTIGATIONS: CBC haemoglobin of 112 g/L; otherwise normal. LDH was 419 IU/L, and CRP was 127 mg/L. Imaging of the chest revealed ground glass shadowing in upper lung fields with subcentimeter pulmonary cysts. PJP and AFB stains and extensive viral testing from a BAL specimen were negative. Routine culture grew +1 *Pseudomonas aeruginosa*, yeast, and +4 fusiform gram variable bacilli. This fusiform was catalase and oxidase negative and the initial differential included *Actinomyces spp*, aerobic actinomycetes and *Capnocytophaga spp*. Mass spectrometry and 16S ribosomal gene sequencing at the National Microbiology Laboratory indicated that the organism was most likely a novel *Leptotrichia* species. The patient improved on beta-lactam-based therapy and was discharged from ICU and went home at her baseline.

DISCUSSION: This is as far as we are aware the fourth case report of *Leptotrichia spp*. causing a severe pneumonia. The organism's atypical gram stain reaction, aerotolerance, and the clinical failure of moxifloxacin are noteworthy features.

P03
LIVER FIBROSIS AS MEASURED BY TRANSIENT ELASTOGRAPHY IN NORTH AMERICAN HIV-INFECTED PATIENTS: DOES METABOLIC SYNDROME MATTER?
V MARTEL-LAFERRIERE, N PARIKH, T KUSHNER, M NG, M GAISA, M MULLEN, DT DIETERICH
Mount Sinai School of Medicine, New York, New York, USA

BACKGROUND: Blanco et al (J. Viral Hepatitis, 2011) found an association between metabolic syndrome parameters and elevated transient elastography (TE) scores in HIV-infected Spanish patients. Since the prevalence of dyslipidemia and diabetes are higher in the North American population, the aim of this study is to determine if the same is true in our HIV-infected population.

METHOD: This is a case-controlled IRB-approved study. A TE measurement was performed on consecutive patients attending a general HIV-clinic. Patients with a BMI >40 were excluded. Cases were defined as patients with an elevated TE (≥9.5 kPa). Data were analysed in SPSS.
RESULTS: The study included 81 patients. Four patients had to be excluded because of invalid TE measurement secondary to chest fat (4.9%). Of the remaining 77 patients, nine (11.7%) had a score ≥9.5 kPa. The difference in the mean TE score was statistically significant between the groups (14.5 (10 to 18.9) vs 4.9 (4.25 to 6.2); p=0.001). Cases and controls were similar for sex and race, but differed for age (56 (52 to 61) vs 51 (43.5 to 57); p=0.03). Diabetes (33.3% vs 4.4%; p=0.02) and active HCV infection (44.4% vs 9.0%; p=0.02) were both associated with higher TE whereas BMI (p=0.39) and total cholesterol level (p=0.57) were not associated with higher TE scores. Dyslipidemia (p=0.15) and random glucose levels (p=0.14) showed a trend toward lower TE score but were not statistically significant.
CONCLUSION: As in the Spanish study, hepatitis C and impaired glucose metabolism have a significant impact on TE score. The impact of dyslipidemia may be blunted by pharmacological control, which does not seem to be the case for diabetes. High BMI limits the ability to perform TE, and it is those patients who are more likely to have elevated TE and impaired glucose tolerance.

P04
ON-TREATMENT RESPONSES TO TELAPREVIR-BASED HEPATITIS C TREATMENT ARE SIMILAR IN HIV/HCV CO-INFECTED AND HCV MONOINFECTED PATIENTS

V MARTEL-LAFERRIERE[1], S BRINKLEY[2], K BICHOUPAN[1], S POSNER[1], A STIVALA[1], P PERUMALSWAMI[1], TD SCHIANO[1], M SULKOWSKI[2], DT DIETERICH[1], AD BRANCH[1]
[1]Mount Sinai Scool of Medicine, New York, New York; [2]John Hopkins University, Baltimore, Maryland, USA

BACKGROUND: Telaprevir is not FDA-approved for HIV/HCV co-infected patients. This study compared virologic responses and side effects of co- and mono-infected patients over the active treatment phase of telaprevir-based therapy.
METHODS: Baseline, four-, 12-, 24- and 48-week data of 33 co-infected and 117 mono-infected patients receiving telaprevir-based therapy at two medical centers were reviewed, with IRB approval. HCV viral load below the limit of detection (Roche Ampliprep test) was considered 'undetectable'. End of treatment (EOT) was week 24 for patients eligible for response guided therapy (RGT) or week 48 for other patients, including all co-infected patients. Severe anemia was defined as an hemoglobin ≤8.9 g/dL or a drop ≥4.5 g/dL. Data were analyzed on an ITT basis.
RESULTS: At baseline, the groups of co-infected (n=33) and mono-infected (n=117) patients did not differ significantly in age, gender, Fib-4 score ≥3.25 [16/33 (48.5%) vs 40/113 (35.4%); p=0.22] or HCV viral load. The median baseline CD4+ count of co-infected patients was 489 cells/μL (IQR 316 to 783); 81% (25/31) had an HIV viral load lower than the level of quantification/detection. The co-infected group had a smaller percentage who were treatment-naïve (12.1% vs 33.8%) and a higher percentage of nonresponders (66.6% vs 43.6%). Seventeen mono-infected patients (14.5%) qualified for RGT. The percentages of co-infected vs mono-infected patients with undetectable HCV viral load during treatment were as follows: week 4, 42.4% vs 43.6%; p=1.00; week 12, 72.7% vs 65.8%; p=0.532; week 24, 72.7% vs 58.1%; p=0.16. At EOT, a higher percentage of co-infected patients were HCV VL negative, but the difference was not statistically significant, 63.6% vs 51.2%; p=0.24. By EOT, 18.2% of co-infected vs 13.7% of mono-infected patients had discontinued treatment due to side effects (p=0.58). The incidence of severe anemia was similar in the two groups (3.3% vs 37.6%; p=0.69); however, the co-infected group showed a strong trend toward a reduced incidence of rash (15.2% vs 34.1%; p=0.05), and they had a lower incidence of rectal symptoms (12.1% vs 43.6%; p=0.001).
CONCLUSIONS: On-treatment virologic responses were comparable in this group of co- and mono-infected patients. Rash and rectal symptoms tend to be less frequent in co-infected patients but discontinuations for side effects are similar. SVR data are needed to determine clinical effectiveness.

P05
RHINO-ENTOMOPHTHOROMYCOSIS IN AN HIV-INFECTED PATIENT: A CASE REPORT

R DHURAT[1], R KOTHAVADE[2,3,4], A DANDLE[1], S GHATE[1]
[1]Lokmanya Tilak Municipal General Hospital and Medical College, Sion, Mumbai; [2]KMC-IC, University of Manipal, Manipal, India; [3]Grant Medical College and Sir JJ Group of Hospitals, Mumbai, India; [4]EPCOR, Edmonton, AB

OBJECTIVE: We describe the significance of second line ART in modulating an antifungal response to itraconazole in an HIV-infected patient with rhino-entomophthoromycosis.
METHODS:CD4 counts, viral load, CT scan, histopathology and mycology
RESULTS: A 32-year old HIV-infected male presented with swelling, redness and blockage of left side of the nasal passage from nine months. Patient history: trauma preceding the lesion and persistent increase in size of the lesion. FNAC of the inferior turbinate mass: remarkable. CT scan of the paranasal sinuses: the middle and inferior turbinate hypertrophy with adhesions causing blockage of nasal pathway. PAS stain: negative for any fungal elements. Acanthosis with dense infiltration of neutrophils, lymphocytes and eosinophils were clearly evident throughout the dermis. Repeated culture of the processed skin biopsy specimens yielded *Conidiobolus coronatus* colonies on SDA. This patient did not respond to a prolonged course of oral itraconazole(200 mg/day) in combination with potassium iodide drops. Sensitivity testing on HIV revealed resistance to zidovudine and nevirapine. Further laboratory investigations suggested increased viral load by 50,000 copies/ml with a decrease in CD4 count of 20 cells/mm^3. Switching to second line ART with continuation of itraconazole resolved the lesions after 3 months.
CONCLUSION: Poor antifungal response in HIV-infected patients needs to be evaluated with first-line ART resistance. Additional studies are needed to determine the basis of therapeutic response to antifungal drugs in conjunction with second-line ART in HIV-infected patients with *rhino-entomophthoromycosis*.

P06
A PROGRAM-BASED APPROACH FOR DEVELOPMENT OF INFRASTRUCTURES AND SERVICE DELIVERY IN A LARGE URBAN COMMUNITY OF CANADA

Y MIRZANEJAD
University of British Columbia/Fraser Health Authority, Vancouver/Surrey; Division of Infectious Diseases, University of British Columbia, Vancouver, BC

OBJECTIVE: Developing a formal community-based infectious diseases (ID) and health care epidemiology program within a regional health care authority by bringing together the consultation services of inpatient care, outpatient care, home IV program, outpatient parenteral antibiotics therapy (OPAT), HIV and Hepatitis C along with research and education for evidence based practice in ID deliverable under one comprehensive leadership.
METHODS: From 2004 the following services are to be formed step by step: 1) ID inpatient services; 2) Home IV antibiotics therapy led by ID specialist and forming a multidisciplinary team to enroll eligible patients to enter the program with management plan and discharge (for long term antibiotics therapy); 3) Outpatient parenteral antibiotics therapy to decongest emergency and acute care beds (for short term intravenous antibiotics therapy); 4) Outpatient clinic for rapid access to ID assessment to support community practitioners or follow-up on discharged patients; 5) Interdisciplinary HIV/ Hepatitis C clinic; 6) Leading management of infected wound care in the hospital and community health.
OUTCOMES: In our five years research in motion the following results were produced: 1) Decongesting acute care beds and emergency room; 2) Diverting inpatient care to outpatient care by providing treatment in OPAT unit; 3) Shortening the length of hospital stay by early involvement of ID in management plan; 4) Providing antimicrobial stewardship; 5) Substantial cost saving and resource utilization and prevention of duplication in our health care system. By developing the above infrastructures

new position opportunities have been created to recruit newly trained ID specialists in this innovative supported framework.

P07

EVALUATION OF ANTIMICROBIAL STEWARDSHIP PROGRAM ON LEUKEMIA SERVICE THROUGH PROSPECTIVE AUDIT AND FEEDBACK

M SO[1], L PIVNICK[2], M STEINBERG[2], T JIVRAJ[2], S LAPINSKY[2,3], A MORRIS[1,2,3], S HUSAIN[1,3]

[1]University Health Network; [2]Mount Sinai Hospital; [3]University of Toronto, Toronto, ON

BACKGROUND: Patients with haematological malignancies are vulnerable to infections and often receive multiple courses of antimicrobials in their disease management. Emergence of multi-drug resistant organisms and *C difficile* infections (CDI) due to widespread and prolonged antimicrobial exposure have highlighted the need for antimicrobial stewardship programs (ASP). University Health Network launched its ASP (prospective audit and feedback) on leukemia service in February 2010. The objective of this study was to assess the impact and sustainability of ASP program in leukemia service at UHN.

METHOD: Data were retrospectively evaluated by comparing the pre-ASP period with post-ASP period using the following outcome measures: antibiotic consumption (defined daily dose [DDD]/100 patient-days [PD]); costs ($/100 PD). Incidence of bacteremias, fungemias and CDI were reviewed. Transfer to the ICU was used as a surrogate marker of patient outcome. Monthly antibiotic DDD and cost/100 PD were compared based on fiscal year using unpaired *t* test (p value <0.05 = statistical significance). Other outcome measures were reviewed for trend.

RESULTS: ASP significantly reduced antibiotic consumption over two years. While monthly antibiotic cost/100 PD decreased, it did not reach statistical significance. Antifungal cost was significantly reduced with marginal increase in consumption, owing to improved consistency in fluconazole prophylaxis. There was a corresponding decrease in more costly antifungals such as voriconazole and liposomal amphotericin. Incidence of fungemia was reduced by 62%. Incidence of gram-positive bacteremia decreased but gram-negative bacilli bacteremia increased. An outbreak in FY 11/12 led to a rise in combined CDI incidence for FY 10-12. Overall percentage of positive cultures remained steady. A detailed analysis of patient characteristics, eg, severity of illness, duration of neutropenia and chemotherapy received, is required to understand the rise in ICU transfer and to assess clinical impact of ASP in leukemia ward.

P08

EFFECT OF PROSPECTIVE AUDIT AND FEEDBACK AS PART OF AN ICU ANTIMICROBIAL STEWARDSHIP PROGRAM ON ANTIMICROBIAL SUSCEPTIBILITY TO *PSEUDOMONAS AERUGINOSA*

K DUPLISEA[1], T JIVRAJ[2], S POUTANEN[2,3], S NELSON[2,3], L DRESSER[1,3], M STEINBERG[2], S LAPINSKY[2,3], N LAZAR[1,3], J SINGH[1,3], C BELL[2,3], A MORRIS[1,2,3]

[1]University Health Network; [2]Mount Sinai Hospital; [3]University of Toronto, Toronto, ON

BACKGROUND: Antimicrobial stewardship programs (ASP) are being implemented in intensive care units (ICU) across Canada as clinicians recognize the importance of preserving our remaining antimicrobial. An ASP that incorporates prospective audit and feedback (PAF) three to five days per week has been operational in three medical-surgical ICUs of two large academic centers for over three years. The purpose of this study was to determine the effect of PAF on ICU susceptibility patterns of *P aeruginosa* and to determine the impact on antimicrobial consumption.

METHODS: A search was performed using the microbiology Laboratory Information System (LIS) to identify isolates of *P aeruginosa* in each of the three ICUs. The post-implementation analysis period began one month after introduction of PAF until July 31, 2012. The pre-implementation analysis period included an equal number of months prior to introduction of PAF. Antimicrobial consumption was reported as Defined Daily Doses (DDD).

RESULTS: Improvement in *P aeruginosa* susceptibility, including a reduction in MDR *P aeruginosa*, occurred with the implementation of PAF at ICU 1. During this time period there was an overall reduction in anti-pseudomonal consumption. No significant improvement in *Pseudomonas* susceptibility was seen at either ICUs 2 or 3 despite reductions in anti-pseudomonal consumption. Further analysis is required to determine if the different patient populations may account for the variations in effect across the three sites.

P09

INVESTIGATION OF CLUSTERS OF BACTERIAL CONTAMINATION OF PRIVATE WELL WATER IN SOUTHEASTERN ONTARIO

J KROLIK[1,5], G HALL[2], K RANDHAWA[1], SYL CHENG[2], A JOYCE[5], G EVANS[1,3], P BELANGER[1,4], A MAJURY[1,5]

[1]Queen's University, Kingston; [2]University of Toronto, Toronto; [3]Kingston General Hospital; [4]Kingston Frontenac and Lennox & Addington Health Unit; [5]Public Health Ontario, Kingston, ON

OBJECTIVE: One in ten Canadians relies on private well water supplies as their primary drinking water source. Well water is susceptible to microbial contamination by *Escherichia coli* (*E coli*), coliforms, and other potentially pathogenic organisms. It is estimated that 90,000 illnesses occur each year in Canada due to contaminated water, but there is currently a lack of knowledge concerning waterborne risks associated with wells. In this study, GIS tools were used to provide a geospatial description of the prevalence of *E coli* and coliform contamination among private wells in southeastern Ontario, for the years 2011. Furthermore, a spatial scan statistic was employed to delineate clusters with increased risk of contamination.

METHODS: Well water data were extracted from the Water Testing Information System (WTIS) and geocoded. Clustering analyses were conducted to determine areas of *E coli* and coliform contamination.

RESULTS: Of 8134 uniquely geocoded well water entries within four public health units submitted to Public Health Ontario Laboratory in Kingston, 8.5% were positive for *E coli*. Clustering analyses identified one significant cluster of increased risk of *E coli* contamination (relative risk = 1.75, p<0.001) at the 50% maximum population size level. It contained two smaller significant clusters (relative risk = 2.38, p=0.017; relative risk = 2.68, p=0.065) when the analysis was performed at the 5% level. Additionally, an area with increased risk for high coliform contamination was also identified (relative risk = 5.09 and 5.27 for 50% and 5% maximum sizes, respectively; p<0.001).

CONCLUSION: A spatial scan statistic revealed two clusters of *E coli* and one cluster of coliform contamination in drinking well waters. The identification of clusters of high-risk areas, at varying cluster population maximum sizes, provides important information for public health practitioners, enabling better assessment and targeted interventions, as well as the opportunity to investigate the causes of microbial contamination in these regions.

P10

HELICOBACTER IN BELUGA WHALES

M LEE[1], L MEASURES[2], S MAN[1], K FERNANDO[1], M MORSHED[1]

[1]BCCDC Public Health Microbiology and Reference Laboratory, Vancouver, BC; [2]Marine mammal health, Fisheries and Oceans Canada, Mont-Joli, QC

OBJECTIVE: The aim of our study was to investigate the presence of *Helicobacter spp* in beluga and its genetic relationship with *Helicobacter spp* found in humans.

METHODS: Eighteen beluga (16 from Hudson Bay, and two from the St. Lawrence) were included in this study. DNA was extracted from different parts of the digestive system (stomachs, intestine) and faeces from beluga using the QIAGEN Extraction Kit. The ABI 9700 was used to amplify the 16S and 23S rRNA genes of *Helicobacter spp*. Cycle sequencing was performed with ABI BigDye® Chemistry and analyzed on the ABI 3130xl. The sequences obtained were analyzed and a phylogenetic tree was built using Geneious software.

RESULT: All beluga (but not all organs) were PCR positive for *Helicobacter spp*. Three stomach samples from each beluga were subjected

to sequencing and analysis. Phylogenetic information showed all sequences were clustered under one clade, *Helicobacter cetorum*, which is closely related to *Helicobacter pylori*, with little variation between clades.
CONCLUSION: All beluga (18/18) examined were positive for *Helicobacter spp*. More than 50% of faecal samples were positive which indicated that faecal testing can be used as a non-invasive method for screening *Helicobacter spp* in beluga. The species of *Helicobacter* found in beluga is genetically closest to human *H pylori* compared with all available animal species examined. Further studies are needed to determine the role of *Helicobacter spp* in the beluga and its pathogenesis.

P11
LEISHMANIASIS IN BRITISH COLUMBIA: A 20 YEAR RETROSPECTIVE REVIEW

M MORSHED[2,3], Q WONG[2], T LO[2], M ROMNEY[1,3], J ISAAC-RENTON[2,3]
[1]Providence Health Care; [2]BC Public Health Microbiology Reference Laboratory; [3]Department of Pathology and Laboratory Medicine, University of British Columbia, Vancouver, BC

INTRODUCTION: *Leishmania* is a blood and tissue protozoan parasite primarily transmitted in tropical regions. In its visceral form (kala-azar), the parasite spreads to spleen, bone marrow and liver, causing high case fatality rates in untreated cases. In its other two forms, spread is limited to skin and mucous membranes with a lower mortality rate but with significant morbidity. The different major clinical syndromes are caused by different species of *Leishmania*. Following the diagnosis of an unusual case in a returned traveler, we retrospectively reviewed province-wide *Leishmania* data for British Columbia (BC).
METHODS: A patient presenting with multiple cutaneous and mucosal lesions refractory to routine antibiotic treatment prompted a review of records for all cases of leishmaniasis diagnosed in BC for the period 1992 to 2012. In BC, unusual parasitological cases are diagnosed by hospital microbiology laboratories partnering with the BC Public Health Microbiology Reference Laboratory (PHMRL). Stained smears from lesions are often examined at larger academic hospital centres with biopsies submitted to PHMRL for culture with Evans Modified Tobies medium**.** Parasites isolated from cultures are submitted to the Centers for Disease Control and Prevention (CDC) for speciation.
RESULTS: Details of the current case are described including an outline of diagnostic procedures and the treatment regimen used. This is a rare infection of returned travelers in BC. Overall during the 20-year study period, 23 cases of leisimanaisis were documented. Twelve cases were in males, 11 in females; mean age was 34 years ranging between 10 and 54**.** The species isolated by culture included: *Leishmania braziliensis*, *L guyanensis*, *L major*, *L mexicana*, *L panamensis*, and *L tropica*. There were no cases of kala-azar detected.
CONCLUSIONS: This case highlights the need to have a collaborative process in place for diagnosing this rare but significant infection, and to consider leishmaniasis in returned travelers presenting with cutaneous/mucocutaneous lesions refractory to conventional antibiotic therapy.

P12
CANWARD 2012: ANTIMICROBIAL RESISTANCE IN PATHOGENS ISOLATED FROM CANADIAN HOSPITAL CLINICS, EMERGENCY ROOMS, MEDICAL/SURGICAL WARDS AND INTENSIVE CARE UNITS

M BAXTER[1], HJ ADAM[1,2], M MCCRACKEN[3], AJ DENISUK[1], G GOLDING[1,3], JA KARLOWSKY[1,2], DJ HOBAN[1,2], GG ZHANEL[1]
[1]University of Manitoba; [2]Diagnostic Services of Manitoba; [3]National Microbiology Laboratory, Winnipeg, MB

OBJECTIVES: The CANWARD study assesses the pathogens causing infections in patients affiliated with Canadian hospitals and evaluates the prevalence of antimicrobial resistance in these isolates.
METHODS: Twelve tertiary care centres across Canada submitted pathogens causing infections from patients attending clinics (C), emergency rooms (ER), medical and surgical wards (W) and intensive care units (ICU) in 2012. Susceptibility testing was performed by CLSI microdilution methods.
RESULTS: 2808 isolates were collected: 41.6%, 38.0%, 10.6%, and 9.8% from blood, respiratory, urine and wound/IV site specimens, respectively. Isolates were from patients on W 35.2%, ER 25.5%, ICU 22.1%, and C 17.2%. The most common pathogens were: *S aureus* (MSSA) 20.1%, *E coli* 17.8%, *P aeruginosa* 9.4%, *K pneumoniae* 6.0%, *H influenzae* 5.3%, and *S pneumoniae* 5.1%. Resistance rates (RR) for *E coli* were: 0% for meropenem (MER), ertapenem (ERT) and tigecycline (TGC), 2.4% piperacillin/tazobactam (PTZ), 8.4% ceftriaxone (CTR), 8.8% gentamicin (GEN), 25.6% ciprofloxacin (CIP) and 27.0% trimethoprim/sulfamethoxazole (SXT). For *P aeruginosa*, RR were 1.1% colistin (COL), 4.9% PTZ, 5.3% GEN, 9.1% MER and 10.2% CIP. RR for MRSA were: 0% vancomycin (VAN) and linezolid (LZD), 0.8% daptomycin (DAP) (1/124 isolates; MIC-DAP 4, VAN 2 µg/mL), 2.4% tigecycline, 4.0% SXT, 31.4% clindamycin, 73.4% CIP and 79.0% clarithromycin. Overall, the prevalence of MRSA, VRE and ESBL- *E. coli* was: 18.0%, 7.8% and 7.8%, respectively.
CONCLUSIONS: RR for *E coli* were lowest with MER, ERT, TGC and PTZ, while RR for *P aeruginosa* were lowest with COL, PTZ, and GEN. For MRSA, no resistance occurred with VAN or LZD.

P13
WITHDRAWN

P14
ALARMING INCREASE IN EXTENDED-SPECTRUM BETA-LACTAMASE (ESBL) PRODUCING GRAM-NEGATIVE BACILLI (GNB) ISOLATED FROM PATIENTS WITH INTRA-ABDOMINAL INFECTIONS IN CANADA

SM POUTANEN[1,2], R RENNIE[3,4], D ROSCOE[5], B TOYE[6], C LEE[7], V LOO[8], D HOBAN[9,10], D GRIMARD[11], P JUTRAS[12], J BLONDEAU[13], JD PITOUT[14], A GALARNEAU[15], J POULIOT[15]
[1]Mount Sinai Hospital/University Health Network; [2]University of Toronto, Toronto, ON; [3]University of Alberta Hospital; [4]Alberta Health Services, Edmonton, AB; [5]Vancouver General Hospital, Vancouver, BC; [6]The Ottawa Hospital, Ottawa; [7]St. Joseph's Healthcare, Hamilton, ON; [8]Montreal General Hospital, Montreal, QC; [9]Health Sciences Centre, Winnipeg, MB; [10]International Health Management Associates Inc, Schaumburg, Illinois, USA;[11]Centre de Santé et de Services Sociaux de Chicoutimi, Chicoutimi; [12]Centre Hospitalier Régional de Rimouski, Rimouski, QC; [13]Royal University Hospital, Saskatoon, SK; [14]University of Calgary, Calgary, AB; [15]Merck Canada, Kirkland, QC

OBJECTIVE: Antimicrobial-resistant GNB have been increasing worldwide, largely due to ESBLs. The Study for Monitoring Antimicrobial Resistance Trends (SMART) is a global surveillance study monitoring the in vitro susceptibility of GNB from intra-abdominal infections. SMART data describing GNB in Canada has not been previously described.
METHODS: SMART includes eleven centres from distinct geographic regions in Canada. Intra-abdominal GNB, one per patient per year, were collected for the period 2008-2011, and sent to a central laboratory for testing. ESBL production and broth microdilution susceptibility were assessed following Clinical Laboratory Standards Institute. Antimicrobials tested included: amikacin, cefoxitin, cefotaxime, ceftriaxone, ceftazidime, cefepime, ampicillin-sulbactam, piperacillin-tazobactam, ertapenem, imipenem, ciprofloxacin and levofloxacin.
RESULTS: A total of 1714 isolates were collected. Five species made up 84% of isolates: *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Enterobacter cloacae* and *K oxytoca*. The most active antimicrobials were amikacin, ertapenem and imipenem. For the four organisms that were tested for the presence of ESBL (*E coli*, *K pneumoniae*, *K oxytoca*, *P mirabilis*), there was a significant increase in those that produced ESBL from 3.8% in 2008 to 11.6% in 2011 (p=0.001), mainly due a rise in ESBL-positive *E coli* from 4.8% to 14.0% (p=0.007).
CONCLUSION: Between 2008-2011, there was an alarming increase in intra-abdominal ESBL-producing GNB in Canada. Increased efforts are needed to limit the spread of ESBL GNB and careful consideration needs to be taken when selecting empiric antibacterial therapy for patients with intra-abdominal infections.

P15

POLYCLONAL OUTBREAK OF KPC-3-PRODUCING *ENTEROBACTER CLOACAE* AT A SINGLE HOSPITAL IN MONTRÉAL, CANADA

L HARAOUI[1], LF MATASEJE[2], S LEVESQUE[3], B LEFEBVRE[3], R BLANCHETTE[1], M TOMKINSON[1], MR MULVEY[2], M MILLER[1]
[1]Jewish General Hospital, Montreal, QC; [2]Public Health Agency of Canada, Winnipeg, MB; [3]Laboratoire di sante publique du Quebec, Ste-Anne-de-Bellevue, QC

OBJECTIVE: *Klebsiella pneumoniae* carbapenemases (KPC) are among the most common enzymes causing carbapenem resistance identified in Enterobacteriaceae causing infections and outbreaks globally. In this report we describe the characterization of KPCs identified from *Enterobacter cloacae* during a single hospital site outbreak in Montréal.

METHODS: Isolates were collected from September 2010 to December 31, 2011. All carbapenem-resistant *E cloacae* from screening and clinical specimens were included in the analysis. PCR was used to identify common carbapenemase-producing isolates. Antimicrobial susceptibility was conducted Vitek2. PFGE was conducted to determine strain relatedness using the restriction enzyme XbaI. Plasmids producing KPCs were replicon-typed using PCR and digested using EcoRI to obtain a fingerprint pattern.

RESULTS: Isolates of KPC-3-producing *E cloacae* (n=26) were identified from 16 patients: 12 men and four women. Seven isolates were recovered from clinical specimens (six patients): two were from blood, three from urine and two from pus. The other 19 isolates were recovered from rectal screens (11 patients). Using PFGE, seven macrorestriction pattern clusters were identified. KPC plasmid analysis revealed numerous Inc types; IncN (n=9); IncN, A/C (n=1); IncP, L/M (n=5) and nontypable (n=11). Fingerprint analysis of the KPC plasmids revealed similar KPC plasmids were found in different *E cloacae* PFGE types. Furthermore, similar *E cloacae* PFGE types contained different plasmid types.

CONCLUSION: This outbreak highlights the potential problems associated with controlling a KPC outbreak at a single site. The multiple PFGE clusters and replicon types associated with the KPC plasmids suggests mobility of the element between strains and the potential mobility of the KPC gene between plasmids. Additional work will be required to sort out the molecular epidemiology of this outbreak.

P16

QUANTIFYING CHANGES IN ANTIMICROBIAL RESISTANCE: A DRUG RESISTANCE INDEX FOR *E COLI*

D PATRICK[1,2], C CHAMBERS[1], D PURYCH[3,4], M CHONG[1], F MARRA[1,2]
[1]BC Centre for Disease Control; [2]University of British Columbia, Vancouver; [3]BC Biomedical Laboratories Ltd; [4]Fraser Health Authority, Surrey, BC

OBJECTIVES: A drug resistance index (DRI) measures the average effectiveness of a set of antimicrobials used to treat a bacterial infection. The objective of this analysis was to derive a DRI for urinary tract infections caused by *Escherichia coli* in British Columbia for years 2007 to 2010 and to examine DRI trends over time and across patient demographics.

METHODS: Indication-specific utilization data for cystitis were obtained from the PharmaNet database of outpatient prescriptions for oral antimicrobials linked to diagnostic data from physician payment files. Resistance data for *E coli* urinary isolates were obtained from BC Biomedical Laboratories. A DRI was calculated by multiplying the rate of resistance to drug k at time t by the proportional rate of use for drug k used to treat cystitis at time t and summing across drug classes. Relevant drug classes included: extended-spectrum penicillins, first- and third-generation cephalosporins, sulfonamides and trimethoprim, fluoroquinolones, and nitrofuran derivatives. Higher index values indicate more resistance.

RESULTS: DRI values remained stable over time around 0.18 (p=1.000). For adults (≥15 years old), there was a significant increasing trend from 0.10 among those 15 to 19 years to 0.32 among those >90 years (p<0.001). For children (<15 years old), there was a significant declining trend from 0.35 among those <1 year to 0.18 among those 10 to 14 years (p<0.001). Males had consistently higher DRIs than females for all age groups.

CONCLUSIONS: Results by age group suggest concerning antimicrobial resistance trends for *E coli* among young children and the elderly. Further work is required to summarize the impact of resistance across all pathogens affecting a population.

P17

SERRATIA MARCESCENS BETA-LACTAMASE (SME): THE NEW CARBAPENEMASE ON THE BLOCK IN CANADA

LF MATASEJE[1], DA BOYD[1], Z HUSSAIN[2], M IMPERIAL[3], B LEFEBVRE[4], P VAN CAESEELE[5], B WILLEY[6], MR MULVEY[1]
[1]Public Health Agency of Canada, Winnipeg, MB; [2]Moncton Hospital, Moncton, NB; [3]British Columbia Center for Disease Control, Vancouver, BC; [4]Laboratoire de sante publique du Quebec, Sainte-Anne-de-Bellevue, QC; [5]Cadam Provincial Laboratory, Winnipeg, MB; [6]Mount Sinai Hospital, Toronto, ON

BACKGROUND: Reports of the class A carbapenemase SME globally are rare. In this report we describe the emergence of SME in Canada from isolates submitted to the National Microbiology Laboratory (NML).

METHODS: *Serratia marcescens* were submitted to the NML for confirmation of a carbapenemase between January 2010 to April 2012 by hospitals and provincial public health laboratories on a voluntary basis. Antimicrobial susceptibility testing was determined using Vitek2 AST-GN25 and Etest. PCR for the detection of the SME carbapenemase gene and the beta-lactamases SHV, TEM, CTX-M, OXA-1 and CMY-2 was conducted. Strain typing was done using pulsed-field gel electrophoresis (PFGE).

RESULTS: A total of 18 nonduplicate *S marcescens* producing SME were confirmed at the NML between 2010 (n=4), 2011 (n=8), and up to April of 2012 (n=6). Isolates originated from BC (10), QC (4), MB (2), ON (1) and NB (1). Fifteen (82%) isolates were from male patients. Sixty-five percent of patients were over the age of 65. The most common site of isolation was the respiratory tract (39%). Sequencing confirmed 13 bla_{SME-1} and five bla_{SME-2}. Using Vitek2 and Etest, 100% were resistant to imipenem. In contrast, by Vitek2, 27.8% and 83.3% and by Etest 72.2% and 94.4% were resistant to meropenem and ertapenem, respectively. All isolates were resistant to cefazolin and nitrofurantoin and 87.5% were susceptible to ceftriaxone and cefipime. One isolate was resistant to tigecycline. PFGE showed two clusters with >85% similarity representing 56% of isolates.

CONCLUSIONS: Increasing reports of *S marcescens* harbouring SME in Canada is of concern. Imipenem appears to be the best drug of choice to detect these organisms. Most *S marcescens* harbouring SME-type carbapenemases in Canada are from one of two clonal clusters.

P18

CURRENT OUTPATIENT ANTIBIOTIC THERAPY (OPAT) PRACTICE: SPECTRUM OF PATIENTS, HIGH USE OF CEFTRIAXONE AND ERTAPENEM, AND A POSSIBLE CLINICAL EXPERIENCE EFFECT ON EARLY ORAL STEPDOWN

L SAXINGER[1], L LYNKOWSKI[2]
[1]University of Alberta; [2]Alberta Health Services, Edmonton, AB

OBJECTIVE(S): To describe the patient population, presenting diagnoses, and therapy offered in a Infectious Diseases Specialist OPAT Clinic at a large urban hospital, to derive recommendations for improving the consistency and quality of care.

METHODS: Real time data from each physician-patient encounter "event" between July 1, 2012 and December 31, 2012 was captured, including source of referral, demographics, comorbidities, diagnoses, therapy, surgical procedures and consultations, radiologic procedures and complex nursing care.

RESULTS: There were 1167 physician patient encounters in the OPAT clinic between July 1, 2012 and December 31, 2012, and average of 47 per week, with 13 ID physicians (range 7 to 202 patient encounters.) The emergency department was the source of 87.6% of new patient visits. New patient visit (n=779) diagnoses included 39.7% cellulitis, traumatic wound infection, bites and abscess (=SSTI), 9.9% odontogenic/ENT-peritonsillar (=Dent-ENT), 3.2% urinary tract, 2.6% tenosynovitis, osteomyelitis, and bursitis (=MSK). Overall 11.5% of patienthad early oral stepdown (EOS,

first visit) whith rates correlating to physician years in practice (>10 yearsin practice, n=6, 11.8% to 29.5%, mean 23.25%; < 10 year practice n=7, 0% to 29.2%, mean 9.23%). Forty percent of EOS patients had cellulitis, 19.5% had dental infections and 7.6% had UTIs. Sixty per cent of IV patient were switched off of their referral IV therapy, of which 26.4% received once daily therapy with ceftriaxone or ertapenem. Ceftriaxone was used in 48.3%, ertapenem in 29.1%, cefazolin in 5.8%, and vancomycin in 5.6%. One hundred and one patients received ertapenem at the first visit, with initial diagnoses of 26% cellulitis and 19% other SSTI, 27% Dent-ENT, 15% UTI/other. Length of treatment of initial Dx 'cellulitis' cases given ceftriaxone (65.5%) versus cefazolin (12.1%) will be presented, as well as trends in local cefazolin and ceftriaxone resistance rates in common pathogens.

CONCLUSIONS: Review of OPAT clinic practice reveals a need to review use ertapenem in 'cellulitis' diagnoses, widespread once daily ceftriaxone use, and a possible 'practitioner experience' effect in provision of early oral stepdown therapy.

P19

BACTERICIDAL ANTIBODY AGAINST *HAEMOPHILUS INFLUENZAE* TYPE A IN NORTHWESTERN ONTARIO ABORIGINAL POPULATION

EB NIX, WG MCCREADY, M ULANOVA

Northern Ontario School of Medicine, Thunder Bay, ON

OBJECTIVE: In the post-*Haemophilus influenzae* type b (Hib) vaccine era, non-type b *H influenzae* emerged as a significant cause of severe invasive disease. We have recently reported an unexpectedly high incidence of invasive *H influenzae* type a (Hia) disease in Aboriginal communities of Northwestern Ontario, affecting both young children and adults with significant underlying medical conditions. Similar to Hib, Hia can be carried by healthy individuals in the nasopharynx, and its carriage may potentially induce protective immunity against the pathogen. Based on similar characteristics of Hib and Hia bacteria, and comparable clinical presentations of both infections, anti-capsular polysaccharide antibodies mediating complement-dependent bacteriolysis are considered the major defense mechanism against Hia infection. In an attempt to understand the reasons for an increased susceptibility of Aboriginal people to invasive Hia disease, we studied natural immunity against this pathogen.

METHODS: Bactericidal antibody levels against Hia were studied using the serum bactericidal assay (SBA) in 61 healthy self-identified First Nations adults and 42 Caucasian individuals of similar age residing in Northwestern Ontario.

RESULTS: Serum bactericidal activity was significantly higher in First Nations compared to Caucasian individuals, ie, geometrical mean titers 376.7 (95% CI 233.1 to 608.9) versus 106.8 (95% CI 55 to 207.2), respectively (p=0.0004).

CONCLUSION: These findings suggest that Hia circulates in First Nations communities and exposure to the pathogen can induce protective immunity in the majority of the population. Lack of anti-Hia capsular polysaccharide antibody in children younger than two years of age and in adults with some immunocompromising conditions is the likely reason for cases of invasive Hia disease in this population.

P20

EXPERIENCE WITH SCREENING STOOL SPECIMENS FROM PEDIATRIC PATIENTS WITH COLOREX STEC MEDIUM IN HALIFAX

D HALDANE[1,2], E GILBERT[1,3], T MAILMAN[1,2,3]

[1]Provincial Public Health Laboratory Network of Nova Scotia; [2]Dalhousie University; [3]IWK Health Centre; Halifax, NS

OBJECTIVE: Shiga toxin producing *E coli* (STEC) sporadic isolates have been uncommonly reported in Nova Scotia. As the clinical presentation of non O157-STEC illness differs from than in *E coli* O157 infection, appropriate diagnostic testing may not be requested.

METHODS: Culture surveillance was used to determine if STEC isolates were not being detected. From May 28, 2012 to October 31, 2012 all stools received at the IWK Health Centre were cultured on MacConkey agar. The following day up to five to six colonies that were "typical for *E coli*" (flat, lactose positive) were subcultured to a 1/12 quadrant of STEC Medium (Colorex STEC MX097B, Alere Canada, Canada). After overnight aerobic incubation at 35°C, any isolates that gave mauve growth was tested for Shiga toxin (ST) production using the ST EIA (Premier EHEC , Meridian Diagnostics, USA). Isolates were grown overnight in MacConkey broth and the supernatant was used for testing. Isolates that tested positive were referred to NML for further confirmation and characterization.

RESULTS: Of 180 stool specimens that were screened, 96 gave mauve colonies at 24 hours and were evaluated for ST production. One gave a positive EIA result but did not confirm as a verotoxin producer. A further 28 specimens gave mauve colonies at 48 hours, of which four were tested by EIA and were negative.

CONCLUSION: Although the STEC medium does not detect all the serotypes reported to produce ST, it is designed to detect the most common. Our data suggest that non O157 STEC strains are uncommon in a pediatric population in Halifax in Nova Scotia. Colorex STEC medium had a specificity of 46.7% at 24 h, and 69% of specimens gave mauve colonies at 48 h.

P21

STABILITY OF CLINICAL STOOL SAMPLES AT 4°C AND ROOM TEMPERATURE FOR *CLOSTRIDIUM DIFFICILE* TESTS INCLUDING GD ANTIGEN, TOXIN A/B ANTIGEN, CPE AND CULTURE

M ALFA[1,2,3], N OLSON[2], P DEGAGNE[1], S HOBAN[1]

[1]Diagnostic Services of Manitoba; [2]St. Boniface Research Centre; [3]University of Manitoba, Winnipeg, MB

OBJECTIVE: Transport of specimens from rural regions often suffers from long delays that may affect diagnostic test results. Detection of *C.difficile* toxin in stool can be assessed by antigen detection, CPE, PCR or culture. The aim of this study was to determine if clinical stool samples could be held at 4°C and Room temperature (RT) for 72 h without adverse affects on diagnostic test results (GD antigen, Toxin A/B antigen, CPE assay and culture).

METHODS: Clinical stool samples that were positive for GD and Toxin A/B antigen were included in the study if they were less than 24 h old and were > 3 mL. Aliquots of these stool samples were placed into sterile tubes and stored at either 4°C or RT. After 24 h, 48 h and 72 h, each aliquot was tested by GD antigen, Toxin A/B antigen, CPE assay and alcohol shock culture using CDMN media. Clinical samples from ten different patients were included in this evaluation.

RESULTS: At 4°C, as well as RT, 100% of the clinical stool samples were stable for 72 h after collection for GD antigen and culture testing. However, the Toxin A/B antigen test showed 90% and 70% of the stool samples were stable for 72 h at 4°C and RT, respectively. At 4°C and RT, 100% and 90% of clinical stool samples were stable for 72 h for the CPE test.

CONCLUSIONS: Our data indicate that when stored at 4°C, clinical stool samples will remain test positive for *C.difficile* GD antigen, Toxin A/B antigen, CPE and culture for up to 48 h after collection. Toxin A/B antigen and CPE are the least stable test as all other tests remain positive at 4°C or RT for up to 72 h.

P22

COMPARISON OF ALERE™ PBP2A TEST AND OXOID PBP2 LATEX AGGLUTINATION TEST FOR THE DETECTION OF METHICILLIN RESISTANCE IN *STAPHYLOCOCCUS* SPP

K PUREWAL, S SMITH, A BELHAJ, H ALMOHRI

LifeLabs Medical Laboratory Services, Toronto, ON

OBJECTIVE: To compare Alere PBP2a Test and Oxoid PBP2 Latex Agglutination for detection and confirmation of Methicillin resistance (by mechanism of PBP2a production) in *Staphylococcus* spp.

METHOD: Alere PBP2a Test is an immunochromatographic membrane assay using monoclonal antibodies to detect PBP2a in *Staphylococcus aureus*. It is not approved for use in coagulase negative *Staphylococci* spp.

Oxoid PBP2 Latex Agglutination test is an assay which detects PBP2a from bacterial isolates. Both tests were performed following manufacturers' instructions. Ten MRSA, 14 MSSA, two Cryptic MRSA (defined as oxacillin MIC of 1 to 2, but carrying mec A gene) and one BORSA (characterized by β-lactamase hyperproduction with oxacillin MIC of ≥4, but negative for mec A gene) and six Coagulase Negative *Stapylococcus* CNS were tested, three susceptible to oxacillin and three resistant. Testing of CNS was performed before and after induction using cefoxitin discs. Comparison was done for ease of use, time required to perform the test and accuracy.

RESULTS: There was 100% agreement between the two methods for confirmation of MSSA and MRSA. The two isolates of cryptic MRSA were positive for PBP2a using both tests. BORSA isolate was negative for PBP2a using both tests. For Methicillin resistant CNS, results were negative before induction and positive after induction with cefoxitin disc using both tests. The Alere method is faster and easier to perform. Results were ready in 6 min and no centrifugation or incubation at 96°C to 100°C was required.

CONCLUSION: Alere and Oxoid PBP2a tests performed equally in detection of MRSA. The Alere test was faster and easier to use. Both tests can be used for confirmation of resistance through PBP2 production in CNS and for confirmation of cryptic MRSA.

P23

EVALUATION OF A RAPID EXTENDED SPECTRUM BETA-LACTAMASES/ CARBAPENEMASES (ESBL/CRO) DETECTION KIT IN A DIAGNOSTIC MICROBIOLOGY LABORATORY

I SEKIROV, D ROSCOE

Department of Pathology and Laboratory Medicine, University of British Columbia, Vancouver, BC

OBJECTIVE: Timely and accurate detection of *Enterobacteriaceae* with ESBL/ CRO is essential to facilitate patient treatment and infection control.We evaluated β LACTA™ (Bio-Rad) kit aimed at rapid detection of these resistance mechanisms.

METHODS: ESBL/CRO detection by β LACTA™ kit occurs through hydrolysis of a chromogenic substrate, which changes from yellow to red within 2 min to 15 min of isolate suspension. A change to orange is non-interpretable (non-I). A retrospective panel (RP) of 74 previously well-characterized *Enterobacteriaceae* isolates (42 ESBL/CRO producing and 32 nonproducing) were tested, followed by testing of a prospective panel (PP) of 166 fresh clinical isolates (simultaneous with routine identification and susceptibility testing using the BD Phoenix). The test was performed according to manufacturer's instructions. Briefly, a fresh well-isolated colony from a blood agar plate was suspended in the reagents, which were examined for color change. The β LACTA™ test was performed by one person blinded to phenotypic and molecular results.

RESULTS: For the RP, the overall sensitivity (Sn) and specificity (Sp) was 88% and 100%, respectively. Excluding non-I isolates, the Sn improved to 90%, while the Sp was unchanged. For the PP, the overall Sn and Sp was 77% and 97%, respectively. Excluding the non-I isolates, the Sn improved to 81% and Sp to 100%. All positive results were apparent within 5 min of isolate suspension in the reagents. CMY-2 type AmpC was not detected by the β LACTA™ kit.

CONCLUSIONS: This user-friendly kit is a rapid and highly specific test of ESBL/CRO production by *Enterobacteriaceae* that has potential use in clinical laboratories.

P24

COMPARISON OF DIFFERENT MIC METHODOLOGY FOR *S PNEUMONIA* AND THE VIRIDANS GROUP OF STREPTOCOCCI USING TREK SENSITITRE SYSTEM ®, MICROSCAN PANEL, E-TEST AND M.I.C. EVALUATOR STRIPS

H ALMOHRI, S SMITH, J LAU, L VAN ZUILEN

LifeLabs Medical Laboratory Services, Toronto, ON

OBJECTIVE: To determine essential (EA) and categorical agreement (CA) between TREK sensititre and Microscan for Penicillin PN, Ceftriaxone CTX and Meropenem MPN in *S pneumonia* and *S viridans*, and to compare E-Test and MICE to TREK Sensititre.

METHOD: For TREK Sensititre; STP6F MIC plate was used, for Microscan the MICroSTREP plus® panel for *Streptococcus* species was used. For E-test and MICE strips, isolates were cultured on MH Agar with Sheep Blood. Incubation and reading of results were according to manufacturers' instructions. CLSI Breakpoint interpretations were used (NM: nonmeningeal, M: meningeal, O: oral). Acceptable performance rates are: EA and CA ≥90%, VME and ME ≤3%.

RESULTS: EA was 96.9% to 100% and CA was 93.7% to 100% between Microscan and TREK sensititre for PN, CTX and MPN testing in *S pneumonia*, and 95.4% to 100% in *S viridans* for PN and CTX.

	PN				CTX			
	S.pneumonia		S.viridans		S.pneumonia		S.viridans	
TREK	MICE	E-test	MICE	E-Test	MICE	E-Test	MICE	E-Test
EA	87.0%	81.8%	92.86%	93.8%	81.2%	66.7%	90.9%	90.9%
	NM 69.6%	NM 81.8%						
	M 95.65%	M 96.97%			NM 65.62%	NM 61.1%		
CA	O 95.65%	O 84.8%	92.86%	87.5%	M 62.50%	M 58.3%	90.9%	90.9%
VME	4.34%	3.03%	0	0	12.5%	27.78%	9.1%	9.1%

CONCLUSION: The agreement between TREK and Microscan is acceptable for *S pneumonia* and *S viridans*. For *S pneumonia*; MICE strips and E-Test are not acceptable for Penicillin and Ceftriaxone susceptibility testing.

P25

EVALUATION OF BIOMÉRIEUX CHROMID VRE (CIDV) VERSION 43004 AGAR USING A COMPREHENSIVE COLLECTION OF WELL-CHARACTERIZED RETROSPECTIVE CLINICAL ISOLATES

BM WILLEY[1], N PRAYITNO[2], A MAZZULLI[1], A TSANG[2], M NG[1], P LO[1], K WONG[1], SM POUTANEN[1,2]

[1]Mount Sinai Hospital/University Health Network; [2]University of Toronto, Toronto, ON

OBJECTIVES: Previous studies found that cIDv version 43002 was very specific (Sp) but lacked sensitivity (Sn) for certain enterococci (Ent) with low expression of acquired vancomycin (V) resistance (R) due to the *vanB* genotype. This study evaluated the updated formulation, 43004, to determine if this had been rectified.

METHODS: The 447 test isolates included 199 *E faecium* (Efe; 55 V-susceptible (S), 144 VR: *vanA* 100 including 14 with *vanR/S* deletions, *vanB* 42, *vanD* 2) and 134 *E faecalis* (Efc; 64 VS, 70 VR: *vanA* 11, *vanB* 45, *vanE* 12, *vanL* two), 24 *E gallinarum*, 13 *E. casseliflavus*, four other *Ent* spp, 20 coagulase-negative staphylococci (CNS) and 53 mixed spp. with intrinsic VR [21 Gram-positive (gpb) and 18 Gram-negative bacilli (gnb), 14 *Candida* (yst)]. Isolates recovered from −80°C were streaked to three lots of cIDv using 10 µL of 0.5 McFarland suspensions for a total of 1341 tests. cIDv plates were read after 24 h and 48 h at 37°C in the dark for purple or blue/green colonies associated with VR Efe and Efc, respectively.

RESULTS: At 24 h/48 h, the three lots of cIDv combined grew 391/391 of 432 VR Efe and 160/167 of 210 VR Efc. Overall Sn at 24 h/48 h were: all VRE (85.8/86.9%); all *vanA* and *vanB* (89.3/91.2%); *vanB* (95.4/97.7%); typical *vanA* (100% by 24 h); *vanA* Efe with *vanR/S* deletions (2.4% by 48 h); *vanD* (100% by 24 h); *vanE* (13.9% by 48 h); and *vanL* (0% by 48 h). 18/19 of 54 gnb, 0/1 of 63 gpb, 0/10 of 42 yst, and no CNS or VS Ent grew at 24 h/48 h. Overall Sp were 97.4%/94.8% at 24 h/48 h, respectively.

CONCLUSIONS: The updated cIDv version 43004 remains highly Sp and detected 100% of typical *vanA* at 24 h and 97.7% of *vanB* by 48 h, an improvement from cIDv version 43002. However, this medium detected only a small proportion of epidemic and important *vanA* Efe with *vanS/R* deletions.

P26

COMPARISON OF A NOVEL CHROMOGENIC GROUP B STREPTOCOCCUS (GBS) MEDIA WITH TWO OTHER CHROMOGENIC GBS MEDIA AND AN ENHANCED BLOOD PLATE FOR THE PURPOSE OF GBS DETECTION FROM BROTH ENRICHED VAGINAL/RECTAL SAMPLES

P KORNHERR, L NGUYEN, R PIZARRO, R BEAUDRY, L ZHU
Gamma-Dynacare Medical Laboratories, Ottawa, ON

OBJECTIVES: The CDC has recommended that laboratories maximize recovery of GBS, including atypical strains, by using a broth sub-cultured to a Blood agar or chromogenic plate. A new chromogenic plate (Oxoid Brilliance GBS) BRI (requiring a read only at 24 h) was compared in its performance to the existing chromogenic Bio-Rad Strep B Select SEL, bioMerieux ChromID Strepto B CID agars and an Oxoid blood agar BAP with an added 10 µgm gentamicin disc.

METHOD: Two hundred seventy-five prenatal vaginal/rectal samples received for GBS detection were inoculated into an Oxoid LIM broth and incubated for 18 h to 24 h. BRI, CID, SEL and BAP were then inoculated in a randomized order. To each BAP 2nd quadrant, a 10 µgm gentamicin disc was added. All plates were incubated for 24 h and read for GBS colonies (BRI, CID – pink; SEL – turquoise; BAP – typical grey, hemolytic and growth up to gent disc). CID, SEL were reincubated for additional 24 h as per manufacturer. Confirmation was via PathoDx B latex agglutination. Catalase, gram, PYR, CAMP and Vitek GP used as necessary. A positive GBS was defined as agglutination etc positive from any one or combination of positive plates.

RESULTS: 70 samples recovered GBS (26%). BRI detected all 70; SEL:67; CID:65; BAP:68 with respective sensitivities of 100%, 96%, 93% and 97%. Specificity was BRI: 86%; SEL:89%; CID: 98%; BAP: 95%. Three samples had GBS and Gp G or C Streptococci present. Gp C+G are indistinguishable from GBS on SEL and CID.

CONCLUSIONS: Gp G, C Streptococci did not grow on BRI. Genta disc can differentiate Enterococci from nonhemolytic GBS. 48 h did not increase the yield of GBS from CID, SEL. Colony size was BRI>SEL>BAP>CID. Breakthrough growth was BRI<SEL=CID<BAP. All grew nonhemolytic GBS well.

P27

SELECTION OF VANCOMYCIN RESISTANCE FROM *VANA*-CONTAINING *ENTEROCOCCUS FAECIUM* SUSCEPTIBLE TO VANCOMYCIN

A LOUGHBOROUGH[1], M KISSOON[1], BM WILLEY[1], S PATEL[2,3], A MCGEER[1,2], DE LOW[1,2], SM POUTANEN[1,2]
[1]Mount Sinai Hospital/University Health Network; [2]University of Toronto; [3]Public Health Ontario, Toronto, ON

OBJECTIVE: *vanA*-containing *Enterococcus faecium* susceptible to vancomycin has been described in Ontario and Quebec, Canada related to deletions of *vanR* and *vanS* genes from Tn1546. All such isolates identified in a Toronto laboratory were exposed to vancomycin to see if vancomycin resistance could be expressed.

METHODS: All *vanA*-positive *E faecium* susceptible to vancomycin detected from rectal swabs from March 2011 through February 2012 were included. PFGE was completed using SmaI. Twenty-one isolates representing the major clonal types were tested further. Two McFarland standards of each was swabbed onto 8 mg/L vancomycin gradient agar and incubated at 37°C for 48 h. Growth was subcultured onto gradient agar a total of 18 times. Vancomycin MICs were done by Etest using two McFarland standards read at 48 h. Sustainability of vancomycin resistance was determined by passing resistant isolates onto sheep blood agar without vancomycin.

RESULTS: Nine of the 21 (43%; 95% CI 24% to 63%) vancomycin-susceptible *vanA*-positive *E faecium* expressed *vanA* after passes onto 8 mg/L vancomycin gradient agar. Four of 21 isolates (19.0%) fully expressed resistance (vancomycin MIC >256 mg/L) by the third pass, while 3/21 (14.3%) did not express full resistance until the seventh or eighth pass. *vanA* was only partially expressed in 2/21 isolates (MIC = 8 mg/L). Six of the seven fully resistant isolates were tested for sustainability and all but one remained resistant after five passes onto plain sheep blood agar.

CONCLUSIONS: 43% of *vanA*-containing *E faecium* originally susceptible to vancomycin were readily selected to express vancomycin resistance. Vancomycin should be used with caution for infections due to such isolates and infection control strategies consistent with those used for VRE should be strongly considered.

P28

USE OF URISWAB IN A COMMUNITY HOSPITAL SETTING

R RENNIE, E HUDYM, S BLONSKI
Banff Mineral Springs Hospital, Covenant Health, Banff, AB

BACKGROUND: This study was designed to compare results of urine culture with Dip N Count (DNC, Starplex) and Uriswab (Copan Innovation, Alere, Canada) for patients in Banff, Alberta presenting to family physicians or the hospital ER.

METHODS: Urines were collected in a sterile container and were then poured onto the DNC. At the same time the sponge containing preservative on the Uriswab stick was dipped into the urine. Both samples were then transported to the laboratory. The Uriswab was centrifuged to remove the urine from the sponge and planted onto BAP and Mac Conkey agar. Both samples were incubated at 35 C for 18 h. Colony counts were performed and pathogens were isolated and identified.

RESULTS: Seventy urine samples were tested by both processing methods. Thirty six percent of DNC samples and 40% of Uriswab samples did not grow. Thirty one percent and 20% of DNC and Uriswab samples respectively were considered to have only contaminants. Of those samples from which pathogens were isolated at either 10×10^6/L or 100×10^6/L, *E coli* was isolated as the sole pathogen from 18 DNC and 17 Uriswabs. Enterococci were isolated from three DNCs and six Uriswabs, and *E cloacae* was isolated once from the same sample by both methods. *E coli* and enterococci in significant numbers were recovered from two Uriswabs but only one DNC. A non-lactose fermenter (NLF), *S agalactiae* and a NLF and *E coli* were recovered only from Uriswabs. Overall, pathogens at significant colony counts were recovered from 23 (33%) of DNCs and 29 (41%) of Uriswabs.

CONCLUSION: These data are consistent with previous studies on the enhanced recovery of pathogens from Uriswab with fewer insignificant contaminants. Uriswab in this setting has provided similar data observed in the crossover studies.

P29

EVALUATION OF ALFRED 60/HB&L – URINE SCREENING FOR CULTURE

M MCCONNELL[1], C LEE[1,2], C MAIN[1,2], M SMIEJA[1,2], D YAMAMURA[1,2]
[1]Hamilton Health Sciences, Hamilton Regional Medical Laboratory Program – Microbiology; [2]Department of Pathology and Molecular Medicine, McMaster University, Hamilton, ON

OBJECTIVE: The Alfred 60/HB&L (Alfred) is a fully automated system able to perform bacterial screening of urine for culture. This novel technology is based on light scattering and detection of live, replicating bacteria. Our study objective was to compare the Alfred to the Clinitek Advantus Multistix 8 SG (Multistix) as a screening method to determine the need for urine culture.

METHODS: Consecutive urine specimens from 1069 inpatients, including paediatric, adult, pregnant and immunocompromised patients, were evaluated. Automated screening of urine was performed on the Alfred using an incubation time of 4 h and 25 min and a detection limit of 250 CFU/mL. Multistix and Alfred results were compared to those obtained by conventional culture using 5% Sheep Blood Agar (SBA) and MacConkey Agar in O_2 at 35 °C for 18 h to 24 h.

RESULTS: Culture of the 1069 urine specimens collected yielded 247 with clinically significant positive cultures. Using culture as the reference standard, the sensitivity and specificity for the Multistix test was 95.0% and 23.3% and for the Alfred was 86.2% and 75.9% respectively. The Alfred had significantly improved specificity as compared to the Multistix method. The Alfred detected clinically significant enteric and afermentative gram-negative bacilli, yeast, and a variety of gram-positives (β-hemolytic streptococci, enterococci, staphylococci, and *Aerococcus*

species) within 3 h and provided an automated colony count. Negative urine samples can be reported in >5 h.

CONCLUSION: The Alfred has demonstrated increased specificity while maintaining an acceptable level of sensitivity for screening urine for culture. Overall, this novel screening method results in far fewer urine specimens requiring culture and offers an increased level of automation for the detection of pathogens causing urinary tract infections.

P30

EFFECTIVENESS OF LIQUEFYING RESPIRATORY (RESP) SPECIMENS PRIOR TO BACTERIAL CULTURE USING COPAN SLSOLUTION (SLS) AND EMD MILLIPORE SPUTOLYSIN® REAGENT (SPR)

P LO[1], C BEZAIRE[1], B MUSTACHI[1], G SMALL[1], BM WILLEY[1], SM POUTANEN[1,2]

[1]Mount Sinai Hospital/University Health Network; [2]University of Toronto, Toronto, ON

OBJECTIVES: This study evaluated the effectiveness of two sputum liquefying agents, SLS and SPR, for use in bacterial culture of clinical RESP specimens with the secondary aim to determine the effects of the reagents on common RESP pathogens.

METHODS: For testing the viability of bacteria when treated with the liquefiers, standardized suspensions (10^4 cfu/mL and 10^6 cfu/mL) of *H influenzae, M catarrhalis, S aureus, S pneumoniae, P aeruginosa* and *E coli* were prepared in a) normal saline, b) SLS and c) SPR, after which the suspensions were allowed to incubate as per manufacturer's instructions. All the tubes were then planted to appropriate culture media using the COPAN Walk Away Specimen Processor (WASP) system. In addition, 20 liquid or purulent RESP samples were randomly selected to determine the effect on yield when specimens were treated with the liquefiers as compared to untreated specimens. Again specimens were processed as per manufacturer's instructions. Colony counts (CC) were done on all cultures.

RESULTS: The bacterial cultures yielded similar CC from saline, SLS and SPR. For the RESP specimens, the CC from liquid samples were equivalent for treated and untreated cultures, while for purulent samples, there was an increased CC among those treated with SLS and SPR compared to untreated samples, in some cases yielding two to three times more colonies in the treated versus the untreated samples.

CONCLUSIONS: Both sputum liquefying agents resulted in enhanced organism recovery, especially in purulent RESP specimens, without compromising the yield of common RESP pathogens. The SLS reagent does not require preparation nor is centrifugation required after treatment and therefore is more easily utilized.

P31

A NOVEL RAPID REAL TIME PCR PANEL FOR THE DETECTION OF PEDIATRIC RESPIRATORY PATHOGENS

C ELLIS[1,2], A MISIR[1,2], J LANGILL[3], C HUI[1,2], M JABBOUR[1,2,3], N BARROWMAN[3], J BOWES[3], R SLINGER[1,2,3]

[1]University of Ottawa; [2]Children's Hospital of Eastern Ontario; [3]CHEO Research Institute, Ottawa, ON

OBJECTIVE: Rapid detection of the wide range of viruses and bacteria that cause respiratory infection in children is important for both clinical management and infection prevention. We designed and evaluated a multiple uniplex, ready to use, room-temperature stable respiratory panel comprised of 24 TaqMan MGB real-time PCR assays dried-down in wells of 96 well plates.

METHODS: The real-time PCR multitarget plates were used to test pediatric nasopharyngeal aspirate (NPA) samples for 20 different viruses, as well as *Bordetella* spp, *Mycoplasma pneumoniae* and *Chlamydophila pneumoniae*. Results obtained by PCR were compared to those obtained by our hospital's standard methods: DFA (for respiratory syncytial virus, influenza, and metapneumovirus), viral culture, and PCR (for *Bordetella* spp and M *pneumoniae*).

RESULTS: 159 pediatric NPA samples were studied. One or more respiratory pathogens were detected in 133/159 (83.6%) samples with the PCR panel. The combined detection rate of the standard methods was only 94/159 (59.1%). The PCR panel was significantly more sensitive than standard methods ($p < 0.001$). A total of 160 pathogens were detected by the PCR panel vs 95 by standard methods. This difference was predominantly due to PCR detection of rhinoviruses, parainfluenza viruses, bocavirus, and coronaviruses.

CONCLUSION: The multiple uniplex PCR panel was able to identify pathogens in a higher percentage of respiratory samples than standard methods, and identified numerous pathogens not routinely detected by viral culture. Advantages of this method are rapid turnaround time, minimal manual steps, and ease of use.

P32

VALIDATION OF DIRECT INOCULATION OF URINARY PATHOGENS FROM ALERE ORIENTATION AGAR TO VITEK 2 AND PHOENIX IDENTIFICATION PANELS AND TO VITEK MS (MALDITOF) (WITH SUSCEPTIBILITY TESTING)

L TURNBULL, R RENNIE

Alberta Health Services, Edmonton, AB

BACKGROUND: Chromagenic agar offers a faster approach to identification (ID) of pathogens from mixed cultures such as urine cultures. Unfortunately, these pathogens must still be subcultured before being tested on automated ID and susceptibility (AST) systems. The purpose of this study is to validate direct inoculation of urine pathogens from Alere Orientation agar into Vitek2, Phoenix and Vitek MS instruments.

METHODS: One hundred urines known to contain a wide range of pathogens were inoculated onto Orientation agar and incubated at 35°C for 18 h. Colonies of the appropriate chromagens were tested by Phoenix (ID), Vitek MS (ID) and Vitek2 (ID and AST). Results of ID and AST were compared to those obtained by the laboratory testing the original sample. Essential and categorical agreement was calculated for each antimicrobial and percentage correct for identification results.

RESULTS: One hundred percent of ID results were correct. Categorical agreement for all 35 antimicrobials tested was greater than 95% except for cefepime (30% minor errors [me]), imipenem (10% me), piperacillin (20% me), ticarcillin (20% me) and ticarcillin/clavulanate (20% me) and all occurred with *P aeruginosa* (PA). All essential agreements were greater than 95% except imipenem and PA(20%).

CONCLUSION: Direct inoculation of urinary pathogens from Alere Orientation agar for ID using Vitek2, Phoenix and Vitek MS is a reliable method to reduce time and costs of ID of many common organisms. Direct inoculation for AST testing using Vitek2 is a reliable method except with *P.aeruginosa*. The lower than expected EA and CA for this group of organisms may be due to the small number tested (n=10) and further testing of this group is needed.

P33

EVALUATION OF VITEK MS FOR THE IDENTIFICATION OF CLINICAL ISOLATES OF BACTERIA AND YEAST FROM TSHWANE ACADEMIC LABORATORY-NHLS, STEVE BIKO ACADEMIC HOSPITAL PRETORIA

K BABA[1,2], MR LEKALAKALA[1,2], AA HOOSEN[1,2]

[1]Department of Medical Microbiology, University of Pretoria; [2]National Health Laboratory Services TAD Steve Biko Academic Hospital, Pretoria, Gauteng, South Africa

OBJECTIVES: Matrix-Assisted Laser Desorption Ionization-Time of Flight Mass Spectrometry (MALDI-TOF) has emerged in recent time as an important tool in the identification of clinically important pathogens. A broad range of micro-organisms has been identified with MALDI-TOF produces a species specific spectral pattern which allows identification of organisms. This technique is rapid compared to biochemical and cheaper compared to molecular methods. The objective of this study was to determine the accuracy of VITEK MS for the identification of clinical isolates in the TAD-NHLS Microbiology laboratory, Pretoria, South Africa.

METHODS: Routinely isolated organisms as well as stored isolates were identified by VITEK MS as well as Vitek 2 according to the manufacturer's instructions. Discrepant results were reconciled with 16S rRNA sequencing.

RESULTS: A total of 720 isolates from clinical specimen organisms were evaluated over a 10-week study period from July 29, 2011 to October 14, 2011. These included 522 Gram negative organisms, 171 gram positive organisms and 27 yeasts with one anaerobe. There was 100% correlation between VITEK MS and Vitek 2 for identification to species level. A total of 73 stored isolates from the Microbiology Department's library were also evaluated. The VITEK MS correctly identified 93% of the stored isolates. The VITEK MS manual acknowledges the misidentification of Shigella as *E coli* and suggests other means of identification like morphology and serology. *Streptococcus pneumoniae* misidentified by VITEK MS was finally identified as *Streptococcus oralis* by 16S rRNA sequencing in favour of VITEK MS. VITEK MS correctly identified 99.4% of the total of 793 isolates in our laboratory.
CONCLUSION: VITEK MS was rapid and accurate for the identification of organisms in a routine laboratory.

P34
MOUNTING A NATIONAL PROFICIENCY TEST FOR 16S RRNA GENE SEQUENCING

K BERNARD[1,2], AL PACHECO[1]
[1]National Microbiology Laboratory-PHAC; [2]University of Manitoba Dept Med Micro, Winnipeg, MB

OBJECTIVE: 16S rRNA gene sequencing (16S) is widely used to characterize bacteria in Canada, but proficiency testing for this assay is not readily available or can be expensive. To address this, the Special Bacteriology Laboratory was asked by the National Molecular Microbiology Diagnostics User Group (NMG, www.nmgroup.ca) to mount a cost-effective, 16S proficiency test for its members. Results for this exercise are presented.
METHODS: NMG members were electronically polled to see if they wished to participate and interested laboratories paid for costs of shipping. Each received two live isolates to test and options to record results via email or faxed on a paper report. Participants were asked to provide: demographics, no. bases obtained, 'BLAST Scores', identification of test samples/BLAST databases used, technical/interpretation methods used, regions targeted and other relevant comments.
RESULTS AND DISCUSSION: Sixteen of 205 (8%) NMG members responded to the initial poll, with 13 sites ultimately participating: five from hospitals (ON 2, SK 1, MB 1, NB 1), four from PHLs (BC, AB, MB, SK) five from three NML labs: (SB 2, Enterics 2, BADD 1). Selected strains *Corynebacterium kroppenstedtii* (EPT-1) and *Chryseobacterium hominis* (EPT-2) were correctly identified to genus and species by 10/13, with others correctly identifying isolates to genus only. Four of 13 obtained nearly full sequence (>1400 bp) with 9/13 results being based on partial sequence (400 bp to 500 bp, 5' end). Philosophies regarding 'cut off values' to delineate a species varied; the test was cost effective. A future exercise is being planned involving more challenging/difficult to interpret identifications than those posed here, which could be made available to additional Canadian laboratories.

P35
PILOT EVALUATION OF THE CEPHEID XPERT *C DIFFICILE*/*EPI* PCR ASSAY FOR DETECTING THE PRESENCE OF 027/NAP1 IN PREVIOUSLY CHARACTERIZED FROZEN STOOLS

BM WILLEY[1], S WEESE[2], MR MULVEY[3], T DU[3], DA BOYD[3], G BROUKHANSKI[4], P LO[1], AM MCGEER[1,5], SM POUTANEN[1,5]
[1]Mount Sinai Hospital/University Health Network; [2]University of Guelph, Guelph, ON; [3]National Microbiology Laboratory, Winnipeg, MB; [4]Public Health Ontario; [5]University of Toronto, Toronto, ON

OBJECTIVES: As *C difficile* infection due to 027/NAP1 isolates may cause severe disease and large outbreaks, it is useful to know at time of diagnosis whether this epidemic type is present. This study evaluated the ability of the Cepheid Xpert *C difficile/Epi* (CD/Epi) assay to identify 027/NAP1-positive stools.
METHODS: Forty-four frozen stools positive by Xpert *C. difficile* assay for toxin B (TcdB) for which toxigenic culture had been completed were tested. Isolates were typed by one or more of the following methods: ribotyping, PFGE, PCR detection of toxin A (TcdA), toxin B (TcdB), binary toxin (CDT), and sequencing of *tcdC*. Twenty isolates were 027/NAP1, while 24 were TcdB-positive strains other than 027/NAP1. The CD/Epi, which targets TcdB, CDT, and the 027/NAP1-specific *tcdC* gene which has a frameshift deletion at position 117 was done as per Cepheid's instructions.
RESULTS: Nineteen of 20 (95%; 95% CI 74.6% to >99.9%) of 027/NAP1 stools were found by CD/Epi to be positive for TcdB, CDT and TcdCΔ117 and interpreted as presumptive 027/NAP1, while 1/20 (5%) 027/NAP1 stools were repeatedly TcdB-positive but CDT-negative/TcdC-wild type. Twenty-three of 24 (95.8%; 95% CI 78.1% to >99.9%) non-027/NAP1 stools were TcdB+ or presumptive TcdB+ (TcdB-negative as per CD/Epi interpretation but with Ct 36.5 to 39.3) while 1/24 non-027/NAP1 stools was reproducibly TcdB+/CDT+/TcdCΔ117. The one 027/NAP1 false-negative may reflect the assay's limit of detection, given that the TcdB result was associated with reproducibly high Ct values. A possible specimen mix-up or data error as an explanation for the one 027/NAP1 false-positive result has not been ruled out.
CONCLUSIONS: The Cepheid Xpert CD/Epi assay performed well with high sensitivity (95%) and specificity (95.8%) for 027/NAP1 detection.

P36
COMPARISON OF CT/GC DETECTION WITH THE BDPROBETEC™ Q^X AMPLIFIED CT & GC DNA ASSAY ON THE BD VIPER XTR USING RESIDUAL SUREPATH SPECIMENS BEFORE AND AFTER THE DENSITY SEDIMENTATION PROCESS

H ALMOHRI, S SMITH, L WHITE, VM WALLEY
LifeLabs Medical Laboratory Services, Toronto, ON

OBJECTIVE: To evaluate the detection of CT/GC on the BD Viper XTR analyzer, using SurePath samples before and after the density sedimentation process.
METHOD: Samples of residual fluid from SurePath vials (519) and of aliquot fluid post density sedimentation process (518) were tested on the BD Viper XTR for detection of CT, and for GC (521 and 520, respectively). Results were compared to urogenital specimens for the same patient, collected the same day. Stability for up to four weeks at room temperature was assessed. All SurePath vials were processed by the density sedimentation method (BD PrepMate; BD Diagnostics-TriPath). Fluid from SurePath sample is centrifuged, vortexed and stain preservative and density reagents added according to manufacturer's instructions. Following aliquoting, the primary SurePath vials with the residual fluid were sent to the molecular lab. Fluid in the aliquot tubes post sedimentation process was also sent for molecular testing.
RESULTS: For CT, of the 519 specimens in SurePath vials, 71 were positive in both the SurePath and the urogenital specimens; five were negative in the SurePath but positive in urogenital samples; and 443 were negative in both, giving 99% agreement. Of 518 SurePath aliquots: 69 were positive for CT in both specimens; seven were negative in the aliquot but positive in the urogenital specimen; and 442 negative in both, giving an agreement of 98.7%. For GC; out of 521 SurePath vial specimens, two were positive in both the SurePath vial and urogenital specimen, giving an agreement of 100%, and the same result was seen in the SurePath aliquots. Both specimen types were stable for NAAT testing for up to four weeks.
CONCLUSION: Both SurePath residual fluid and SurePath aliquots post density sedimentation are acceptable specimen types for NAAT testing for CT and GC on the BD Viper XTR.

P37
DETECTION OF INFLUENZA (FLU) A & B AND RESPIRATORY SYNCYTIAL VIRUS (RSV) BY AN IN-HOUSE 4-PLEX RT-PCR ASSAY VERSUS THE XTAG RESPIRATORY VIRAL PANEL (XTAG RVP)

K GUNADASA[1], N LOWTHER[1], A MCNABB[1], P TANG[1,2], M KRAJDEN[1,2]
[1]BCCDC PHMRL; [2]University of British Columbia, Vancouver, BC

OBJECTIVE: Rapid and sensitive identification of the viral causes of

influenza-like illness is important for antiviral treatment initiation (influenza) and for cohorting patients to contain virus transmission. Influenza (Flu) A & B and Respiratory Syncytial Virus (RSV) are clinically important adult and childhood viral pathogens. Given the reported reduced sensitivity and high cost of the xTAG Respiratory Viral Panel (xTAG RVP), the Virology lab developed a real time (4-plex) RT-PCR assay to identify Flu A, B and RSV. The purpose of the evaluation was to assess the target specific sensitivity of the 4-plex RT-PCR assay versus the xTAG RVP.

METHODS: Between October 2011 and April 2012, the Virology lab tested 3700 samples by both the xTAG RVP and the 4-plex assays.

RESULTS: The 4-plex was more sensitive than the xTAG RVP detecting 475 (12.8%) Flu A, 122 (3.3%) Flu B, and 353 (9.5%) RSV. In comparison, the xTAG RVP detected 413 (11.4 %), 81 (2.2%) and 274 (7.6%) Flu A, B and RSV, respectively. However, of samples that were negative for Flu A, B or RSV by the xTAG RVP assay, 1157 (32.7%) were positive for at least one other respiratory virus.

CONCLUSIONS: These data confirm that the 4-plex assay was significantly more sensitive for the detection of Flu A, B and RSV, was more rapid and required less labour to perform (4 h vs 7 h) and was substantially cheaper than the xTAG RVP. The 4-plex assay will be used for primary respiratory screening and the xTAG RVP will be used for outbreak and epidemiological surveillance during the 2012/2013 respiratory season. Improvements in xTAG RVP assay sensitivity and costs will be required for broader utilization for routine respiratory virus screening.

P38

FOCUS SIMPLEXA AND SIMPLEXA DIRECT ASSAYS FOR THE DETECTION OF INFLUENZA A, INFLUENZA B, AND RSV

C KERN[1], L SULLIVAN[1], L SHAW[1], T KARNAUCHOW[1,2]

[1]Regional Virology Laboratory, Children's Hospital of Eastern Ontario; [2]University of Ottawa, Ottawa, ON

BACKGROUND: Molecular detection, the gold standard for the diagnosis of influenza and RSV infections, is limited to laboratories with appropriate resources/expertise. Time-to-reporting is impacted by the need for nucleic acid extraction. The SimplexaFluA/B & RSV Direct test (DAD) (Focus Diagnostics; BND Inc., Toronto) is an 8-well triplex realtime (rt) RTPCR assay that uses the proprietary 3M Integrated Cycler and amplification disks that contain and process nasopharyngeal (NP) swab samples, and does not require nucleic acid extraction. A 96-well assay format requiring nucleic acid extraction (SimplexaFluA/B & RSV; 96-NA) is also available. We evaluate the DAD and 96-NA tests plus an off-label direct method for the latter (96-Direct) vs. DFA on NP swabs and aspirates (NPS/As).

METHODS: NPS/As submitted to the Regional Virology Laboratory (RVL) from adult and pediatric patients and tested for FluA/B/RSV by DFA and culture were retrospectively analyzed by DAD, 96-Direct, and 96-NA assays (NPA=148, NPS=151). Nucleic acids were extracted by MagNAPure Compact (Roche Diagnostics, Laval). Discordant results were resolved by retesting by discordant method and testing by RVL rtRTPCRs.

RESULTS: Sensitivity and specificity of Simplexa assays relative to DFA for FluA, FluB, and RSV respectively: DAD (97.5/99.1%, 98.8/100%, and 100/97.7%), 96-NA (97.5/99.1%, 98.8/100%, and 100/92.1%), and 96-Direct (96.3/99.1%, 98.8/100%, and 98.8/96.8%). There were 23 (16 NPA, 7 NPS) discordant results. Using an expanded gold standard (DFA/culture/RVL rtRTPCRs) assay sensitivity and specificity for FluA, FluB, and RSV respectively: DAD (100/100%; 100/100%, and 89.8/100%), 96-NA (100/100%, 100/100%, and 99/99.5%), and 96-Direct (98.8/100%, 100/100%, and 90.8/100%).

CONCLUSIONS: Focus Simplexa assays provide accurate and reliable detection of influenza A, B, and RSV. The DAD and 96-Direct assays were simple to use and obviate the need for nucleic acid extraction. The conventionally-extracted 96-NA assay offered enhanced sensitivity for RSV detection over the direct assays.

P39

COMMUNITY PREVALENCE OF INFLUENZA C IN ALBERTA, CANADA FROM 2008 TO 2012

K PABBARAJU[1], S WONG[1], A WONG[1], J DUNBAR[1], R TELLIER[1,2], K FONSECA[1,2]

[1]Provincial Laboratory For Public Health; [2]Department of Microbiology, Immunology and Infectious Diseases, University of Calgary, Calgary, AB

BACKGROUND: Influenza C (IFVC) can cause both upper and lower respiratory tract disease. Previous studies have predominantly focused on IFVC infections in children and in hospitalized populations. Traditionally, IFVC infections have been underdiagnosed and epidemiological data is limited. A real-time reverse-transcription PCR (rtRT-PCR) assay was developed for the detection of IFVC virus and used for screening community respiratory samples from children and adults. Positive samples were characterized by sequencing the hemagglutinin esterase (HE) and matrix (M) genes.

METHODS: Respiratory samples (n=1568) collected from community patients (age ranging from newborn to 92 years) visiting their family physician with influenza like illness between Sept 1, 2008 and June 30, 2012 were screened for IFVC. Most of the samples (99.62%) were from patients >5 years old. All specimens had tested negative for respiratory viruses detected by the xTAG® Respiratory Viral Panel assay. Nucleic acid extraction was performed using the MagMAX™ automated extractor. The HE and M genes were sequenced using Sanger sequencing.

RESULTS: A total of eight positive samples were detected (0.51%), of which three were from patients less than five years old, one from a 27-year-old and four from patients between the ages of 40 and 53. The detection of positive samples peaked during the months of February and March in 2009 and 2011. Typing by sequencing the HE gene showed that in 2009, only viruses belonging to the Aichi lineage were circulating, whereas in 2011 there was co-circulation of the Aichi and Kanagawa lineages.

CONCLUSIONS: A positive rate of 0.51% was determined for IFVC infections using a large number of community samples from different age groups over a period of four years; positive rates peaked in late winter. Strains from two lineages were shown to co-circulate, which can in theory result in strain replacement and frequent reassortment, thus allowing the virus to persist and spread.

P40

DEVELOPMENT OF A REAL-TIME, REVERSE TRANSCRIPTASE (RT) MULTIPLEX PCR ASSAY FOR THE DETECTION OF ROTAVIRUS AND ADENOVIRUS IN FAECAL SPECIMENS

M MACPHERSON[1], C RUTHERFORD[1], P JAYARATNE[1,2], SE DALE[3]

[1]St Josephs Hospital; [2]McMaster University, Hamilton, ON; [3]ACM Medical Laboratory, Rochester, New York, USA

OBJECTIVE: Rotavirus A and adenovirus are responsible for acute viral gastroenteritis. Although the incidence of disease is lower in industrialized nations than developing countries, they are still implicated in institutional outbreaks making them a significant problem for infection control. This report describes the development of an RT- PCR assay for the detection of rotavirus A and adenovirus which can be performed on faecal samples.

METHODS: Rotavirus positive and negative stools were tested (n=125), in parallel, by the Rotavirus/Adenovirus multiplex RT-PCR assay and RIDAQUICK™ (R-Biopharm, Darmstadt, Germany) Rotavirus EIA assay. Six adenovirus positive stools and nine clone dilutions were tested using a uniplex in-house PCR assay. All 131 stools were tested by xTAG™ Gastrointestinal Pathogens Panel (xTAG GPP [Luminex Corp., Texas]), an FDA approved commercial multiplex PCR.

RESULTS: For rotavirus, RT-PCR detected one more positive than EIA. Analytically, RT-PCR shows a 2 log greater sensitivity over EIA. All 14 stools negative by PCR and positive by EIA were negative by GPP. One sample negative by EIA and positive by PCR confirmed positive by GPP. The differences of CT's for adenovirus, multiplexed with rotavirus, was negligible when compared to the uniplex in-house PCR assay.

CONCLUSION: This assay shows far better performance for rotavirus A detection than EIA. Efficiency was not lost when compared to uniplex PCR for adenovirus. It incorporates an internal control MS2, an RNA

phage that monitors the efficiency of RT-PCR and detects inhibitors in stool specimens. Heat labile UNG destroys carryover contamination due to amplicons, a major concern in PCR technology.

P41

PREVALENCE OF HIV-2 IN HIV POSITIVE ADULTS, PORT HARCOURT, NIGERIA

M SILVERMAN[1], S BABTUNDE[2], T MEYERS[1], D ALLMAN[1], C AKANI[2], S CHINENYE[2], A YARMOSHUK[1], F MARLATT[2], P SANDSTRUM[3], J BROOKS[3], E IGBIGBI[2], O OBUNGE[1]

[1]Dalla Lana School of Public Health, Toronto, ON; [2]University of Port Harcourt, Port Harcourt, Nigeria; [3]Public Health Agency of Canada, Ottawa, ON

BACKGROUND: Approximately 3.1 million people in Nigeria are living with HIV. The prevalence of HIV2 in Nigeria is presently unknown. National guidelines recommend first line ARV therapy with zidovudine/3TC and a NNRTI. As HIV2 is intrinsically resistant to NNRTI's clarification of the prevalence of HIV2 is important. Present national testing algorithms utilize the Determine® HIV1/2 (ABBOTT), Clearview® Stat-pak HIV1/2 (Inverness medical) and Unigold HIV1/2 (Trinity Biotech) all of which do not differentiate HIV1 and HIV2 infection. Port Harcourt has a large number of immigrant oil workers from other West African countries and so may be an entry point of HIV2 into Nigeria.

METHODS: Between March 1 and May 1, 2012, pregnant HIV-positive women attending the PMTCT clinic and non-pregnant HIV-positive adults attending the ARV clinic at a tertiary hospital in Port Harcourt were selected for study participation by random sampling using clinic numbers. HIV1 and HIV2 infections were differentiated using the Bio-Rad Laboratory Multispot HIV-1/HIV-2 Rapid Test, an FDA-approved assay for differentiating HIV1 and HIV2 as well as co-infection.

RESULTS: A total of 206 ARV clinic attendees were screened. One of 206 (0.5%) had HIV2 mono-infection and 1/206 (0.5%) had HIV1/2 co-infection [total HIV2 infected 2/206 (1%)]. In the PMTCT clinic, 1/53 (2%) HIV positive pregnant women had HIV2 mono-infection, and 0/53 had co-infection.

DISCUSSION: Previously unrecognized HIV2-infected patients were found in both the ARV clinic and the PMTCT clinic, although at low prevalence. Confirmation of HIV type prior to starting empiric ARV therapy would be important in this setting.

P42

EVALUATION OF THE SEEGENE ANYPLEX™ MTB/NTM REAL-TIME PCR FOR IDENTIFICATION OF *M. TUBERCULOSIS* (MTB) AND NON TUBERCULOSIS MYCOBACTERIA (NTM) FROM ACID FAST BACILLI (AFB) SMEAR POSITIVE, PROCESSED CLINICAL SPECIMENS

VN CHIRIP[1], M DESJARDINS[1,2]

[1]The Ottawa Hospital; [2]The Ottawa Hospital Research Institute, Ottawa, ON

OBJECTIVE: To evaluate the performance of the Anyplex™ MTB/NTM real time PCR (RTPCR) a multiplex PCR assay, for the detection of MTB and NTM from processed smear positive clinical specimens.

METHOD: 188 AFB smear pos clinical samples (152 sputa, 30 endoscopic, one lung aspirate, five tissues) processed for cultures were tested by RTPCR. Samples were centrifuged, re-suspended in PCR water, sonicated for 15 min and boiled for 30 min at 95°C. 2.5 µL of this extract was amplified on the BioRad CFX96 thermocycler as per manufacturer's instructions.

RESULTS: Of the 188 smear pos specimens tested by RTPCR, 18 (10%) were invalid due to internal control failure. Seventy-five of the 77 (97%) MTB culture pos samples were pos by RTPCR. Rare AFB was seen in both MTB false neg (FN) PCR samples. Thirty-four of the 48 (71%) *M. avium* (MAC) pos cultures were pos by RTPCR. Eleven of 14 FN were from samples with rare or few AFB seen. For the remaining 27 samples, NTM were detected in 0/8 *M. xenopi*, 6/8 *M. fortuitum*, 3/5 *M. kansasii*, 2/2 *M. chelonae*, 2/2 *M. abscessus*, 1/1 *M. bovis*, 0/1 *M. lentiflavum* pos cultures by RTPCR. Eleven of the 18 culture neg samples were from patients with a history of MTB, nine of which were RTPCR pos. For 2/3 culture neg samples from patients with a history of MAC, PCR was neg and three samples from patients with no history of mycobacterial infections were pos for NTM only. The Anyplex™ MTB/NTM PCR identified 123 of the 152 (81%) *Mycobacterium* positive samples.

CONCLUSION: The Anyplex™ MTB/NTM PCR assay is an effective platform for the detection of MTB and NTM from clinical specimens. The Diagnostic accuracy of a direct MTB RT-PCR assay will be improved when combined with the direct MAC RT-PCR on smear positive samples.

P43

CLINICAL VALIDATION OF THE DIAGENODE™ REAL-TIME MTB PCR ASSAY FOR THE DETECTION OF *M TUBERCULOSIS* IS6110 FROM CLINICAL SAMPLES

P LAGACÉ-WIENS[1,2], HJ ADAM[1,2], K NICHOL[1], P DEGAGNÉ[1], A RENDINA[1], M ALFA[1,2]

[1]Diagnostic Services of Manitoba; [2]University of Manitoba, Winnipeg, MB

BACKGROUND: PCR for the detection of *M. tuberculosis* (MTB) from specimens is recommended by the American Thoracic Society for the rapid diagnosis of tuberculosis. Although several commercial assays exist for the detection of MTB, costs may be significant. The purpose of this study was to validate the performance of an analyte-specific reagent kit for the detection of IS6110 on the Rotorgene™ real-time PCR cycler.

METHODS: Specimens were processed according to a routine MTB culture protocol. A total of 219 specimens were selected on the basis of their final mycobacterial culture result and Kinoun stain results. A variety of culture positive, culture negative, smear positive and smear negative specimens were selected for the validations study. Real-time PCR was performed with the Diagenode™ MTB real-time PCR assay on the Rotorgene™ Q cycler according to manufacturer's instructions.

RESULTS: Sixty-one MTB culture positive, Kinyoun smear positive; 38 MTB culture positive, smear negative; 26 non-tuberculous mycobacterium (NTM) culture positive, smear positive; 58 NTM-culture positive, smear negative; 34 culture negative (no mycobacteria); smear negative and two culture negative, smear positive specimens were analysed. Among smear positive specimens, sensitivity was 100% and specificity was 100%. For smear negative specimens, sensitivity was 50% and specificity was 100%. Excluding follow-up samples from patients on treatment, sensitivity for smear negative specimens increased to 78.6%.

CONCLUSION: Sensitivity and specificity of this assay is comparable to other commercial assays available and has a very high sensitivity and specificity. The assay could serve to rule-in or rule-out MTB in smear positive specimens and enhances rapid detection of MTB from smear-negative specimens.

P44

USEFULNESS OF PROBETEC ET DIRECT TB ASSAY (DTB) (PROBETEC, BD DIAGNOSTIC SYSTEM) FOR DIRECT DETECTION OF MYCOBACTERIUM TUBERCULOSIS COMPLEX IN CLINICAL SPECIMENS

A SOMILY

King Saud University, Riyadh, Saudi Arabia

BACKGROUND: Tuberculosis is still a major health problem in third world countries. Current convention diagnostic methods are slow and lacking sensitivity which might affect the patient outcome and disease control. Nowadays there are several molecular techniques available in the market for diagnosis of tuberculosis. In this study we are evaluating the performance of ProbeTec ET Direct TB assay (DTB) (ProbeTec, BD Diagnostic System) in detecting the Mycobacterium Tuberculosis Complex (MTB) directly from clinical specimens.

METHOD: In this prospective study we performed ProbeTec ET Direct TB assay (DTB) (ProbeTec, BD Diagnostic System) on clinical specimens submitted to King Khalid University Hospital, Riyadh, Saudi Arabia mycobacteriology laboratory for Acid Fast Smear (AFB) and mycobacterium culture during the period from June 2009 to December 2011. All received samples are processed according to laboratory policy for AFB smear and mycobacterial culture using both solid (LJ) and liquid (MIGT)

media. Biochemical tests are performed for final identification of mycobacterium tuberculosis complex. We performed molecular testing using ProbeTec ET Direct TB (DTB) assay directly on specimens following manufacture protocol.

RESULT: During the period of the study we received 96 clinical samples. Of these 46 were respiratory samples and 50 samples were non-respiratory which includes sterile body fluid, tissues and pus (27, 20, 30), respectively. The rate AFB smear positivity were 84% for respiratory and 32% for non-respiratory.

ProbeTec ET Direct TB assay (DTB) assay achieved sensitivity and specificity for respiratory samples of 97.4% and 100%, respectively. For non-respiratory, its sensitivity overall was 51% with highest sensitivity for pus and body fluid of 100% and 65% respectively and the lowest for tissues 25%.The high sensitivity of ProbeTec ET Direct TB assay (DTB) assay in case of pus can be explained by small sample size (only three samples tested). The lowest sensitivity in case tissues could be due decrease in the rate AFB smear positivity 25%

CONCLUSION: ProbeTec ET Direct TB assay (DTB) assay has high sensitivity and specificity for smear positive respiratory samples. For non-respiratory samples, it has lower sensitivity especially for smear negative samples and the result should be interpreted with caution.

P45

ANTIFUNGAL SUSCEPTIBILITY OF RESPIRATORY *ASPERGILLUS* ISOLATES FROM CANADIAN HOSPITALS: RESULTS OF THE CANWARD 2012 STUDY

J FULLER[1], S JIWA[1], S SHOKOPLES[1], A BULL[1], R RENNIE[1], H ADAM[2], M BAXTER[2], D HOBAN[2], G ZHANEL[2]

[1]Provlab, University of Alberta, Edmonton, AB; [2]Medical Microbiology and Infectious Diseases, University of Manitoba, Winnipeg, MB

BACKGROUND: CANWARD is an ongoing national surveillance study that assesses pathogens causing infections in patients attending Canadian hospitals, as well as determines the prevalence of antimicrobial resistance in these isolates. Here we present the antifungal susceptibility data for *Aspergillus* isolates collected in 2012.

METHODS: Clinical *Aspergillus* isolates recovered from respiratory specimens at 10 participating medical centres during the 2012 study period were tested against amphotericin B (AMB), itraconazole (ITRA), posaconazole (POSA), voriconazole (VORI) and caspofungin (CASP) by broth microdilution using the CLSI M38-A2 method. Growth endpoints were measured at 24 h of incubation and values above the epidemiological cutoff (ECOFF) were scored as non-wildtype (non-WT).

RESULTS: Of the 569 *Aspergillus* isolates recovered, *A. fumigatus* represented 73% of the population; *A. flavus*, *A. niger*, *A. nidulans* comprised 7%, 6%, and 3%, respectively. *A. fumigatus* isolates were recovered primarily from Medicine inpatients (55%) and Clinic outpatients (31%) from sputa (53%) and bronchoscopy (30%) specimens. Only three isolates of *A. fumigatus* had non-WT VORI MICs (2 to 4 mg/L) and three others had non-WT CASP MECs of 1 mg/L to 2 mg/L. In contrast, 10 *A. flavus* (25%) isolates showed non-WT MICs to VORI.

CONCLUSION: Invasive aspergillosis is often difficult to diagnose and the clinical correlation of in vitro susceptibility testing is not known. ECOFF values have been proposed to facilitate the detection of microbiological resistance. WT isolates of *A. fumigatus* were most prevalent in this study and evidence of non-WT isolates of other species was very limited. These results will serve as an important baseline to monitor temporal changes as national *Aspergillus* surveillance continues.

P46

EVALUATION OF DERMATOPHYTE IDENTIFICATION MEDIUM (DIM) AND DERMATOPHYTE TEST MEDIUM (DTM) FOR THE RAPID DETECTION OF DERMATOPHYTES ON CLINICAL SPECIMENS

B GANDHI, S RICHARDSON

Division of Microbiology Paediatric Laboratory Medicine, The Hospital for Sick Children, Toronto, ON

OBJECTIVE: To evaluate the Dermatophyte Identification Medium (DIM) and modified Dermatophyte Test Medium (DTM) as an alternative to traditional fungal culture medium for detection of dermatophytes from superficial clinical specimens.

METHOD: Where adequate material was available, 96 consecutive clinical specimens were tested by KOH-calcofluor microscopy and culture. Specimens were inoculated onto two routine agar plate media (IMA w/ciprofloxacin and SAB w/ccg) and two test tube media (DIM and DTM) medium and incubated at 30°C aerobically for up to 14 days. *T. mentagrophytes* ATCC 9533 was used as a positive control while *A. flavus* (clinical), *S. aureus* ATCC 25923, *E. coli* ATCC 25922 and *P.aeruginosa* ATCC 27853 served as negative controls. All media were observed three times a week for growth on primary culture plates in addition to colour change (and growth,) from straw yellow-green to purple on DIM slants and from orange -yellow to red on DTM slants

RESULTS: 29/96 (30.2%) KOH-calcofluor (+), 36/96 (37.5%) routine culture (+), 28/96 (29.2%) DIM (+), 22/96 (22.9%) DTM (+), three non-dermatophytes on routine culture (= false positive), zero DIM false (+), five DTM false (+).

CONCLUSIONS: This study demonstrates that the DIM and DTM media look promising despite a lower positivity rate compared to traditional methods. However a better controlled study is warranted whereby inoculation is done by the same person and the sample as divided evenly three ways. DIM was superior to DTM due to greater sensitivity and no growth of non dermatophytes.

STUDENT POSTERS

Room 200B: Poster Viewing: Thursday April 4, 1100 – 1430 and 1835 – 2000 Friday April 5, 1100 – 1430

SP01

POST MALARIA NEUROLOGICAL SYNDROME IN A SIX-YEAR-OLD CHILD

H AL HASHAMI, A BITNUN

University of Toronto, Toronto, ON

BACKGROUND: Post malaria neurological syndrome (PMNS) is a rare complication of malaria characterized by neurological and psychiatric symptoms and signs that occur after clearance of infection. Symptoms typically develop within two months of acute illness.

The **objective** of this case report and English literature review is to describe the first reported case of PMNS in a North American child and summarize the salient features of PMNS in children.

A six-year-old girl with Plasmodium falciparum malaria acquired in Nigeria was successfully treated with a three day course of Malarone. Two days after treatment completion she developed ataxia, tremor and slurred speech. She was afebrile. Blood smears were negative for malaria parasites. Within a week, her symptoms resolved without intervention.

Of the 41 cases of PMNS (including our case) reported in the English language literature only five were children. Three of these five cases involved children living in southeast Asia. Two, including ours, were travel-related. Age ranged from six to 17 years. Onset of symptoms ranged from three to 14 days (median four days) after treatment completion and clearance of malaria parasites. Parasitemia prior to treatment, reported in two of five cases was 9% in one (17 year old) and 0.9% (our case). Blood

smears were negative at the time of PMNS presentation. Fever, confusion and seizures were observed in four of five children. Ataxia was observed in only one of the five children (our case). Complete recovery was noted in all pediatric cases.

CONCLUSION: Post malaria neurological syndrome is a rare and generally benign condition. This syndrome should be considered in individuals developing neurological symptoms after successful treatment of malaria

SP02

NEURODEVELOPMENTAL OUTCOMES IN INFANTS WITH CONFIRMED CONGENITAL SYPHILIS

V VERGHESE[1], J ROBINSON[1,2], A SINGH[1,2], J GRATRIX[2], T GUENETTE[2], S PLITT[1,2], L HENDSON[1,2]

[1]University of Alberta; [2]Alberta Health Services, Edmonton, AB

OBJECTIVES: A resurgence of infectious syphilis in Alberta in 2002 resulted in many infants being exposed in-utero. The absence of published literature on neurodevelopmental outcomes in congenital syphilis prompted us to do this study in babies born to mothers with reactive syphilis serology.

METHODS: Identification of eligible subjects from the provincial STI database followed by chart review of children seen at the Neonatal and Infant Follow-up Clinic at the Glenrose Rehabilitation Hospital, Edmonton.

RESULTS: 20 babies born to pregnant women with reactive syphilis serology between 2005 and 2010 had neurodevelopmental assessment at 18 months of age; of these, nine were diagnosed with confirmed congenital syphilis (CS) at birth. All were assessed to have been adequately treated between birth and 26 days (median age at treatment three days). At 18 months of age any impairment or delay was documented in 4/9 (44%) with CS, including microcephaly (1), visual insufficiency (1), developmental delay (2) and delayed speech (2), compared with 4/11 (36%) without confirmed CS (p=0.6). Mental Developmental Index (MDI) scores on administration of the Bayley Score of Infant Development were lower in children with CS compared to the rest (mean MDI scores 91.1 vs 96.4; p=0.08). Five of the children with CS had a further assessment at 36 months of age, when 4/5 (80%) had impairments that included severe language delay (4), low IQ scores on intelligence testing (3), and attention-deficit hyperactivity disorder and fetal alcohol spectrum disorder in one child.

CONCLUSIONS: Babies with congenital syphilis are at high risk of neurodevelopmental impairment even with adequate treatment at birth.

SP03

CABIN FEVER: A CASE OF BORRELIA HERMSII IN PREGNANCY

H Hussein, A Showler, DHS Tan

University of Toronto, Toronto, ON

Tick-borne relapsing fever (TBRF) should be considered in patients presenting with fever and a history of exposure to rustic housing in endemic areas. Non-specific symptoms including headache, nausea, arthralgias and myalgias typically coincide with febrile episodes. Although thrombocytopenia is common in TBRF, it is rarely associated with clinically important bleeding. Pregnant patients with acute TBRF may experience significant complications, including maternal ARDS, spontaneous abortion, premature labour and neonatal demise. Patients must be monitored for the Jarisch-Herxheimer reaction, which frequently accompanies initial antibiotic treatment and can be life-threatening. We describe a case of TBRF in a pregnant woman who presented to an Ontario hospital after travel to Southern British Columbia.

SP04

A CASE SERIES OF WEST NILE VIRUS NEUROINVASIVE DISEASE IN THE SUMMER OF 2012 IN HAMILTON, ONTARIO, CANADA

L JIAO[1], C MAIN[1,2]

[1]McMaster University; [2]Hamilton Regional Laboratory Medicine Program, Hamilton, ON

OBJECTIVE: West Nile neuroinvasive disease is a severe infectious disease worldwide, which carries a high mortality rate especially in immunocompromised hosts. An increased incidence of infection was noted in Hamilton in the summer of 2012, prompting a retrospective case series of neuroinvasive cases.

METHODS: Patients admitted to Hamilton Health Sciences hospitals in Hamilton, Ontario, with laboratory evidence of West Nile virus infection were reviewed to identify cases of neuroinvasive disease, following ethics approval. Clinical symptoms, radiological imaging, serological diagnosis, treatment and recovery were evaluated in this group.

RESULTS: 5 cases of encephalitis, 1 case meningitis and 1 case meningomyelitis were identified. The patients ranged from 36 to 82 years old and the ratio of male to female cases was is 3:4. Co-morbidities identified include hypertension, atrial fibrillation, diabetes mellitus, osteoporosis and depression. Common non-specific clinical features include fever (100%), nausea and vomiting (57.1%), diarrhoea (42.9%), rash (42.9%) and headache (28.6%). Two patients were admitted to ICU due to respiratory failure and/or decreased level of consciousness. The mortality rate in this report is approximately 14.3%, and 57.1% of the patients had residual symptoms upon discharge and in the follow up.

CONCLUSION: The information contained in this case series may be helpful in aiding clinicians in making a clinical diagnosis of Neuroinvasive West Nile Virus infection.

SP05

THE RELATIONSHIP BETWEEN HCV HEPATITIS RECURRENCE (HCV-R) POST-LIVER TRANSPLANTATION (LT) AND CMV VIREMIA: A RETROSPECTIVE CHART REVIEW

I SEKIROV[1], N AL NAHDI[1,2], P DOYLE[1], E YOSHIDA[1,2]

[1]Department of Pathology and Laboratory Medicine; [2]Department of Gastroenterology, University of British Columbia, Vancouver, BC

OBJECTIVE: End stage liver disease due to chronic HCV infection is the leading cause of LT in our institution. HCV-R in the transplanted graft can decrease the success of LT, and CMV viremia has been linked to severity of HCV-R. We undertook a retrospective chart review of our LT patients to examine HCV-R and associated factors during the first 12 months post-LT.

METHODS: Charts of all LT recipients (178) from 2006 to 2010 were reviewed and 78 patients with LT due to HCV were identified. Rates and timing of HCV-R, presence of CMV viremia (pre- and post- CMV prophylaxis protocol instituted in 2008) and functionality of transplant for 12 months post-LT were examined.

RESULTS: Overall, 28/78 (35.9%) HCV LT patients experienced HCV-R in first 12 months post-LT. 2/78 recurred in the first month, 18/78 at one to six months and 8/78 at six to 12 months post-LT. Nine of 28 (32.1%) HCV-R occurred in patients with CMV viremia and 14/28 (50%) in patients with acute transplant rejection. Six of 28 (21.4%) patients with HCV-R progressed to fibrosing cholestatic hepatitis (FCH), 2/6 in association with CMV viremia. Two patients experienced graft failure unrelated to HCV-R. Four of 78 (5.1%) patients deceased within first year post-LT; two of them experienced HCV-R. The rates of CMV viremia in HCV LT patients were 50% in 2006, 16.7% in 2007, 14.3% in 2008, 0% in 2009 and 8.7% in 2010. The rates of HCV-R were 42.9% (2006), 38.9% (2007), 42.9% (2008), 27.3% (2009) and 26.1% (2010). The differences in HCV-R rates did not reach statistical significance.

CONCLUSIONS: This data sheds light onto the rates of HCV-R, progression to FCH and graft failure for other reasons in our institution. It gives insight into time periods at high risk for HCV-R, as well as some associated risk factors.

SP06
WITHDRAWN

SP07
CAUTIOUS OPTIMISM REGARDING HIV PRE-EXPOSURE PROPHYLAXIS AMONG FRONT-LINE SERVICE PROVIDERS FROM CANADIAN AIDS SERVICE ORGANIZATIONS

H SENN[1], J WILTON[2], M SHARMA[3], S FOWLER[4], DHS TAN[1,3]
[1]Faculty of Medicine, University of Toronto; [2]Canadian AIDS Treatment Exchange; [3]Division of Infectious Diseases, University of Toronto; [4]Hassle Free Clinic; [5]Division of Infectious Diseases, St Michael's Hosptial, Toronto, ON

OBJECTIVE: Oral daily tenofovir/emtricitabine (Truvada®) is approved in the United States for HIV pre-exposure prophylaxis (PrEP) but has generated controversy in the media and within HIV-affected communities. PrEP acceptability among Canadian AIDS service organizations (ASOs) has not previously been assessed.

METHODS: We developed and pilot tested an online survey about PrEP-related knowledge, experience, opinions, and learning needs, and distributed it to service providers at Canadian ASOs through the Canadian AIDS Treatment Information Exchange between January 2012 and May 2012. The primary objective was to determine the level of support for PrEP approval by Health Canada. Multivariable logistic regression was used to identify characteristics associated with such support.

RESULTS: Overall, 160 complete surveys were received from across Canada. Half (49%) of respondents believed that PrEP warranted Health Canada approval. Support for PrEP approval was associated with more years spent working in HIV (adjusted odds ratio, aOR=1.89 per decade, 95% CI 1.10 to 3.25), low baseline familiarity with PrEP (aOR 3.24 [95% CI 1.01 to 10,41]) and knowing someone who had used PrEP (aOR 4.39 [95% CI 1.28 to 15.08]). Overall, respondents were cautiously optimistic about PrEP with 57% agreeing that "PrEP would be an appropriate use of healthcare resources". Many (26%) had been asked about PrEP in the past year, but most participants thought that they (61%) or their organization (63%) did not have enough current knowledge about PrEP.

CONCLUSIONS: Service providers from Canadian ASOs in this study were cautiously supportive of PrEP, but most felt they did not have enough current knowledge. Since community groups play an important role in PrEP counseling, further education is needed to better prepare ASOs for this novel HIV prevention strategy.

SP08
TRANSMISSION OF *BRUCELLA MELITENSIS* TO A HEALTHCARE WORKER OUTSIDE OF THE MICROBIOLOGY LABORATORY

C LOWE[1], A SHOWLER[1], S MCINTYRE[2], R QURESHI[1,2], V ALLEN[1,3], R DEVLIN[1,2], M MULLER[1,2]
[1]University of Toronto; [2]St. Michael's Hospital; [3]Public Health Ontario, Toronto, ON

OBJECTIVES: *Brucella* is the most common laboratory-acquired infection but transmission to non-laboratory healthcare workers has not been reported. We describe transmission of *Brucella melitensis* to a non-laboratory healthcare worker.

METHODS: Synovial fluid was sent to the laboratory from a joint aspiration; *Brucella* was not suspected. Planting and gram staining were performed in a biosafety cabinet but subsequent testing was conducted on the open bench. Identification of *Brucella* prompted an exposure investigation conducted using CDC exposure definitions. High-risk workers were those who directly manipulated the culture or were within 5 feet on the open bench. Low-risk workers included all others in the laboratory at the time. All exposed workers were offered serial serology (2, 4, 6 and 24 weeks). Prophylaxis with doxycycline and rifampin (3 weeks) was recommended for high-risk workers. Although not recommended by the CDC, we defined the healthcare workers that performed the aspiration as high-risk.

RESULTS: Thirty-two healthcare workers were exposed: 11 high-risk (nine laboratory and two radiology) and 21 low-risk. Ten of 11 high-risk workers took prophylactic antibiotics. No seroconversions were detected among laboratory staff. However, a radiology technician seroconverted with titres of 1:160 and 1:320 at 3 and 8 weeks post-exposure, respectively. This technician injected the synovial fluid from a syringe into a container. The technician was initiated on prophylaxis which was extended to six weeks.

CONCLUSION: Transmission of *B. melitensis* can occur in healthcare workers performing aerosolizing procedures outside of the microbiology laboratory. When *Brucella* is identified, exposed non-laboratory healthcare workers should be identified and monitored in the same manner as laboratory workers.

SP09
APPARENT ELEVATED INFECTION RISK ASSOCIATED WITH TITANIUM PLATE STERNOTOMY CLOSURE AFTER CARDIAC SURGERY

M CHARLES, L SAXINGER, S SMITH
University of Alberta, Edmonton, AB

OBJECTIVES: Sternal wound infections are an important cause of morbidity and mortality in patients undergoing cardiac surgery. Wire cerclage is currently the standard for sternotomy closure. However, recent studies have suggested that sternal closure with rigid titanium plates is associated with greater sternal stability, shorter length of stay, and decreased postoperative complications. We report on our institution's preliminary experience with this technique.

METHODS: This is a descriptive retrospective study performed through Infection Prevention and Control at one institution. From August 2010 to August 2012, 1476 CABG procedures were completed and titanium plate fixation was used in 60 patients. In this group, data were collected on the deep and superficial infection rate, known infection risk factors, microbiologic results, length of hospitalization and overall mortality rate.

RESULTS: 48 males and 12 females, with an average age of 65 years, had plate fixation. Primary plate fixation closure was performed on 32 patients, with a sternal wound infection (deep and superficial) rate of 25%. In comparison, the infection rate for all CABG procedures was 3.4%. More detailed risk factor data using a matched cerclage comparator group will be presented. The overall mortality rate was 6.3% in the plate fixation group, and 7.0 % in the cerclage closure group.

CONCLUSION: In this small sample, the use of rigid plates in primary sternal closure appeared to be associated with a 25% infection rate rather than decreased complication rates and better outcomes. Long-term follow up and larger scale studies will be needed to assess the benefits, complications and indication for primary and secondary sternal closure with titanium plate fixation.

SP10
NATIVE VALVE INFECTIVE ENDOCARDITIS DUE TO MICROCOCCUS LUTEUS

A GHOSH, K SHAH, N SINGHAL
McMaster University, Hamilton, ON

OBJECTIVE: To present a case of a rare cause of infectious endocarditis and literature review of endocarditis due to *Micrococcus species*.

METHODS: A search of the MEDLINE database using the Medical Subject Heading terms "micrococcus" and "endocarditis" was conducted to identify previously reported cases. Furthermore, the references of all identified studies were manually searched for additional reports.

RESULTS: A 23-year old male intravenous drug user with type 1 diabetes mellitus presented with a 1-month history of fevers. He was found to have vegetations on the aortic and mitral valves, as well as a left inguinal mycotic aneurysm. He underwent aortic and mitral valve replacement surgery and aneurysm repair. Tissue cultures from the mycotic aneurysm grew *Candida albicans*. Cultures of the aortic valve grew *Micrococcus luteus* and pathological examination of the valve revealed pseudohyphae. A previous blood culture, one month prior had grown *Enterococcus faecalis*, suggesting a polymicrobial endocarditis. He was initially treated with vancomycin, tobramycin and fluconazole and subsequently successfully completed his course with ampicillin and fluconazole.

A review of the seventeen reported cases of endocarditis due to Micrococcus

species reported found that five were due to *Micrococcus luteus*. All of these 17 cases occurred in patients with prosthetic cardiac valves, with the exception of one case of native valve endocarditis due to *Micrococcus luteus* in an immunosuppressed patient.
CONCLUSION: This is, to our knowledge, the first reported case of native valve endocarditis due to *Micrococcus luteus* in an immunocompetent host and the only reported case of a polymicrobial infective endocarditis involving *Micrococcus luteus*.

SP11
LIVER TOXICITY OF VORICONAZOLE AND POSACONAZOLE IN HAEMATOLOGICAL CANCER PATIENTS

N BELZILE[1], S DUFRESNE[1,2], A LABBÉ[1,2], M LUONG[1,2], S BÉLANGER[2], M LAVERDIÈRE[1,2]
[1]Université de Montréal; [2]Hôpital Maisonneuve-Rosemont, Montréal, QC

OBJECTIVES: To compare the liver toxicity of voriconazole and posaconazole in haematological cancer patients.
METHODS: A retrospective chart review was conducted in haematological cancer patients fulfilling the following criteria: (1) admitted for at least one day at Hôpital Maisonneuve-Rosemont between December 2009 to December 2010, (2) received voriconazole or posaconazole for at least 3 consecutive days, (3) had hepatic function monitored with liver function tests (LFT) for a minimum of seven days after the introduction of the azole. Liver toxicity was defined as an elevation of one or many LFT three times the upper limit of normal range. For those who had abnormal values at baseline, it was defined as three times the baseline value.
RESULTS: Fifty-six patients were included, among which 30 were allogeneic hematopoietic stem cell transplant (HSCT) recipients and one was an autologous HSCT recipient. Posaconazole was administered to 32 (57%) patients for 3-207 days (median 17 days), and voriconazole to 24 (43%) for 8-88 days (median 24 days). Twenty-nine patients had abnormal liver enzymes at baseline (14 [25%] with AST>40 and 27 [48%] with ALT>40). Liver toxicity occurred in 16 (29%) patients: 12 (38%) patients treated with posaconazole and four (17%) with voriconazole (p=0.09). Patients on voriconazole were significantly older than patients on posaconazole (median 57 years old vs 46 years old; p=0.049). Liver toxicity was not associated with age; it was more frequent in HSCT recipients (37%) than in non-transplant patients (20%) but the difference was not statistically significant.
CONCLUSION: Although the power of the study was limited, posaconazole does not seem to be less hepatotoxic than voriconazole in hematological cancer patients.

SP12
INVESTIGATING WELL WATER SUBMISSION TRENDS IN SOUTHEASTERN ONTARIO IN 2011

K RANDHAWA[1], J KROLIK[2,3], A MAJURY[3,4]
[1]Department of Community Health & Epidemiology; [2]Department of Pathology & Molecular Medicine, Queen's University; [3]Public Health Ontario; [4]Department of Biomedical & Molecular Sciences, Queen's University, Kingston, ON

OBJECTIVE: Ensuring the safety of drinking water is a public health priority. Drinking water quality management is a crucial component for the prevention and control of waterborne diseases. With over four million Canadians consuming water from private water supplies, which are mostly groundwater wells, it is important to ensure water from private wells is safe for consumption through testing. The objective of this study was to provide a comprehensive, geospatial description of well water submissions, focusing on well water submitters who did not contribute the recommended number of samples for testing in southeastern Ontario, in 2011.
METHODS: Well water data were extracted from the Water Testing Information System and entries were geocoded. Addresses containing more than one bacteriological test result in the given year were considered as multiple submissions. Clustering analyses were conducted to determine the distribution of well water submitters.
RESULTS: Of the 8,134 well water entries within four public health units (PHUs) submitted to Public Health Ontario Laboratory in Kingston, 4698 were single submissions and 3166 were multiple submissions. There were no significant differences in terms of number of submissions per unique address between the four PHUs. Within the four PHUs, 61.08% of well owners submitted one sample and 81.20% of well owners submitted less than the recommended number of annual sample submissions. Cluster analyses revealed no significant clusters of submission trends within the four PHUs.
CONCLUSION: A lack of significant clusters indicates that poor sample submission behaviours are widespread. Findings from this study indicate a need for greater education on the importance of well water testing and increased availability and accessibility to well water testing programs.

SP13
AN EVALUATION OF SOUTHEASTERN ONTARIO BEACH WATER QUALITY

MC BENNELL[1,2], A JOYCE[2], E GARDNER[3], A MAJURY[1,2]
[1]Queen's University; [2]Public Health Ontario; [3]KFL&A Public Health, Kingston, ON

The presence of enteric pathogens in recreational waters is a potential threat to human health. *Escherichia coli* (*E. coli*), the current faecal indicator bacteria for fresh waters in Canada, is an indirect indicator of the possible presence of enteric pathogens. An evaluation was conducted to assess *E. coli* contamination of Southeastern Ontario beach waters through time and by location, and by non-compliance with the Ontario bathing beach standard. Beaches were sampled approximately bi-weekly or more frequently, and *E. coli* were enumerated by a membrane filtration method. The results indicated that the average beach had low levels of contamination, the year 2010 was a more contaminated year than 2011, and monthly *E. coli* contamination was equal across months during the study years. Lake Ontario beaches exhibited higher *E. coli* contamination than inland beaches. Seven beaches showed high non-compliance to the Ontario standard, which could be of concern for the health of recreational beach users in the surrounding area. Comparisons of this study to the broader geographic area were difficult, as similar studies are not prominent in the literature. The authors call for greater beach sampling standardization across jurisdictions, provinces, territories, and across North America. Wider accessibility of beach management data and analyses that are reflective of a single *E. coli* contamination standard are needed to produce accurate comparative studies. Additionally, it is suggested that public health agencies work in collaboration with municipalities and government agencies to establish bird management programs, improved methods for beach grooming, education programs for the public to refrain from feeding birds, and the reconstruction of wetlands to filter agricultural and urban runoff.

SP14
MOLECULAR EPIDEMIOLOGY OF EXTENDED-SPECTRUM β-LACTAMASE-, AMPC β-LACTAMASE-, AND CARBAPENEMASE-PRODUCING *ESCHERICHIA COLI* (EC) AND *KLEBSIELLA PNEUMONIAE* (KP) ISOLATED FROM CANADIAN HOSPITALS: CANWARD 2007 – JUNE 2012

AJ DENISUIK[1], PRS LAGACÉ-WIENS[1,2], PJ SIMNER[3], MR MULVEY[1,4], M BAXTER[1], HJ ADAM[1,2], DJ HOBAN[1,2], GG ZHANEL[1]
[1]University of Manitoba; [2]Diagnostic Services of Manitoba, Winnipeg, MB; [3]Mayo Clinic, Rochester, Minnesota, USA; [4]National Microbiology Laboratory, Winnipeg, MB

OBJECTIVE: To assess the prevalence and molecular characteristics of ESBL-, AmpC-, and carbapenemase-producing EC and KP isolated from Canadian hospitals.
METHODS: 5,842 EC and 1,795 KP were collected from January 2007 to June 2012, as part of the ongoing CANWARD national surveillance study. Antimicrobial susceptibility testing was performed in order to detect putative ESBL-, AmpC-, and carbapenemase-producers. Isolates were characterized by PCR and sequencing to detect resistance genes, and clonal spread was assessed.
RESULTS: The prevalence of ESBL-EC [2007: 3.4%, 2012: 7.2%], AmpC-EC [2007: 0.7%, 2012: 2.6%], and ESBL-KP [2007: 1.5%,

2012: 2.9%] increased significantly during the study period. Antimicrobials demonstrating the greatest activity against ESBL-EC, AmpC-EC, and ESBL-KP in this study were colistin, amikacin, ertapenem, and meropenem. Isolates were generally unrelated by PFGE (<80% similarity); however, ST131 was identified among 55.8% and 28.7% (P<0.001) of ESBL-EC and AmpC-EC, respectively. CTX-M-15 was the dominant genotype in both ESBL-EC (66.2%) and ESBL-KP (50.0%), while the dominant genotype in AmpC-EC was CMY-2 (55.7%). Carbapenemase production was identified in 0.03% (n=2) of EC and 0.11% (n=2) of KP, with KPC-3 representing the dominant genotype.
CONCLUSIONS: The prevalence of ESBL- and AmpC-producing EC and KP increased significantly from 2007 to 2012, while the prevalence of carbapenemase-producers in Canada remained low (<1%). Compared to AmpC-EC, ESBL-EC were significantly associated with multi-drug resistance and the ST131 clone.

SP15

EVIDENCE OF EXTENDED-SPECTRUM BETA-LACTAMASE PRODUCING ORGANISMS EMERGING IN ONTARIO HOSPITALS AND COMMUNITIES

L GOODLIFFE[1,2], P BOERLIN[4], G CHALMERS[4], J VANDERLAAN[2], R SCHERTZBERG[2], W CICCOTELLI[2,3]
[1]University of Toronto, Toronto; [2]Grand River Hospital; [3]St. Mary's General Hospital, Kitchener; [4]University of Guelph, Guelph, ON

OBJECTIVE: Recently, the regional prevalence of extended-spectrum beta-lactamase (ESBL) has risen at an alarming rate, including an increase in serious community infections requiring hospitalization. Our objective was to define the local epidemiology of ESBL-producing *Enterobacteriaceae* in an effort to guide infection control policies of local hospitals.
METHODS: ESBL positive patients were identified by the Regional Microbiology Laboratory using standard methodology. Only the first isolate per patient identified from 1 January 2008 to 30 June 2012 were included and were classified as hospital, community, or other-health care associated. Patient characteristics, ESBL-case counts and rates were analyzed. Forty-six available *Escherichia coli* isolates were typed by pulsed-field gel electrophoresis (PFGE) and screened for major ESBL and carbapenemase genes using standard PCR protocols. Research Ethics Board approval was obtained prior to the study.
RESULTS: ESBL-producing isolates were recovered from 281 unique patients (267 *E. coli*, 11 *Klebsiella pneumonia* and 3 *Proteus mirabalis*). Patients were predominantly female (68.0%) and the average age was 62.4 years. Annual case counts rose steadily from 2008 to 2012. The rate of ESBL-producing E. coli increased from 4.71 to 41.8 per 1000 E. coli (p<0.001) during the study period. There was significant multi-drug resistance among isolates. PFGE showed a large genetic diversity among the isolates tested (≥60% pattern similarity). All but one isolate carried a CTX-M gene. Preliminary results suggest that most isolates belong to the pandemic *E. coli* ST131 lineage.
CONCLUSIONS: There has been a sudden emergence of genetically diverse CTX-M ESBL-producing multidrug resistant *E. coli* in this area over the last 4.5 years. Given the limitations of classifying cases, the exact cause of this is uncertain; future research is required.

SP16

ISOLATION OF VANCOMYCIN HETERORESISTANT *STAPHYLOCOCCUS CAPITIS* IN A CANADIAN NEONATAL INTENSIVE CARE UNIT

G POLIQUIN[1], L MACKENZIE[1], A WALKTY[1,2], HJ ADAM[1,2]
[1]University of Manitoba; [2]Diagnostic Services of Manitoba, Winnipeg, MB

OBJECTIVE: Coagulase-negative *Staphylococcus* is a common cause of sepsis in infants in the neonatal intensive care unit (NICU). We recently encountered a case of persistent *Staphylococcus capitis* bacteremia in an infant despite a seemingly vancomycin-susceptible isolate. On further testing, this isolate was found to be vancomycin heteroresistant by Macro E-test. We sought to determine whether this was a unique case or a broader finding within our NICU.
METHODS: *S. capitis* blood culture isolates covering a five year period from one NICU in Winnipeg, Manitoba (n=16) were compared to blood isolates from non-NICU units at a separate hospital (n=10). Vancomycin susceptibility was determined by the E-test method. Heteroresistance was tested using the Macro E-test method with vancomycin and teicoplanin E-tests on Brain-Heart Infusion agar for a total incubation period of 48 h. Heteroresistance was defined as either 1) both vancomycin and teicoplanin MIC ≥8 µg/mL or 2) teicoplanin MIC of ≥12 µg/mL.
RESULTS: A total of 26 isolates were tested from 22 different patients. All 26 isolates were vancomycin susceptible by standard E-test. The Macro E-tests revealed that 16/16 isolates (100%) from the NICU were positive for heteroresistance whereas none (0/10) of the other isolates were positive. Within the heteroresistant group, 7/12 infants had multiple positive blood cultures (range 2-5) whereas in the non-heteroresistant group, 0/10 patients had multiple positive blood cultures. Pulse-field gel electrophoresis of the heteroresistant isolates revealed two clones.
CONCLUSIONS: Vancomycin heteroresistant *S. capitis* have been described in other areas of the world but not in Canada to the best of our knowledge. The detection of this phenotype in the *S. capitis* from our NICU is of concern as the phenotype may be associated with prolonged periods of bacteremia. Further study is required to elucidate why some infants remain bacteremic and to explore the heteroresistance phenotype.

SP17

MORTALITY OF COMMUNITY-ONSET *STAPHYLOCOCCUS AUREUS* BACTEREMIA: A MULTI-NATIONAL POPULATION-BASED STUDY

S TOM[1], JC GALBRAITH[2,3], L VALIQUETTE[3,4], G JACOBSSON[3,5], P COLLIGNON[3,6], HC SCHØNHEYDER[3,7], M SØGAARD[3,8], KJ KENNEDY[3,6], JD KNUDSEN[3,8], C OSTERGAARD[3,9], O LYYTIKÄINEN[3,10], KB LAUPLAND[3,11]
[1]Department of Medicine, University of Calgary, Calgary, AB; [2]Microbiology Laboratory, Vancouver Island Health Authority, Royal Jubilee Hospital, Victoria, BC; [3]The International Bacteremia Surveillance Collaborative, Calgary, AB; [4]Department of Microbiology-Infectious Diseases, Université de Sherbrooke, Sherbrooke, QC; [5]Department of Infectious Diseases, Skaraborg Hospital, Skövde, Sweden; [6]Department of Infectious Disease and Microbiology, Canberra Hospital and School of Clinical Medicine, Australian National University, Woden, Australia; [7]Department of Clinical Microbiology, Aalborg Hospital, Aarhus University Hospital, Aalborg, Denmark; [8]Department of Clinical Microbiology, Copenhagen University Hospital, Hvidovre Hospital; [9]Department of Clinical Microbiology, Copenhagen University Hospital, Herlev Hospital, Copenhagen, Denmark; [10]Department of Infectious Disease Surveillance and Control, National Institute for Health and Welfare, Helsinki, Finland; [11]Departments of Medicine, Critical Care Medicine, Pathology and Laboratory Medicine, and Community Health Sciences, University of Calgary, Calgary, AB

OBJECTIVE: The epidemiology of *Staphylococcus aureus* bacteremia has been changing worldwide. The objective of this study was to assess secular trends in the mortality associated with community-onset *S. aureus* bacteremia.
METHODS: Population-based surveillance for community-onset *S. aureus* bacteremia was conducted in Canada, Australia, Sweden and Denmark during 2000-2008. Case fatality (deaths per cases) and mortality rates (deaths per 100,000 population) were age- and gender-standardized to the European Union 27-country 2007 population.
RESULTS: A total of 11,058 cases of community-onset *S. aureus* bacteremia were included; 10,624 (96%) were methicillin-sensitive *S. aureus* (MSSA) and 434 (4%) were methicillin-resistant *S. aureus* (MRSA). Although the annual incidence of MRSA did not significantly change during the study period, the incidence of MSSA increased. The overall case fatality was 19% (MSSA 19%, MRSA 22%). Although the MSSA case fatality rates were comparable year-to-year, the MSSA mortality rate (2.86 per 100,000 overall) increased during the study. On the other hand, MRSA case fatality rates decreased progressively but the MRSA mortality rate (overall 0.22 per 100,000) was similar throughout the study.
CONCLUSION: Although the emergence of MRSA has attracted great

concern, community-onset MSSA is associated with a much higher and increasing mortality rate. This study highlights the importance of interpreting case fatality rates in the context of disease incidence and population mortality rates when establishing the burden of an infectious disease.

SP18

STREPTOCOCCUS PNEUMONIAE BLOODSTREAM INFECTION AMONG PATIENTS WITH HEMATOLOGICAL MALIGNANCIES: WHAT IS THE CLINICAL IMPACT IN AN ERA OF ANTI-PNEUMOCOCCAL VACCINATION AND ANTIBIOTIC RESISTANCE?

S BARKATI[1], S BÉLANGER SAVARY[1], F FILION[1], V LAVERGNE[2,1], SH SU[3,1], V LOUNGNARATH[4,5], L VALIQUETTE[6,7], K WEISS[1,8], B LEVEBVRE[9], A LABBÉ[1,8]

[1]Université de Montréal;[2]Hôpital du Sacré-Coeur de Montréal; [3]CHUM-Hôpital Notre-Dame, Montreal; [4]Université Laval; [5]Hôpital Enfant-Jésus, Quebec; [6]Université de Sherbrooke; [7]CHUS-Hôpital Fleurimont, Sherbrooke; [8]Hôpital Maisonneuve-Rosemont, Montreal; [9]Laboratoire de Santé publique du Québec (LSPQ), Sainte-Anne-de-Bellevue, QC

OBJECTIVES: Haematological malignancies are associated with a higher risk of pneumococcal bloodstream infections (BSI) and vaccination remains the cornerstone of prevention. The objectives of this study were to assess the clinical impact of BSI in this population and to investigate serotype distribution and antibiotic susceptibility profiles of *S. pneumoniae* isolates.

METHODS: We conducted a multicenter (three tertiary-care centers in Montreal) retrospective study of all consecutive pneumococcal BSI in adult patients with haematological malignancies between 2003 and 2011. Medical charts were reviewed. *S.pneumoniae* serotypes and antibiotic susceptibility results were provided by the LSPQ.

RESULTS: A total of 78 BSI occurred in 75 patients. Multiple myeloma (42.9%) and non-Hodgkin lymphoma (26.0%) were the most common malignancies. There were 22 hematopoietic stem cell transplant (HSCT) recipients. Pneumonia was the major diagnosis (76.9%). The 30 day-mortality rate was 14.1%. Twenty-eight different serotypes were encountered: 62.8% were included in the pneumococcal polysaccharide vaccine; 24.4%, 30.8% and 44.9% were included in the Pneumococcal conjugate vaccines 7, 10 and 13 respectively. Association with non-vaccine isolates was more often seen in HSCT recipients (50.0% vs 23.2%; $p<0.05$). There was no resistance to parenteral penicillin. Macrolide, TMP-SMX and respiratory fluoroquinolone resistance rates were 29.5%, 17.9% and 1.3%.

CONCLUSION: Pneumococcal BSI in patients with haematological malignancies carries an important mortality rate. Continuous efforts should be made to optimize immune response and vaccination coverage of *S. pneumoniae* particularly in HSCT recipients. Penicillin remains an excellent treatment choice of pneumococcal BSI.

SP19

MICROBIOLOGY OF MRSA IN PEDIATRIC AND ADULT INPATIENTS AT HOSPITALS PARTICIPATING IN THE CANADIAN NOSOCOMIAL INFECTION SURVEILLANCE PROGRAM (CNISP), 1995-2007

T LOCKE[1], C QUACH[2,3], G GOLDING[4], A MOUNCHILI[1,5], L PELUDE[5], D COYLE[1], D GRAVEL[1,5], A SIMOR[6], CNISP Program

[1]University of Ottawa, Ottawa, ON; [2]McGill University; [3]Montreal Children's Hospital, Montreal, QC; [4]National Microbiology Laboratory, Winnipeg, MB; [5]Public Health Agency of Canada, Ottawa; [6]Sunnybrook Research Institute, Toronto, ON

OBJECTIVE: We aimed to compare the microbiology of methicillin-resistant *Staphylococcus aureus* (MRSA) in pediatric and adult inpatients from participating CNISP facilities.

METHODS: We assessed patient/clinical characteristics, molecular laboratory testing, rates, and hospital profiles using data from the CNISP MRSA surveillance programs. Descriptive statistics and non-parametric tests were used to compare adult and pediatric MRSA microbiology. MRSA hospital rates were also compared.

RESULTS: MRSA annual incidence rates were higher in adult hospitals (0.73 to 14.01 [adult] and 0.08 to 3.88 [pediatric] /10,000 patient-days). The major PFGE strains differed between adults (CMRSA2) and children (CMRSA7 and CMRSA10). SCCmec type II (1606, 53%) was more common in adults while SCCmec type IVa (177, 70%) was more common in children. A higher proportion of pediatric isolates were PVL positive (277, 59% vs. 1174, 24%; $p<0.01$). In comparison to isolates from pediatric patients, adult patients' isolates showed significantly higher non-susceptibility to most antibiotics (clindamycin, erythromycin, ciprofloxacin, tetracycline and TMP-SMX; Multi-resistance (non-susceptibility >2 antibiotics): 5659, 94% [adult] vs. 271, 71% [children], $p<0.01$). Healthcare-associated (HA) isolates exhibited higher resistance to the majority of antibiotics compared to community-associated (CA) in both populations. (Multi-resistance: 4031, 96% [HA] vs. 941, 86% [CA], $p<0.01$).

CONCLUSION: The microbiology of MRSA differed significantly in adults and children. This may support a need for different treatment and control strategies in pediatric patients.

SP20

MSSA AND MRSA PEDIATRIC OSTEOMYELITIS AT THE ROYAL UNIVERSITY AND VICTORIA HOSPITALS, SASKATOON AND PRINCE ALBERT, SK, 1996-2010

B KOZUN, A MCCONNELL, A RAMJI

University of Saskatchewan, Saskatoon, SK

OBJECTIVE: A retrospective study was performed to describe the changing epidemiology of, and differences in clinical features, laboratory investigations and outcomes in pediatric MSSA vs MRSA osteomyelitis at the Royal University Hospital (RUH) in Saskatoon, SK and Victoria Hospital (VH) in Prince Albert, SK for the period1996-2010.

METHODS: Pediatric patients (<17 yoa) admitted to RUH or VH with osteomyelitis between January 1996 and December 2010 were identified using the ICD-10 code for osteomyelitis (M86.9), and cross-referenced with microbiological data for confirmed *S. aureus* etiology. If microbiological data was not available for cross-referencing, then a manual search of the chart was performed. A standard case form was used to abstract epidemiological and clinical data from the hospital chart. Patients with chronic osteomyelitis or with osteomyelitis not due to *S. aureus*, were excluded. Descriptive statistics were performed using SPSS v18.

RESULTS: Forty-seven patients had *S. aureus* acute or subacute osteomyelitis, with seven MRSA cases. The prevalence of MRSA increased over time (16.7% 1996 to 2000, 0% 2001 to 2005, 31.3% 2006 to 2010). The majority of patients presented with fever and site pain (76.1% and 95.7% respectively), with preceding trauma reported by < 10%. Although elevations in white blood cells or neutrophils were present in less than 50% of patients, >90% had elevations of ESR and/or CRP. Patients with MRSA osteomyelitis had more severe clinical manifestations, were more likely to have complications and were more likely to have abnormal laboratory findings.

CONCLUSIONS: In the past 15 years *S. aureus* osteomyelitis in pediatric patients at the RUH and Victoria Hospitals has been increasing, with those due to MRSA becoming more prevalent. Osteomyelitis, secondary to MRSA, appears to cause a more severe clinical course.

SP21

ANTIMICROBIAL RESISTANCE AND SEROTYPE DISTRIBUTION OF *STREPTOCOCCUS PNEUMONIAE* IN EASTERN ONTARIO

C ELLIS[1,2], R ROBINSON[3], M DESJARDINS[1,2,3], B TOYE[1,2,3]

[1]The Ottawa Hospital; [2]University of Ottawa; [3]Ottawa Hospital Research Institute, Ottawa, ON,

OBJECTIVES: We determined the prevalence of antimicrobial resistance and the mechanism of macrolide resistance in Sp and the serotype distribution of invasive isolates in Eastern Ontario.

METHODS: Single patient clinical isolates of Sp were collected from all 14 acute hospitals and one community laboratory in Eastern Ontario during 2011. Oxacillin, norfloxacin (Nor), clindamycin and erythromycin (Ery) susceptibility were determined by disk diffusion testing. PCR for both

erm(B) and *mef* genes was done on all Ery resistant isolates. Penicillin (Pen) and Levofloxacin (Lev) MICs were determined by E-test. Serotype data was collected for blood and CSF isolates from 2011-12.
RESULTS: A total of 412 isolates of Sp (including 42% respiratory, 27% sterile sites) were collected. Of these, 84 (20%) were resistant to Ery: 39% due to *erm(B)*, and 54% due to *mef*; 7% were found to carry both genes. Forty-seven isolates (11%) had Pen MIC of 0.12-1 mg/L and only two isolates (0.5%) had Pen MIC ≥2 mg/L. Resistance to Ery (24% vs 19%) and Pen MIC >0.06 mg/L (16% vs 9%) were slightly higher in the tertiary care hospitals than in the community hospitals, respectively. 3/321 (1%) isolates tested were resistant to Nor and had a Lev MIC >2 mg/L. 49% of the 171 invasive Sp isolates had serotypes that are contained in the 13-valent conjugate vaccine (PCV-13). The most common serotype in both children and adults was 7F (24%).
CONCLUSION: The prevalence of macrolide resistance emphasizes the need for continued surveillance, especially for isolates with a dual mechanism of Ery resistance which have been associated with multidrug resistance. The serotype distribution of invasive Sp isolates supports the introduction of PCV-13 in Ontario in 2011.

SP22

A SYSTEMATIC REVIEW OF AVAILABLE MEASURES OF ANTIMICROBIAL USE IN HOSPITALIZED PEDIATRIC POPULATIONS

E FORTIN[1,2], PS FONTELA[3], AR MANGES[1,4], DL BUCKERIDGE[1], RW PLATT[1], C QUACH[1,2,3]
[1]Departement of Epidemiology, Biostatistics and Occupational Health, McGill University; [2]Direction des risques biologiques et de la santé au travail, Institut national de santé publique du Québec, Québec and Montréal, QC; [3]Department of Pediatrics, The Montreal Children's Hospital, McGill University, Montréal, QC; [4]School of Population and Public Health, University of British Columbia, Vancouver, BC

OBJECTIVES: Measures quantifying antimicrobial (AM) use in a population have been described, but the measure to privilege for surveillance of AM resistance in hospital settings, especially when including pediatric populations, is unknown. This systematic literature review aims to 1) describe existing measures of AM use in pediatric inpatient populations; 2) understand how well they correlate; and 3) compare their ability to predict subsequent AM resistance levels.
METHODS: We searched the MEDLINE, EMBASE, CINAHL and LILACS databases for cohort and repeated point-prevalence studies presenting data on AM use in a population of hospitalized children (1975 to 2011). Two reviewers independently screened all eligible studies, studies published in 2011 in two journals and two proceedings, studies suggested by experts in the field, and references of all included studies.
RESULTS: From the 79 selected studies, 26 measures (numerator/denominator) were identified; the most frequently used being defined daily doses / patient-days (DDD / pd, 26%) and exposed patients / total patients (% exposed, 23%). Only two studies quantitatively compared different measures. One study used data from 40 pediatric hospitals and reported a positive correlation between % exposed and AM-days / pd. The other study was the only one to compare AM use measures (quinolones) to resistance rates (in Gram-negative rods). Over 7 years, a strong correlation was observed between doses / pd and agent-days / pd (ρ=0.98), but doses / pd correlated more with resistance rates (ρ=0.80 vs 0.55).
CONCLUSION: Our results highlight the need for an exhaustive comparison of known measures and their ability to accurately predict resistance in pediatric populations.

SP23

POINT PREVALENCE STUDY OF ANTIMICROBIAL UTILIZATION IN A CANADIAN TERTIARY-CARE TEACHING HOSPITAL

C LEE, S WALKER, N DANEMAN, M ELLIGSEN, L PALMAY, B COBURN, A SIMOR
Sunnybrook Health Sciences Centre, Toronto, ON

OBJECTIVES: Inappropriate antimicrobial use can promote antimicrobial resistance, which is associated with increased patient morbidity and mortality. Identification of the pattern of hospital antimicrobial use can provide baseline data from which target interventions to optimize antimicrobial utilization may be identified. The primary objective was to identify the prevalence of antimicrobial use at a tertiary care teaching hospital with both acute care patients and long-term complex care residents. The secondary objective was to identify the pattern of prescribing and extent of infectious diseases (ID) and/or antimicrobial stewardship (ASP) involvement in antimicrobial prescribing.
METHODS: A point prevalence study was conducted on July 19, 2012. Patients were identified using an integrated database (SPIRIT), and information on antimicrobial use, diagnostic imaging, and ID/ASP consultations were collected by prospective chart review.
RESULTS: One or more antimicrobials were ordered in 178 (17%) of 1021 hospitalized patients. Antibiotics were the most common class of antimicrobials used (91% of all antimicrobial orders). Fluoroquinolones, cephalosporins, and penicillins were the most common classes of antibiotics ordered (62% of all antibiotics ordered). The majority of orders (89%) were for a definite indication; and the most common indications for treatment were pneumonia, symptomatic urinary tract infection, *C. difficile* infection, secondary bacteremia, and febrile neutropenia.
CONCLUSION: This prospective point prevalence study provided important baseline information about antimicrobial use in a large tertiary care teaching hospital, which identified antibiotics and indications that may be targeted to optimize antimicrobial use.

SP24

RECURRENT GENITAL CHLAMYDIAL INFECTIONS: A NOVEL METHOD FOR TYPING AND MOLECULAR DETECTION OF AZITHROMYCIN RESISTANCE

M TADROS[1], S ZITTERMANN[2], R MELANO[2], V ALLEN[1,2]
[1]University of Toronto; [2]Public Health Ontario, Toronto, ON

BACKGROUND: High rates of recurrence of *Chlamydia trachomatis* genital infection have been associated with the use of macrolides, suggesting treatment failure or reinfection. A novel multilocus sequence typing (MLST) system was created by Klint et al. This system targets five different genetic regions, which are amplified, sequenced and subsequently assigned a genetic profile through a curated database http://mlstdb.bmc.uu.se/ .
OBJECTIVES: To evaluate the discriminatory power of the MLST 5 method for *C. trachomatis* from genital and urine specimens submitted for NAAT, to assess, whether isolates from patients with persistent infection have identical MLST 5 types, and to perform molecular evaluation for potential macrolide resistance.
METHODS: Eight reference specimens and 50 urine/genital specimens positive for *C. trachomatis* by NAAT from 25 patients (two samples from each patient) were used in our study. MLST 5 targets were amplified by PCR and sequenced. Patient samples were tested for potential mutations conferring resistance to macrolides by sequencing the two copies of 23S rRNA as well as the L22 and L4 regions.
RESULTS: MLST 5 targets were successfully amplified in all reference strains but in only 11/25 (44%) patient NAAT samples. Eight different MLST patterns were identified among the eight reference strains, and in 12/22 of patients' specimens. Identical MLST 5 patterns were identified in duplicate samples from 10/ 11 patients. Reported mutations conferring macrolide resistance were not found.
CONCLUSIONS: MLST 5 is a novel typing method for *C. trachomatis* with a good discriminatory power to investigate clinical failure and as a preliminary step for molecular evaluation of macrolide resistance.

SP25

THE USE AND INTERPRETATION OF MLVA FOR *E. COLI* O157:H7 OUTBREAKS IN CANADA

J RUMORE[1,2], L TSCHETTER[2], C TABING[2], C PETERSON[2], C GOODMAN[2], A KEARNY[2], C NADON[1,2]

[1]University of Manitoba; [2]National Microbiology Laboratory, Public Health Agency of Canada, Winnipeg, MB

OBJECTIVE: Multiple Locus Variable Number Tandem Repeat Analysis (MLVA) is a molecular subtyping method for use with Pulsed-field Gel Electrophoresis (PFGE), the current gold standard method for subtyping *E. coli* O157:H7. Ideally, MLVA should provide further specificity to outbreak investigations by helping to differentiate outbreak and sporadic cases. The objective of this study was to determine the usefulness of MLVA for the identification and investigation of three recent *E. coli* O157:H7 outbreaks in Canada.

METHODS: Using the subtyping methods PFGE and MLVA, a descriptive analysis of three recent and separate *E. coli* O157:H7 outbreaks associated with walnuts and beef burgers was conducted.

RESULTS: Forty-six isolates were subtyped during the time period of a 2011 outbreak associated with walnuts; 14 were found with the PFGE pattern combination EXCAI.1194, ECBNI.0895, which was not common at that time. The MLVA results were useful in differentiating this outbreak from a concurrent outbreak associated with hazelnuts in the United States. Of the 148 isolates analysed for a 2012 outbreak associated with beef burgers, 18 cases were found to have the highly common PFGE pattern EXCAI.0001, ECBNI.0012. The MLVA results were integral in separating outbreak-related and sporadic cases, and for confirming the food source. During a second 2012 outbreak associated with beef burgers, five cases out of 139 isolates were confirmed with PFGE pattern EXCAI.2850, ECBNI.0202, which had not been seen in Canada before. The MLVA results confirmed the findings of PFGE; however, PFGE was used to define the outbreak and to confirm the food source.

CONCLUSION: The utility of MLVA was dependent on the prior frequency of the PFGE pattern of the outbreak strain; it provided specificity when the PFGE pattern is common. A thorough analysis of the distribution of MLVA profiles among surveillance and outbreak data in Canada is needed and is currently underway.

SP26

REAL-TIME PCR ASSAY TO DETECT SHIGA TOXIN-PRODUCING *ESCHERICHIA COLI* IN ONTARIO WELL WATERS

L CHENG[1], J KROLIK[2,3], A JOYCE[3], G HALL[4], E GARDNER[5], G EVANS[2,6], A MAJURY[3,7]

[1]Dalla Lana School of Public Health, University of Toronto, Toronto; [2]Department of Pathology and Molecular Medicine, Queen's University; [3]Public Health Ontario; [4]Department of Civil Engineering, Queen's University; [5]Kingston, Frontenac and Lennox & Addington Public Health; [6]Division of Infectious Diseases, Kingston General Hospital; [7]Department of Biomedical and Molecular Sciences, Queen's University, Kingston, ON

OBJECTIVES: Shiga toxin-producing *Escherichia coli* (STEC) are emerging foodborne and waterborne pathogens that can cause significant disease in humans. STEC produce Shiga-like toxins, which are coded by the genes *stx1* and *stx2*. While O157:H7 is often implicated in STEC outbreaks, non-O157 strains, which are not routinely detected by traditional culture methods, may comprise up to 50% of all STEC infections. The Public Health Ontario (PHO) Laboratories do not currently employ standard protocols for the detection of O157 or non-O157 STEC in water. Thus, the purpose of this study was to optimize a real-time quantitative PCR (qPCR) assay for the rapid detection of *stx1* and *stx2* from DNA extracted from well waters. The test was then applied to 100 well water extracts.

METHODS: The STX qPCR assay for stool samples used by the PHO Laboratory in Toronto was selected for use with water samples. Serial dilutions of O157:H7 genomic DNA were tested using the assay to determine the detection limit and efficiency of the test. The assay was then applied to 100 DNA extracts from well water samples which were previously tested for species-specific fecal contamination.

RESULTS: The detection limit of the qPCR assay was one O157:H7 genome copy per PCR reaction, and the assay demonstrated high (102% to 104%) PCR efficiency. The assay detected low numbers of O157:H7 despite the presence of PCR inhibitors inherent in well waters. Of 100 well water samples, two were positive for both *stx1* and *stx2*, and one was positive for *stx2* only.

CONCLUSION: A rapid, sensitive, and efficient qPCR assay capable of detecting STEC from well water was optimized for use in PHO Laboratories. This test can facilitate outbreak investigations, implementation of interventions, and the timely diagnosis and treatment of infected patients.

SP27

EVALUATION OF DIFFERENT COMMERCIAL AGAR MEDIA FOR THE DETECTION OF SHIGA TOXIN-PRODUCING *ESCHERICHIA COLI* AND THE USE OF MOLECULAR TYPING TO RAPIDLY IDENTIFY SEROTYPES

N ZELYAS[1], A POON[1], L CHUI[1,2]

[1]University of Alberta; [2]Provincial Laboratory for Public Health, Edmonton, AB

OBJECTIVE: Both O157 and non-O157 Shiga toxin-producing *Escherichia coli* (STEC) are responsible for major outbreaks worldwide. Non-O157 STEC are under reported because it is difficult to identify them using the current culture protocol. The goals of this study were to evaluate 1) different agar media; 2) molecular serotyping methods for non-O157 STEC.

METHODS: 167 non-O157 STEC isolates were inoculated onto Colorex® STEC, Rainbow® O157, BD CHROMagar™ O157, Colorex® O157, and MacConkey agar. The growth characteristics of the isolates on each of the selected media were recorded. Representatives of isolates were spiked in different quality of stools (watery, mucoid, semi-formed and bloody), enriched 16h in MacConkey broth, and then subcultured onto the selective plates. Molecular serotyping of 22 non-O157 STEC isolates was performed by PCR followed by sequencing of the *gnd* locus.

RESULTS: Colorex® STEC and Rainbow® O157 agar identified 88.6% and 76.0% of the non-O157 isolates respectively. 100% of STEC isolates grew on Colorex® O157 and MacConkey agar and only 89.2% of them grew on BD CHROMagar™ O157 agar. In the spiked stool samples, growth of the organisms depended on the quality of the spiked stools. Molecular typing correctly identified the serotype of 22 different non-O157 STEC isolates.

CONCLUSIONS: Colorex® STEC provides an option for the screening of STEC from clinical samples. Molecular typing is a fast and accurate method to serotype isolates. A diagnostic algorithm incorporating the use of these media and molecular typing could correctly identify STEC serotypes from specimens within three days of arriving at the clinical laboratory.

SP28

DEVELOPMENT OF URINARY TRACT INFECTION (UTI) PREDICTION RULES BASED ON AN AUTOMATED URINE CHEMISTRY ANALYZER AND A FLOW CYTOMETRIC URINE CELL ANALYZER

A GHALY[1], K THOMPSON[1], K FORWARD[2,3]

[1]Department of Medicine; [2]Departments of Pathology, Microbiology & Immunology and Medicine, Dalhousie University; [3]Department of Pathology and Laboratory Medicine, Queen Elizabeth II Health Sciences Centre, Halifax, NS

OBJECTIVES: Urinalysis results may improve diagnostic accuracy and can be used to help guide microbiologic workup when UTI is suspected. We developed prediction rules based on the CLINITEK® AUWi system to facilitate diagnosis, guide empiric antibiotic use and limit processing of low yield urine samples.

METHODS: Over a three-month period, we examined results of 1483 paired urine specimens collected within two hours of each other. Automated urinalysis was performed using the AUWi. Cultures were performed using sheep blood agar +/− MacConkey agar. Culture was considered significant if a single morphotype grew 10^8 cfu/L. AUWi determined the number of bacteria, WBCs, RBCs, epithelial cells, leukocyte esterase

(LE) and the presence of nitrites. We identified cut offs for the continuous variables using receiver operator curves (ROC), and identified the most useful variables to predict significant growth.

RESULTS: 549 samples had significant growth; 934 had no growth. Samples with mixed growth were excluded. Using ROC, thresholds for AUWi were identified for bacteria (270), WBCs (14), RBCs (0), epithelial cells (27) and LE (500). Using these thresholds, bacteria (PPV 79%, NPV 92%), WBCs (PPV 73%, NPV 86%), LE (PPV 81%, NPV 74%) and nitrites (PPV 95%, NPV 74%) were the most predictive of infection. Two models were developed using these parameters. Epithelial and RBCs were not useful predictors. When combined with bacteria and nitrites, WBCs were minimally more predictive than LE (AUC 0.9339 vs. 0.9176). Fewer than 4% (25/718) of samples were culture positive in the absence of three variables (bacteria, white blood cells, nitrites), whereas 98% (177/180) of samples with all three variables had significant growth.

CONCLUSIONS: Prediction rules using select variables determined by AUWi may be useful for the clinical and laboratory assessment of suspected UTIs.

SP29

IMPACT OF URINE CULTURES WITHOUT CLINICAL INDICATION ON ANTIMICROBIAL PRESCRIPTION FOR ASYMPTOMATIC BACTERIURIA AMONG MEDICAL AND SURGICAL INPATIENTS

J LEIS[1,2], N DANEMAN[2], K SHOJANIA[3], W GOLD[2], D LOW[1], A MCGEER[1,2]

[1]Department of Microbiology, University Health Network & Mount Sinai Hospital; [2]Division of Infectious Diseases, Department of Medicine, University of Toronto; [3]University of Toronto Centre for Patient Safety, Toronto, ON

OBJECTIVE: A prospective audit of urine culture ordering practices was undertaken to assess the proportion of urine cultures ordered among medical/surgical inpatients without an appropriate clinical indication, and the proportion which resulted in inappropriate antimicrobial therapy of asymptomatic bacteriuria (ASB).

METHODS: Urine cultures from non-pregnant patients on medical/surgical services at two acute care hospitals were identified within 24 h of test ordering during August and September, 2012. Indications for test ordering were recorded and patients assessed for presence of urinary tract infection (UTI) using CDC criteria. Culture results and antimicrobial prescriptions were documented 48 hours later.

RESULTS: Sixty-eight per cent (76/112) of tests ordered did not meet CDC clinical criteria for UTI. Detection of ASB led to antimicrobial therapy in 57% (12/21) of cases. Forty-two per cent (32/76) of urine cultures without clinical indication were sent from non-catheterized patients from the ward, 30% (23/76) from non-catheterized patients on admission, 21% (16/76) from catheterized patients on the ward, and 7% (5/76) from catheterized patients on admission. After applying the culture results to the CDC criteria, a UTI was confirmed in 38% (3/8) of catheterized patients cultured on admission, 12% (4/34) of non-catheterized patients on admission, 5% (1/21) of catheterized patients from the ward and 2% (1/49) of non-catheterized patients from the ward.

CONCLUSION: Medial/surgical inpatients without an indwelling catheter represented nearly half of urine cultures ordered without clinical indication despite carrying the lowest risk of UTI. These patients represent a potential target population for future quality improvement initiatives aiming to decrease antimicrobial therapy for ASB.

SP30

EVALUATION OF TWO CHROMOGENIC MEDIA FOR THE ISOLATION AND IDENTIFICATION OF URINARY TRACT PATHOGENS (UTP)

M PAYNE[1], D ROSCOE[1,2]

[1]University of British Columbia; [2]Vancouver General Hospital, Vancouver, BC

OBJECTIVE: Chromogenic media (CM) are available for urine specimens (US) to enable rapid identification of common pathogens. Two CM, chromID CPS 4 (bioMérieux) and UriSelect (URS) 4 (Bio-Rad) were compared to the standard media (SM) used for US.

METHODS: The performance of CM was compared to a routine protocol (BAP and MAC) for the isolation and identification of UTP. In June/July 2012, all US received between 9am and 5pm on 10 specific days were inoculated to CPS 4, URS 4, BAP and MAC plates using 1 µL and 10 µL for non-invasive and invasively collected US, respectively. CM interpretation was done according to the product inserts by one person blinded to the results of SM. SM were read by experienced technologists according to protocol and isolates were identified using BD Phoenix™. Results were grouped into significant (SG), mixed (MG), and no significant growth (NSG). SG was defined as $\geq 10^7$ CFU/L, MG as ≥3 species, and NSG as $<10^7$ CFU/L for standard specimens.

RESULTS: A total of 903 US were studied. SM identified 239 SG, 112 MG, and 552 NSG cultures. The most common pathogens were *E.coli* (38%) and *Enterococcus spp.* (11%). Comparing CM to SM, the exact agreement (same species isolated) was 89.3% and 89.5% for URS 4 and CPS 4, respectively. When grouped by clinical significance (MG, NSG, and SG-same species), agreement with SM was 93.0% and 93.1% for URS 4 and CPS 4, respectively. CM were equivalent with respect to processing time, both required less than SM.

CONCLUSION: Both CM compared well to SM and allowed for rapid preliminary identification of many UTP. Advantages include decreased labour and need for other identification of certain species, particularly *E.coli*. In terms of workflow, CM enables same-day identification for almost 50% of significant UTP.

SP31

EVALUATION OF ALERE PBP2A KIT FOR RAPID PRELIMINARY IDENTIFICATION OF MRSA DIRECTLY FROM BLOOD CULTURES FLAGGED BY BACT/ALERT 3D

Y YU[2], L JIAO[2], A JISSAM[1], M MCCONNELL[1], C MAIN[1,2]

[1]Hamilton Regional Laboratory Medicine Program; [2]McMaster University, Hamilton, ON

OBJECTIVE: *Staphylococcus aureus* bacteremia is associated with high morbidity and mortality and therefore, all cases identified in our institution are seen by an Infectious Disease physician. The increasing prevalence of methicillin resistance in *Staphylococcus aureus* (MRSA) has led to the widespread use of vancomycin as empiric therapy. Thus, more rapid characterization of *S. aureus* isolates from blood cultures as MRSA or MSSA (methicillin sensitive) would be beneficial for patients. The objective of this study was to evaluate the use of the Alere PBP2a kit (Inverness Medical Innovations, Inc. Waltham, MA) for rapid identification testing of MRSA directly from blood culture broth (BCB) media (BacT/ALERT 3D, bioMerieux).

METHODS: Previously characterized *Staphylococcus* isolates (both MRSA and MSSA) and fresh clinic isolates from the blood cultures identified as *Staphylococcus aureus* based on gram stain and tube coagulase were evaluated. 3mls of BCB was centrifuged in a serum separating tube (SST). The sediment on top of the gel in SST was used for testing with Alere PBP2a kit. The final identification of the isolates (MRSA or MSSA) was based on off line tests (including Bio-Rad MRSASelect agar, colony PBP2a testing) and/or Vitek 2 results.

RESULTS: In this study, the sensitivity of the PBP2a kit is 100% (15/15) in identifying MRSA. The specificity is 77.3% (17/22). The reporting time for MRSA was 4 h to 6 h from the time the blood culture was flagged by BacT/ALERT 3D, compared to 18 h to 24 h using routine methods.

CONCLUSION: The Alere PBP2a kit is a sensitive method to shorten the turn-around-time of reporting MRSA directly from blood culture broth. This may reduce unnecessary vancomycin use at our institution. The implementation of this method in our lab may positively impact patient care, however incorporating the test into current workflow poses some challenges.

SP32

THE EFFECT OF SCC*MEC* TYPE AND COLONY SPREADING ABILITY OF METHICILLIN-RESISTANT *STAPHYLOCOCCUS AUREUS* ON THE SURVIVAL OF *CAENORHABDITIS ELEGANS*

M REA[1], GR GOLDING[2], PM CHONG[2], AE SIMOR[3], A MOUNCHILI[4], D GRAVEL[4], K KATZ[5], M LOEB[6], A MCGEER[7], P KIBSEY[8], K WEISS[9], J EMBIL[10], S SMITH[11], C FRENETTE[12], MR MULVEY[2]

[1]Department of Medical Microbiology, University of Manitoba; [2]National Microbiology Laboratory, Public Health Agency of Canada, Winnipeg, MB; [3]Sunnybrook Health Sciences Centre, Toronto; [4]Healthcare-Associated Infections Surveillance and Epidemiology, Public Health Agency of Canada, Ottawa; [5]North York General Hospital, Toronto; [6]McMaster University, Hamilton; [7]Mount Sinai Hospital, Toronto, ON; [8]Royal Jubilee Hospital, Victoria, BC; [9]Maisonneuve-Rosemont Hospital, Montreal, QC; [10]Health Sciences Centre, Winnipeg, MB; [11]University of Alberta, Edmonton, AB; [12]McGill University Health Centre, Montreal, QC

OBJECTIVE: Methicillin-resistant *Staphylococcus aureus* (MRSA) is a common pathogen in both the hospital and community settings. CMRSA2 (USA100/800, ST5) has been identified in both settings and is currently the most common MRSA strain type in Canada. It was observed that *S. aureus* has the ability to spread on soft agar plates and this may be beneficial its survival. This study examined CMRSA2 with different SCC*mec* types to determine spreading ability and differences in virulence characteristics using a *Caenorhabditis elegans* infection model.

METHODS: A selected number of CMRSA2 isolates (n=40) collected from the Canadian Nosocomial Infection Surveillance Program (CNISP) were analysed. SCC*mec* typing was conducted on all isolates. Colony spreading ability of MRSA was tested using a spread plate assay. *C. elegans* slow killing assays were performed on all isolates.

RESULTS: A total of 40 CMRSA2 isolates were characterized from clinical isolates identified over a 10 year period. An equal number of CMRSA2 isolates harbouring SCC*mec* II (n=20) and IV (n=20) were characterized. The SCC*mec* type II isolates had an average spreading distance of 16.8 mm, while the SCC*mec* type IV isolates had a spreading distance of 17.4 mm. *C. elegans* killing assays were also performed on this selection of CMRSA2 isolates. It was found that MRSA isolates that had the ability to spread (n=28) on a soft agar surface had greater nematocidal activity than the isolates that did not spread on soft agar plates (n=12) (p=0.0051). There was not a significant correlation between SCC*mec* type and nematode death in CMRSA2 isolates (p=0.1358).

CONCLUSION: The ability of MRSA to spread on a soft agar surface was correlated with the killing ability of nematodes by MRSA.

SP33

A PRELIMINARY REPORT OF INFANT NASAL MICROBIOME DIVERSITY APPLYING *CPN60* UNIVERSAL TARGET (UT) AS A PHYLOGENETIC MARKER

S PETERSON[1], N KNOX[1,2], G GOLDING[1,2], J EMBREE[1], F FLEMING[1], S FANELLA[1], M GRAHAM[1,2], G VAN DOMSELAAR[1,2], MR MULVEY[1,2]

[1]Dept. of Medical Microbiology, University of Manitoba; [2]National Microbiology Laboratory, Public Health Agency of Canada, Winnipeg, MB

OBJECTIVE: A protocol was developed to study the nasal microbiome applying a *cpn60* universal target (UT) amplicon as a bacterial phylotyping marker using next generation sequencing. The UT can be used to identify organisms in metagenomic samples with greater taxonomic specificity than 16S rRNA. We aimed to determine the typical composition of healthy infant anterior nares microbiota, and to explore the relation to microflora observed within their primary caregivers.

METHODS: Ethics approval was obtained from the U of Manitoba and PHAC. Nares swabs were collected from 40 two-week old infants and their primary caregivers using a single swab in one nare for each subject. Samples were treated with MoLysis (Molzym) to diminish contaminating human DNA, and bacterial template DNA was augmented using the GenomiPhi (GE Healthcare) whole-genome linear augmentation procedure. The *cpn60* UT target was PCR amplified using degenerate primers and sequenced using 454/GS FLX pyrosequencing. Reads were quality-filtered and matched to the *cpn60* database using an 80% nucleotide identity cutoff.

RESULTS: Reads were assigned to 23 unique phylotypes, with the 12 most abundant phylotypes accounting for >98% of the community. Infant/caregiver samples contained 63%/76.5% Actinobacteria, 35%/18% Firmicutes and 2%/5.5% Proteobacteria and trace amounts of Bacteroidetes. 17% of infant reads were streptococci, compared to 3.9% in caregivers. Lactobacilli represented 0.04%/0.003% (infants/caregivers) and Acinetobacter represented 0.4%/2.7%.

CONCLUSIONS: Lactobacilli and streptococci were more commonly found in infants, while Acinetobacter was more likely to be associated with caregivers. This data is in agreement with previous studies of healthy adult human nares microflora.

SP34

HUMAN RHINOVIRUS (HRV) INFECTION IN CHILDREN PRESENTING TO A PAEDIATRIC EMERGENCY DEPARTMENT

N NASHID[1], A GRANADOS[2,3], M SMIEJA[2,3], J MAHONY[2,3], JM PERNICA[1]

[1]McMaster University, Department of Pediatrics; [2]St. Joseph's Healthcare; [3]McMaster University, Department of Pathology and Laboratory Medicine, Hamilton, ON

OBJECTIVES: To determine whether human rhinovirus (HRV)C is over-represented in children with asthma and to investigate whether HRV viral load (VL) correlates with the severity of respiratory symptoms in children.

METHODS: Nasopharyngeal swabs from children aged three months to five years presenting to McMaster Children's Hospital Emergency Department in September-October 2010 and 2011 were screened using PCR for influenza A/B, RSV A/B, parainfluenza I-III and adenovirus. Negative swabs (n=116) underwent further PCR testing to identify HRVA/B/C samples (n=75), and HRV viral loads were determined. Chart reviews identified children at "high risk" for developing asthma, using the modified (loose) Asthma Predictive Index (API). Severity of respiratory symptoms at presentation was quantified using the Paediatric Respiratory Assessment Measure (PRAM). Patients with significant cardiac or pulmonary disease or malignancy were excluded from analysis.

RESULTS: Of the 75 HRV-positive samples, clinical data were missing for seven and 21 were excluded due to co-morbidities. The median age of the remaining 48 participants was 16 months and 33% were girls. The mean VL was 6.5 $\log_{10}$ genome copies/ml (4.0-10.1). There were 15 HRVA, two HRVB, and 31 HRVC infections. The association between VL and respiratory disease severity was neither statistically nor clinically significant. The association between HRVC infection (as compared to HRVA/B) and children at higher risk for asthma was not statistically significant (OR 1.7 [95% CI 0.5 to 6.1).

CONCLUSIONS: HRV VL was not associated with severity of respiratory disease. However, our results suggest that the odds of developing asthma are higher in children with HRVC infections, as compared to HRVA/B. This association was not statistically significant, which may be due to a lack of power in this small exploratory investigation. Further studies are needed.

SP35

COST EFFECTIVE REAL-TIME PCR FOR THE DETECTION OF VARICELLA ZOSTER VIRUS

K BINKHAMIS[1], T AL-SIYABI[1], T HATCHETTE[1,2], J LEBLANC[1,2]

[1]QEII Health Sciences Centre; [2]Dalhousie University, Halifax, NS

OBJECTIVE: Molecular-based assays have become gold standard for the detection of viruses; however, their cost is far more expensive than virus culture and direct fluorescent antibody. Here we evaluate a low cost in-house real-time polymerase chain reaction (PCR) for varicella zoster virus (VZV) and replaced nucleic acid extraction (NAE) with a homogenization and heat (HH) treatment.

METHODS: Using 10-fold dilutions (n=12), the limit of detection (LoD) and inter- and intra-run variations were calculated using virus culture and DFA, real-time PCR following HH treatment, and real-time PCR following NAE. Specificity was assessed using a number of organisms including various herpes viruses. The three methods were then compared using 200 clinical specimens. Performance was compared to a modified gold standard (2/3 concordant results). Discordant results were resolved by manual DNA extraction and a commercial real-time PCR.
RESULTS: The real-time PCR assay was highly specific for VZV. Using NAE, the analytical sensitivity was between 3 and 18 copies per reaction. Using HH treatment it was ~18 copies per reaction, suggesting a slight decrease in sensitivity. Virus culture and DFA was ~10,000-fold less sensitive than either molecular method. With clinical specimens, all methods were highly specific (100%). The sensitivity for NAE with real-time PCR was 100%, 97.2% for HH with real-time PCR, and virus culture and DFA was significantly less sensitive (54.8%).
CONCLUSION: While slightly less sensitive than NAE, HH treatment coupled to an in-house real-time PCR provides a cost effective method for the detection of VZV with performance characteristics far exceeding traditional virus culture and DFA.

SP36

A COST EFFECTIVE REAL-TIME PCR ASSAY FOR THE DETECTION OF ADENOVIRUS

T AL-SIYABI[1], K BINKHAMIS[1], M WILCOX[2], W SALLENE[3], P KANTI[3], R TELLIER[3,4], T HATCHETTE[1,2], J LEBLANC[1,2]
[1]QEII Health Science Center; [2]Dalhousie University, Halifax, NS; [3]Provincial Laboratory for Public Health; [4]University of Calgary, Calgary, AB

OBJECTIVE: Real-time polymerase chain reaction (PCR) is a highly sensitive and accurate method for detection of human adenoviruses. However, cost prohibits its use in many laboratories. To improve costs, an in-house real-time PCR was evaluated and nucleic acid extraction (NAE) was replaced with homogenization and heat treatment (HH).
METHODS: The performance of virus culture with direct fluorescence antibody (DFA) testing was compared to the real-time PCR assay following HH or NAE on an automated instrument (MagNAPure). The limit of detection (LoD), dynamic range, inter- and intra-experimental variations were assessed by 10-fold serial dilutions of cultured adenovirus (n=24). Analytical specificity was evaluated against a wide range of organisms including various adenovirus types. 196 clinical specimens [swabs in universal transport media (UTM)] were tested in parallel using all three methods. A modified gold standard (2 of 3 concordant results) was used to determine sensitivity, specificity, accuracy, and precision. Discordant results were resolved using manual DNA extraction followed by a commercial real-time PCR.
RESULTS: The real-time PCR was highly specific and detected all serogroups of adenovirus. The LoD for the in-house real-time PCR following HH treatment or NAE were equally sensitive at 12 and 18 copies/reaction, respectively, and was ~100-fold more sensitive than cell culture and DFA. Compared to the modified gold standard, sensitivity of each assay was 100%, 97.5 % and 69.2%, respectively.
CONCLUSION: The in-house real-time PCR following HH treatment is an effective strategy for the detection of adenovirus, at a cost comparable to virus culture and DFA.

SP37

IDENTIFICATION OF A PERMISSIVE MUTATION RESTORING THE FITNESS OF THE H275Y OSELTAMIVIR RESISTANCE MUTATION IN 2009 PANDEMIC INFLUENZA A/H1N1 VIRUSES

A PIZZORNO, Y ABED, X BOUHY, G BOIVIN
Research Center in Infectious Diseases of the CHUQ-CHUL and Laval University, Québec, QC

OBJECTIVE: Neuraminidase (NA) mutations conferring resistance to oseltamivir generally compromise the fitness of influenza viruses. The only oseltamivir-resistant virus that widely spread in the population, the A/Brisbane/59/2007 (H1N1) strain with the H275Y substitution, contained additional permissive mutations that counteracted the detrimental effect induced by H275Y. The latter substitution is still infrequent in 2009 pandemic influenza viruses (A/H1N1pdm09), but computational approaches have predicted a set of compensatory NA mutations, with T289M as the best candidate. We sought to investigate the effect of T289M on the fitness and virulence of A/H1N1pdm09 viruses containing the H275Y oseltamivir resistance mutation.
METHODS: Recombinant A/H1N1pdm09 wild-type (WT), H275Y and H275Y+T289M NA mutants were generated by reverse genetics. The enzymatic parameters *Km* and *Vmax* were determined by fluorometric-based assays whereas replicative capacities were evaluated by yield assays on ST6GalI-MDCK cells. Infectivity and virulence were assessed in C57BL/6 mice.
RESULTS: The H275Y substitution significantly reduced the NA affinity (*Km*) and activity (*Vmax)* by 3-fold and 5-fold, respectively, compared to the WT. The T289M mutation partly restored both parameters, with the H275Y+T289M mutant showing 1.4-fold reduction in both affinity and activity compared to the WT. *In vitro*, the T289M mutation completely restored the detrimental effect caused by H275Y on the early stage of the viral replication curve. In mice, the H275Y group had reduced mean weight loss (6.2% vs 8.1%) and mortality (37.5% vs 50%) compared to the WT group. Conversely, the H275Y+T289M group had the highest mean weight loss (13.2%) and unaltered mortality rate (50%). Determination of lung viral titers on days 3 and 6 p.i. is in progress.
CONCLUSIONS: As predicted, the T289M NA mutation seems to enhance the fitness and virulence of A/H1N1pdm09 viruses containing the H275Y mutation both *in vitro* and *in vivo*. The potential emergence and dissemination of these variants should be carefully monitored.

SP38

THE MARS PILOT PROJECT: USING LABORATORY DATA TO ESTIMATE THE PERFORMANCE OF NATIONAL MEASLES AND RUBELLA SURVEILLANCE

T EISBRENNER[1,2], A SHANE[3], S DESAI[3], Y LI[3], J MACEY[3], T LIPSKIE[3], N KHALIL[3], J ISAAC-RENTON[4,5], M PETRIC[4,5], M KRAJDEN[4,5], Y CHANG[4], K MACDONALD[1,4], M NAUS[5,6], V SAHNI[6], K FONSECA[7], J MAY-HADFORD[7,8], C GRANT[7], T ST JEAN[9], L LACHANCE[9], S NOLT[9], L ROBBERTS[10], L GILBERT[10], C O'KEEFE[11], G BUTLER[11], J BEIRNES[1], J HIEBERT[1], A SEVERINI[1,2], S MUKHI[1,2], G TIPPLES[1,2,9]
[1]National Microbiology Laboratory, Public Health Agency of Canada; [2]Department of Medical Microbiology, Faculty of Medicine, University of Manitoba, Winnipeg, MB; [3]Centre for Immunization and Respiratory Infectious Diseases, Public Health Agency of Canada, Ottawa, ON; [4]BC Public Health Microbiology and Reference Laboratory; [5]Faculty of Medicine, University of British Columbia; [6]BC Centre for Disease Control, Vancouver, BC; [7]Alberta Provincial Laboratory for Public Health, Calgary/Edmonton, AB; [8]Office of Public Health Practice, Public Health Agency of Canada, Ottawa, ON; [9]Alberta Health, Edmonton, AB; [10]Newfoundland and Labrador Public Health Laboratory; [11]Newfoundland and Labrador Department of Health and Community Services, St. John's, NL; [12]Canadian Network for Public Health Intelligence, National Microbiology Laboratory, Public Health Agency of Canada, Winnipeg, MB

OBJECTIVES: Measles and rubella are nationally notifiable, vaccine-preventable diseases targeted for elimination by the Pan American Health Organization (PAHO). To support elimination efforts, surveillance optimization is important to ensure rapid case detection, document endemic transmission interruption, identify susceptible populations and inform immunization strategies. While current national enhanced surveillance captures confirmed-case data, its performance cannot be assessed using PAHO-recommended surveillance indicators as suspect-case investigation data are required for their estimation. In Canada, the investigation of clinically-suspect cases of measles-like illness (MLI) is very dependent on laboratory evidence. To address surveillance challenges, the Measles and Rubella Surveillance (MARS) pilot was developed to

explore the feasibility of implementing real-time integrated measles/rubella surveillance, and the use of augmented laboratory data to estimate surveillance performance.

METHODS: A MARS pilot application was developed using the web-based Canadian Network for Public Health Intelligence (CNPHI) platform to support real-time federal/provincial alerting upon initiation of a measles/rubella investigation at participating pilot sites (BC, AB and NL). Augmented laboratory data were collected via centralized reports to support surveillance indicator estimation during the June 2011 to May 2012 pilot year. Measles IgM serology testing was used as a laboratory-based proxy for MLI investigation.

RESULTS: Real-time, integrated surveillance was successfully implemented in MARS pilot provinces. 'Timeliness' and 'level of investigation' surveillance indicators were estimated using laboratory data, and compared with international performance targets. Indicator-based analysis demonstrated that 100% of real-time measles/rubella investigation reports met PAHO-recommended sample collection and laboratory receipt timelines, and 91.7% met laboratory result timelines (Targets: ≥80%). An estimated 99.8% of MLI investigations were laboratory-discarded (Target: ≥95%). Baseline performance was also estimated for indicators without defined target values in the literature.

CONCLUSIONS: Surveillance in MARS pilot provinces successfully met all laboratory-related PAHO surveillance targets evaluated. Laboratory data plays an important role in evaluating and documenting surveillance performance to support measles and rubella elimination efforts.

SP39

BOTH TRIF AND IPS-1 ADAPTOR PROTEINS CONTRIBUTE TO THE CEREBRAL INNATE IMMUNE RESPONSE AGAINST HERPES SIMPLEX VIRUS TYPE-1 (HSV-1)

R MENASRIA[1], N BOIVIN[1], M LEBEL[2], J PIRET[1], J GOSSELIN[2], G BOIVIN[1]

[1]Research Center in Infectious Diseases-CHUL, Quebec, QC; [2]Research Center in Endocrinology and Genomic-CHUL, Quebec, QC

OBJECTIVE: Toll-like receptors (TLRs) and RNA helicases (RLHs), namely the retinoic acid-inducible gene 1 (RIG-I) and the melanoma differentiation-associated gene 5 (MDA-5), are important cell sensors involved in the immunological control of viral infections mainly through interferon (IFN)-β production. Herein, we investigated the relative contribution of theTLR3 signalling pathway via its adaptor molecule TIR-domain-containing adaptor inducing IFN-β (TRIF) and RIG-I/MDA-5 signalling pathways via their adaptor molecule IFN-β promoter stimulator-1 (IPS-1), for the control of HSV-1 infection in the central nervous system (CNS) of mice.

METHODS: TRIF$^{-/-}$, IPS-1$^{-/-}$ and C57BL/6 wild-type (WT) mice were infected intranasally with 7.5×10^5 pfu of HSV-1. Mice were monitored for signs of encephalitis and survival. The viral DNA load, infectious viral titers and IFN-β levels were determined in mouse brains obtained on days 3, 5 and 7 following the infection. Transcriptional induction of IFN genes was evaluated by measuring the activation level of interferon regulatory factor 3 and 7 (IRF-3 and 7) in the brains.

RESULTS: TRIF$^{-/-}$ and IPS-1$^{-/-}$ mice exhibited increased sickness signs and higher mortality rates compared to WT (20%, 64% and 50% for WT, TRIF$^{-/-}$ and IPS-1$^{-/-}$ mice, respectively) (p=0.02 for TRIF$^{-/-}$ vs. WT and p=0.09 for IPS-1$^{-/-}$ vs. WT). Viral replication was significantly increased in brains of TRIF$^{-/-}$ (on days 5 and 7) and IPS-1$^{-/-}$ (on day 7) mice compared to WT. In addition, impaired IFN-β production was noted in brains of TRIF$^{-/-}$ and IPS-1$^{-/-}$ mice on day 5 along with decreased activation of IRF-3 (TRIF$^{-/-}$) and IRF-7 (IPS-1$^{-/-}$) compared to WT.

CONCLUSION: These data suggest that TRIF and, to a lesser extent, IPS-1 signalling pathways are important for the control of HSV-1 replication in the CNS. Moreover, the higher susceptibility of both types of knockout mice seems to be related to an impaired IFN-β production.

SP40

ANALYSIS OF RECEPTOR FOR ADVANCED GLYCATION END-PRODUCTS (RAGE) EXPRESSION BY REAL-TIME PCR IN NASOPHARYNGEAL ASPIRATES FROM CHILDREN WITH RSV INFECTION

K LEIFSO[1,2], R SLINGER[1,2]

[1]University of Ottawa; [2]Children's Hospital of Eastern Ontario, Ottawa, ON

OBJECTIVE: RSV is a common pathogen responsible for significant morbidity in the pediatric population, especially in children under 6 months of age. Recent studies have suggested that RAGE may be critical to determining RSV severity. RAGE knockout mice showed reduced nasal RSV viral load, reduced inflammatory cell number in lung digests and earlier resolution of RSV induced weight loss relative to wild-type mice. Our objective was to quantify mRNA expression levels of RAGE in nasopharyngeal aspirates (NPA) from children with respiratory infection.

METHODS: We obtained NPA samples from children who presented to the emergency department with respiratory illness. mRNA was isolated and analyzed for membrane RAGE (mRAGE), endogenously secreted RAGE (esRAGE), S100A8, S100A12 and S100B by quantitative real-time PCR. Signals were normalized against β-actin gene expression.

RESULTS: mRAGE mRNA was found in 30 of 43 RSV positive samples. There was no difference in cycle threshold between RSV A (ΔΔCt mean 4.55, SD 1.19) and RSV B (ΔΔCt mean 4.20, SD 1.54) samples. mRAGE was detectable in aspirates positive for rhinovirus, with relative quantities (ΔΔCt mean 4.72, SD 2.12) similar to RSV positive samples. No mRAGE signal was detected in samples positive for *Bordetella pertussis*, but β-actin mRNA was present at levels similar to samples positive for RSV.

CONCLUSION: mRNA transcripts for RAGE and S100 are detectable in NP aspirates from children with RSV and rhinovirus infection. Further study will pair expression levels with clinical data to investigate if levels of RAGE or S100 protein ligands at presentation are predictive of further illness severity and patient disposition.

SP41

KILLER IMMUNOGLOBULIN-LIKE RECEPTOR GENES IN TUBERCULOSIS PATIENTS

K BRAUN[1], J WOLFE[2], C LOPEZ[1], S KIAZYK[1,2], T B BALL[1,2], M SHARMA[1,2]

[1]University of Manitoba, Winnipeg, MB; [2]Public Health Agency of Canada, Winnipeg, MB

OBJECTIVE: Although tuberculosis (TB) rates in Canada are declining, incidence in Canadian Aboriginal populations and foreign born individuals living in Canada remains high. The objective of this study was to compile killer immunoglobulin-like receptor (KIR) gene profiles found in Canadian born Aboriginals, Canadian born non-Aboriginals, and foreign born individuals from Manitoba with active, latent, and uninfected TB status.

METHODS: DNA was extracted from whole blood using the Qiagen DNA Mini kit according to manufacturer's instructions. KIR profiles were determined in 209 patients (59 active, 46 latent, and 104 uninfected TB controls; 54 Aboriginal, 77 non-Aboriginal, and 78 foreign born individuals) using the Miltenyi Biotec KIR typing kit. TB disease status was determined by culture and/or clinical diagnosis.

RESULTS: Many significant differences were seen in KIR gene frequencies between individuals with and without tuberculosis. KIR2DL2/S2 (P=0.0005/0.0010) and 2DS3 (P=0.0009) were present less often in active TB cases compared to uninfected individuals. In addition, KIR2DSLB was present more often in latent TB cases compared to uninfected individuals (P=0.0125). These trends remain consistent when broken down into Aboriginal, non-Aboriginal and foreign born population groups.

CONCLUSION: This study shows that there are differences in the KIR gene frequency in tuberculosis patients of different population groups. Further investigation is needed to explore the subtleties of these differences using sequencing and/or KIR-HLA association studies.

SP42

IDENTIFICATION OF *MYCOBACTERIA* SPP AND *MYCOBACTERIUM TUBERCULOSIS* COMPLEX (MTBC) RESISTANCE DETERMINANTS USING THE MDR-TB ASSAY ON THE PLEX-ID PCR-ELECTROSPRAY IONIZATION MASS SPECTROMETRY (ESI-MS) SYSTEM

P SIMNER, S BUCKWALTER, N WENGENACK
Mayo Clinic, Rochester, MN, USA

OBJECTIVES: We evaluated the PLEX-ID MDR-TB assay for detection and identification of *Mycobacteria* spp and MTBC resistance determinants from culture.

METHODS: The MDR-TB assay was compared to traditional culture using previously characterized MTBC (n:68) and nontuberculous mycobacteria (NTM; n:97) isolates and 107 clinical MGIT™ broth cultures (MBC). Resistance profiles were compared to the indirect agar proportion method (IAPM) for MTBC isolates. The data was analyzed with two databases (V1 & updated V2). Discordant results were resolved by 16S rDNA sequencing.

RESULTS: The PLEX-ID identified all MTBC isolates regardless of the database used. However, database V1 co-identified 32.4% of the MTBC as both MTBC and a NTM. This was attributed to an algorithm issue that was resolved with database V2. Database V1 identified 38.1% and 72.9% of the NTM from characterized isolates and MBC, respectively. Improvements to database V2 resulted in 99% and 100% concordance for the NTM characterized cultures and MBC, respectively. In comparison to IAPM, the sensitivity and specificity for the detection of MTBC drug resistance using the MDR-TB assay was 100% and 92.3% for rifampin, 100% and 93.8% for isoniazid, 91.0% and 94.4% for ethambutol, and 100% and 100% for fluoroquinolones, respectively.

CONCLUSIONS: The MDR-TB assay appears to be a rapid and accurate method for the detection and identification of mycobacterial species and resistance determinants to MTBC directly from culture using the updated database V2. Implementation of this assay in a Clinical Microbiology laboratory would improve TATs and provide physicians with a prompt diagnosis and the ability to tailor the treatment regimens for *M. tuberculosis* prior to receiving phenotypic AST results.

SP43

THE INTRODUCTION OF MOLECULAR DETECTION OF MUTATIONS ASSOCIATED WITH FIRST-LINE DRUG RESISTANCE IN *MYCOBACTERIUM TUBERCULOSIS* IN MANITOBA

S SEPEHRI[1], J KARLOWSKY[1,2], S CHRISTIANSON[3], M SHARMA[3,1], M ALFA[1,2]
[1]Department of Medical Microbiology and Infectious Diseases, University of Maniotba, Winnipeg, MB; [2]Diagnostic Services of Manitoba, Winnipeg, MB; [3]National Reference Centre for Mycobacteriology, National Microbiology Laboratory, Winnipeg, MB

OBJECTIVE: Drug-resistant strains of *Mycobacterium tuberculosis* pose a serious threat to infection prevention and control efforts. Timely drug susceptibility testing is critical to ensure the effective treatment of patients and to stop the transmission of the disease. It is known that mutations in *katG* and/or *inhA*, *rpoB* and *pncA* genes are present in approximately 80% of Isoniazid (INH), 95% of Rifampin (RIF) and >95% of Pyrazinamide (PZA) resistant isolates respectively. The objective of this study was to assess the accuracy of molecular detection of resistance and its impact on turn-around time (TAT) compared to phenotypic testing.

METHODS: Since April 2012, detection of mutations in the *katG*/*inhA*, *rpoB* and *pncA* genes has been performed in conjunction with culture-based susceptibility testing. Only the mutations with high-confidence associations to resistance in the literature are being reported.

RESULTS: Of 93 *M. tuberculosis* isolates tested, 9 (9.8%) were resistant to INH via phenotypic testing, no other drug resistance was identified. Two isolates were resistant at the high concentration of INH (0.4 ug/mL), both were identified through mutations. Seven isolates were resistant at the low concentration of INH (0.1 ug/mL), one of which was identified by an *inhA* mutation. The remaining 6 isolates lacked mutations and were indistinguishable by MIRU-VNTR genotyping. For resistant isolates, the median TAT from receipt of the positive culture was 19 days for phenotypic testing compared to 3 days for molecular testing.

CONCLUSION: Detection of mutations associated with drug resistance in *M. tuberculosis* reduces the TAT significantly and provides specific drug susceptibility information which aids in prompt and appropriate treatment of tuberculosis patients and ceases the spread of the disease.

SP44

PARADOXICAL REACTIONS: PROGRESSIVE MULTIFOCAL LEUKOENCEPHALOPATHY IN THE SETTING OF IMMUNE RECONSTITUTION INFLAMMATORY SYNDROME

S PEERMOHAMED, O LARIOS
University of Saskatchewan, Saskatoon, SK

Progressive multifocal leukoencephalopathy (PML) is one of the most devastating opportunistic infections that can occur amongst the HIV-infected population. While the incidence of other opportunistic infections has decreased substantially because of advances in highly active antiretroviral therapy (HAART), PML continues to occur at a similar frequency amongst HIV patients. PML may actually worsen with HAART in the setting of immune reconstitution inflammatory syndrome (PML-IRIS), which is a paradoxical worsening of symptoms or infection despite a recovery of the immune system.

Here, we describe a 67 year old male who was started on HAART when his CD4 count and viral load were measured as 301 cells/mm^3 and 252,139 copies/mL, respectively. Six weeks later, his viral load decreased dramatically to 2240 copies/mL. He later developed progressive weakness in his extremities, increased spasticity, tone and myoclonic jerks. Magnetic resonance imaging revealed multiple, hyper-intense subcortical lesions in both frontoparietal lobes without mass effect. While examination of his cerebral spinal fluid was negative for the presence of the John Cunningham virus, a brain biopsy was performed and confirmed the diagnosis of PML with findings also consistent with IRIS. He was started on a course of steroids and transferred to a rehabilitation unit, where his symptoms continued to improve.

Further research is needed regarding the role of steroids in treating PML-IRIS and further characterizing this disease. Improvements in HIV viral loads following HAART and the development of neurological symptoms should raise suspicion of an underlying infection and the diagnosis of PML-IRIS should be entertained.

SP45

ESTIMATED RATE OF TREATMENT FAILURE IN FIRST-LINE ANTIRETROVIRAL TREATMENT IN KINSHASA: CASE OF ACS-AMO CONGO

E KAMANGU[1], E KAWILA[2], H MUKUMBI[2], G MVUMBI[1]
[1]University of Kinshasa, Kinshasa, Congo; [2]Acs Amo-Congo, Kinshasa, Congo

BACKGROUND: In the Democratic Republic of Congo (DRC), the first line of treatment for HIV adopted since 2008 for the PLHIV is a combination of single-dose Azydothimidine (AZT), Lamivudine (3TC) and Nevirapine (NVP). In 2009, 34,947 PLHIV were on the first line HAART in the DRC, of which 28,918 or 82.75% were adults.

OBJECTIVE: Estimate the rate of treatment failure in first-line antiretroviral therapy in treatment centers in Kinshasa.

METHODS: This study is a cross-sectional study in three outpatient treatment centers of the NGO ACS AMO-Congo in the city of Kinshasa in the Democratic Republic of Congo in January 2009. Any patient on antiretroviral therapy in first intension followed regularly in one of three centers of study for more than six months in January 2009 was considered. Blood sampling was done in a tube with EDTA anticoagulant, from a venous puncture. The numbering of CD4 has been made in the laboratory of ACS/AMO-Congo Kasa Vubu on FACSCount™ and viral load at the National Reference Laboratory AIDS / STI (LNRS) by NucliSens Easy Q - HIV1, Version 1, 2. Data were entered using Excel and SPSS software. The Student test was used for quantitative variables and Chi-square (X2) for

categorical variables. The significance (p) was chosen for the probability of $p<0.05$. Results are expressed as mean ± standard deviation. The tables have been reformatted in Excel. Respect for the individual and the confidentiality of records were found.

RESULTS: A total of 102 patients were included in this work with a female predominance (66.7%) and a mean age of 41.4±9.4 years. The mean CD4 count of third control (395.5±145.2 cells/L) were significantly higher than those of CD4 at baseline (252.1±128.7 cells/L). Controls were performed CD4 at 1 month (control 1), 3 months (control 2) and ± 6 months (control 3). The viral load (VL) average of 20,258.3±10,209.0 RNA copies/mL. Eighteen patients (17.6%) had a CD4 count lower than the third control values before treatment, 16 patients (15.7%) had a viral load above 1000 RNA copies/mL and 7 patients (6.9%) evolved at the AIDS stage.

CONCLUSION: The estimated rate of treatment failure of patients on ARV first line in Kinshasa conducted in three treatment centers for AMO-Congo gave a rate of 17.6%.

AUTHOR INDEX

A

Abed Y SP37
Adam H D3,P45
Adam HJ P12,P43,SP14,SP16
Agarwal K A5
Ahmed R D4
Akani C P41
Al Hashami H SP01
Al Nahdi N SP05
Alenazi T B5
Alfa M E3,G2,P21,P43,SP43
Al-Ghazawi F A1
Allen R F6
Allen V D4,H2,SP08,SP24
Allman D P41
Almohri H E5,P22,P24,P36
Al-Rawahi G J3
Al-Siyabi T SP35,SP36
Amihod B G6
Ang L D4
Angel J A1
Arnott W C1,C3
Auk B F2,I1

B

Baba K P33
Babtunde S P41
Ball TB B4,SP41
Barkati S K1,SP18
Barrowman N P31
Baxter M D3,P12,P45,SP14
Beaudoin M IA2
Beaudry R P26
Bechard C E5
Beirnes J SP38
Bekal S D4
Belanger P P09
Bélanger S K1,SP11
Bélanger Savary S SP18
Belhaj A E5,P22
Bell C H1,H3,P08
Belzile N SP11
Bennell MC SP13
Bergeron MG I3
Bernard K B3,J4,P02,P34
Bertrand C G5
Bezaire C P30
Bichoupan K P04
Binkhamis K SP35,SP36
Bitnun A SP01
Blanchette R P15
Blondeau J P14
Blonski S P28
Boerlin P SP15
Boivin G I3,I4,SP37,SP39
Boivin N SP39
Bouhy X SP37
Bourgault A D1
Bowes J P31
Boyd D C2,E2
Boyd DA D5,G4,P17,P35
Bramberger D E5
Branch AD P04
Braun K B4,SP41
Bright AT B5
Brinkley S P04
Brooks A H8
Brooks J P41
Broukhanski G C4,P35
Bryce E D5
Buckeridge DL SP22
Buckwalter S SP42
Bull A P45
Buller-Taylor T K3
Burdz T J4
Burry L H1
Butler G SP38
Butt G K2,K3

C

Campsall P IA3
Canadian Nosocomial Infection Surveillance Program (Cnisp) . . . C2,D2,SP19
Cantin P G8
Carbonneau J I3,I4
Carey A E8
Carrasco-Medina L A1
Chahil N A4
Chalmers G SP15
Chambers C P16
Chang Y SP38
Charest H F1,G8
Charles M SP09
Chaubey V IA3
Cheng L SP26
Cheng SYL P09
Chiao-Ling Kuo J F7
Childs K A5
Chinenye S P41
Chirip VN P42
Chong M P16
Chong P F4
Chong PM SP32
Christianson S E4,SP43
Chui L F6,F7,IA5,J2,SP27
Ciccotelli W SP15
Coady A J1
Coburn B SP23
Collignon P SP17
Conly J IA3
Cook D K2,K3
Corbeil J F1,G8
Costiniuk C A1
Cote S A1
Courtemanche J J1
Coyle D SP19
Craig J G5

D

Dale SE P40
Daley P E7
Dalgleish S G5
Dalton B IA3
Dandle A P05
Daneman N H2,IA4,SP23,SP29
Daniel J E8
Darwish I G7
Degagne P G2,P21
Degagné P P43
Demczuk W K5
Denisuik AJ SP14
Denisuk AJ P12
Desai S K5,SP38
Desbiens F F1,G8
DeSerres G I3
Desjardins M P42,SP21
Desouza E G5
Devlin R E1,SP08
Dewhurst N H6,H7
Dhurat R P05
Dieterich DT A5,P03,P04
Dion R G8
Doyle P SP05
Dresser L H3,H4,H6,H7,P08
Drews SJ J2
Dromanraju D A5
Du T C2,F3,P35
Dufresne P K4
Dufresne S K4,SP11
Dunbar J P39
Duplisea K H4,P08

E

Edagiz S E3
Edwards I B3
Eisbrenner T SP38
Eisler D F2
ElBailey S D4
Elligsen M H2,IA4,SP23
Ellis C P31,SP21
El-Sherif M F5
Embil J D2,SP32
Embree J D5,SP33
Evans G P09,SP26

F

Fanella S SP33
Fathima S J2
Faucher S F1
Fernando K F8,P10
Ferrato C J2
Fiel M A5
Filion F SP18
Finley R D4
Fisher G G5
Fleming F SP33

Fonseca K P39,SP38
Fontela PS SP22
Fortin C D1
Fortin E SP22
Forward K SP28
Fowler S SP07
Frenette C C2,D2,F3,G6,SP32
Fuller J D3,E6,P45
Fullerton J G5
Fung J F2,I1
Fung T G4
Furness C G1

G

Gaisa M P03
Galarneau A P14
Galbraith JC SP17
Gall R J4
Gallant V J1
Gandhi B E5,P46
Gardam M G1,IA1
Gardner E SP13,SP26
German G P02
Gerson M C1
Gerull-Upton A E8
Ghaly A SP28
Ghate S P05
Ghazy NH P01
Ghosh A SP10
Gilbert E P20
Gilbert L SP38
Gill Y E7
Gilmour M D4,K5
Gitterman L IA1
Gold W SP29
Golding G P12,SP19,SP33
Golding GR D2,SP32
Goodliffe L SP15
Goodman C SP25
Gorbach S C1,C3
Gosselin J SP39
Goupil-Sormany I F1,G8
Graham M F3,SP33
Granados A SP34
Grant C SP38
Grant J H5
Gratrix J SP02
Gravel D C2,D2,D5,F3,F4,G6,SP19,SP32
Gregson D IA3
Griffith A K5
Grimard D P14
Guenette T SP02
Gunadasa K P37

H

Haider S H8
Haldane D D4,P02,P20
Hall G P09,SP26
Hamandi B H6,H7
Hamelin ME I3,I4
Haraoui L P15
Harris C H1
Hasan M J3
Hasing E I2
Hatchette T F5,SP35,SP36
Hendson L SP02
Hiebert J SP38
Hizon R C2
Hoang L D4,F2,J4
Hoban D D3,P14,P45
Hoban DJ P12,SP14
Hoban S P21
Holmes L C2,F3
Hoosen AA P33
Horsman G D4
Houston S B5
Hsiao W F2
Hudym E P28
Hui C P31
Hum C G4
Huot C G8
Husain S P07
Hussain Z E1,P17
Hussein H SP03

I

Igbigbi E P41
Imperial M F2,P17
Isaac-Renton J I1,P11,SP38
Isaac-Renton JL F2
Isabel S I3

J

Jabbour M P31
Jacobsson G SP17
Jansen B E6
Jayaratne P P40
Jiao L SP04,SP31
Jissam A SP31
Jivraj T H1,H4,P07,P08
Jiwa S D3,E6,P45
Joyce A P09,SP13,SP26
Jutras P P14

K

Kaki R G3
Kamangu E SP45
Kanti P SP36
Kao D H5
Karlowsky J E3,SP43
Karlowsky JA P12
Karnauchow T P38
Katz K C2,D2,D5,F3,G6,SP32
Kawila E SP45
Kearny A SP25
Keen P H5
Kennedy KJ SP17
Kern C P38
Khalil N SP38
Kiazyk S B4,SP41
Kibsey P C2,D2,D5,F3,F4,SP32
Kissoon M P27
Knox N F3,SP33
Knudsen JD SP17
Kong C I1
Kornherr P P26
Kothavade R P05
Kozun B SP20
Krajden M A4,K2,K3,P37,SP38
Krolik J P09,SP12,SP26
Kuhn M D5
Kushner T A5,P03

L

Labbé A D1,SP11,SP18
Lachance L SP38
Lagacé-Wiens P E3,P43
Lagacé-Wiens PRS SP14
Lambert G D1
Lang A F5
Langill J P31
Langner S D4
Lapinsky S H1,H4,P07,P08
Larcombe L B4
Larios O SP44
Larocque M G4
Lau J P24
Laupland KB SP17
Laverdière M K1,SP11
Lavergne V SP18
Lazar N H4,P08
Lebel M SP39
Leblanc J C5,F5,SP35,SP36
Lee B IA5
Lee BE I2,J2
Lee C C1,P14,P29,SP23
Lee M F8,P10
Lefebvre B D1,K5,P15,P17
Leifso K SP40
Leis J SP29
Lekalakala MR P33
Levebvre B SP18
Levesque S P15
Lévesque S F1,G8
Li A I1
Li L I1
Li V IA5,J2
Li Y SP38
Lin A H6,H7
Lipskie T SP38
Lo P B2,B3,G4,P25,P30,P35
Lo T P11
Locke T SP19
Loeb M D2,SP32
Loo V P14
Lopez C B4,SP41
Loughborough A P27
Louie M D4
Louie T C1,C3

Loungnarath V. SP18
Low D. SP29
Low DE. B3,G4,P27
Lowe C. SP08
Lowe D. H6,H7
Lowther N P37
Luong M. SP11
Lynch T F3,F4
Lynkowski L P18
Lyytikäinen O SP17

M

Mabon P. F3
Macdonald K SP38
Macdonald KA. F2
Macey J. SP38
Mackenzie L. SP16
Macpherson M. P40
Mahmoud Ahmed S P01
Mahony J SP34
Mailman T P20
Main C. P29,SP04,SP31
Majury A P09,SP12,SP13,SP26
Mak A A4
Man S. F8,P10
Manges AR SP22
Marchand G. G8
Marlatt F P41
Marra F. P16
Martel-Laferriere V A5,P03,P04
Martin I K5
Mataseje L G4
Mataseje LF D5,P15,P17
May-Hadford J SP38
Mazzulli A G4,P25
Mcauley J K2
Mccabe MN F2
Mcconnell A SP20
Mcconnell M P29,SP31
Mccorrister S F4
Mccracken M. D4,P12
Mccready WG P19
Mcgeer A C2,D2,F3,G6,P27,SP29,SP32
Mcgeer AJ G4
Mcgeer AM P35
Mcintyre S SP08
Mcnabb A F2,P37
Mcneil S. F5
Measures L P10
Melano R SP24
Menasria R. SP39
Mendis N F1
Mertz D G3,G5,H8
Meyers T P41
Mian F A2
Midodzi W E7
Milburn S. E8
Miller L C4
Miller M. C2,F3,F4,P15
Mirzanejad Y P06
Misir A. P31
Missaghi B IA3
Mitchell R G6
Mohiuddin Q. H1
Moore D. C2,F3
Morris A. H1,H3,H4,P07,P08
Morshed M. F8,P10,P11
Morsi M P01
Mounchili A D2,D5,SP19,SP32
Mubareka S A2,H2
Mukhi S IA5,SP38
Mukhi SN J2
Mukumbi H SP45
Mullen M. P03
Muller M SP08
Mulvey MR C2,D2,D4,D5,E2,F3,F4,G4,
. . . . . G7,P15,P17,P35,SP14,SP32,SP33
Musa J. E5
Mustachi B. P30
Mvumbi G SP45

N

Nadon C. SP25
Nashid N SP34
Nault V IA2
Naus M. SP38
Nelson S. H1,H4,P08
Nevala J J1
Ng B. J4,P02
Ng M G4,P03,P25
Nguyen L P26
Nichol K. P43
Nigh S E5
Nix EB P19
Nolt S. SP38
Noorpour L. E5
Nuri K G5

O

O'Keefe C. SP38
O'Neill C G3,G5
Obunge O. P41
Okasha HS. P01
Olson N G2,P21
Orr P. B4
Ostergaard C SP17

P

Pabbaraju K A3,P39
Paccagnella A F2
Pacheco AL J4,P34
Paganotti G B5
Palmay L. H2,IA4,SP23
Pan B IA5
Pang XL I2
Papenburg J I3
Parfitt E IA3
Parikh N. A5,P03
Patel S P27
Patrick D H5,P16
Payne M SP30
Peermohamed S SP44
Peltsch P. G5
Pelude L SP19
Pernica JM SP34
Perron J IA2
Perry P G5
Perumalswami P. P04
Peterson C SP25
Peterson S SP33
Petkau A F3
Petric M SP38
Petrich A B1
Pinto R. H2
Pipkin T G4
Piret J SP39
Pitout JD P14
Pitre M H6,H7
Pivnick L P07
Pizarro R. P26
Pizzorno A SP37
Plante P F1,G8
Platt RW. SP22
Plitt S SP02
Poliquin G SP16
Poon A. SP27
Posner S P04
Pouliot J P14
Poutanen S. H4,P08
Poutanen SM B2,B3,G4,H6,H7,
. . . . . P14,P25,P27,P30,P35
Pradinuk J. F2
Prayitno N P25
Preiksaitis JK I2
Prystajecky N F2,I1
Purewal K. P22
Purych D P16

Q

Quach C. SP19,SP22
Qureshi R. SP08

R

Rachlis A H2
Ramji A SP20
Randhawa K. P09,SP12
Raymond F. F1
Rea M. SP32
Reason P. IA1
Rendina A P43
Rennie R D3,P14,P28,P32,P45
Richardson S G7,P46
Robberts L D4,SP38
Robinson J SP02
Robinson R SP21
Robitaille L F1
Romney M P11
Roscoe D D5,P14,P23,SP30
Ross T. IA3

Roussy JF I4
Rumore J SP25
Rutherford C P40

S

Sahni V SP38
Sallene W. SP36
Sandstrum P. P41
Saxinger L H5,P18,SP09
Scales D H3
Schertzberg R. SP15
Schiano TD A5,P04
Schønheyder HC. SP17
Schwartz K G7
Sekirov I. P23,SP05
Senn H. SP07
Sepehri S SP43
Severini A SP38
Shah K SP10
Shane A SP38
Sharma M. B4,E4,SP07,SP41,SP43
Shaw L P38
Sherrard L K5
Shojania K SP29
Shokoples S B5,D3,E6,P45
Showler A SP03,SP08
Shurgold J. D2
Silverman M P41
Simner P. SP42
Simner PJ SP14
Simor A C2,D5,F3,SP19,SP23
Simor AE D2,SP32
Singh A SP02
Singh J H4,P08
Singhal N. SP10
Slinger R P31,SP40
Small G B3,P30
Small W E1
Smieja M P29,SP34
Smith S D2,P22,P24,P36,SP09,SP32
So M. H3,P07
Søgaard M SP17
Somily A P44
Srigley JA. G1,H8
St Jean T SP38
Steinberg M H1,H3,H4,P07,P08
Stivala A P04
Su SH. SP18
Suh K C2,F3,G6
Sulkowski M. P04
Sullivan L. P38
Sung M. H8

T

Tabing C. SP25
Tabor H D4
Tadros M B1,E1,SP24
Tam B. E1
Tan DHS SP03,SP07
Tan R J3
Tang P F2,P37
Tarning J. B5
Taylor C A5
Taylor G C2,D5,F3,G6
Tellier R A3,I2,P39,SP36
Thampi N. G7
Thomas E D5,J3
Thompson K SP28
Tilley P J3
Tipples G SP38
Tom S. SP17
Tomkinson M. P15
Toye B P14,SP21
Tremblay C. D1,F1,G8,K4
Tsang A G4,P25
Tschetter L SP25
Turgeon N D5
Turnbull L D3,P32

U

Ulanova M. P19

V

Vachon ML A5
Vadnais B K1
Valiquette L C3,IA2,SP17,SP18
Van Caeseele P. J4,P17
Van Domselaar G. F3,SP33
Van Zuilen L P24
Vanderlaan J. SP15
Varga L H1
Venne S D1
Verghese V SP02
Villeneuve J G8
Voth D E4

W

Walker S. H2,IA4,SP23
Walkty A E3,SP16
Walley VM. P36
Webber CG G5
Weese S P35
Weiss K. C1,C3,D2,SP18,SP32
Wengenack N SP42
Westmacott G F4
White L P36
White N. B5
Wiebe D J4
Wilcox M SP36
Wilkinson K. G6
Willey B P17
Willey BM B2,B3,G4,P25,P27,P30,P35
Williams JV I3
Wilton J SP07
Winzeler E B5
Wolfe J B4,E4,SP41
Wong A G6,P39
Wong B I1
Wong H A2
Wong K B3,P25
Wong Q P11
Wong S. A3,P39
Wong T J1
Wylie J D4

Y

Yamamura D. H8,P29
Yan L IA5,J2
Yanow S B5
Yarmoshuk A P41
Yoshida E SP05
Young C A1
Yu A A4,K3
Yu Y G3,SP31

Z

Zarra D A3
Zelyas N SP27
Zhanel G P45
Zhanel GG. D3,P12,SP14
Zhu L P26
Zittermann S SP24