LAB Connections

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HAMILTON REGIONAL LABORATORY MEDICINE PROGRAM

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As co-editors of the HRLMP newsletter, we would like to say a sincere **thank you** to **Dr. Elizabeth McCready** for her work and commitment to the HRLMP Lab Connections as an active member on our Editorial Board. Dr. McCready stepped down from this role this past July.

We would also like to **welcome Dr. Daria Grafodatskaya** to our Editorial Board, as she has taken over this role from Dr. McCready. We are looking forward to working with Dr. Grafodatskaya and are pleased that she has joined our team.

Dr. Cathy Ross and Karen Moffat

A Novel Isothermal DNA Amplification Method for Detecting Group B Streptococci (Streptococcus agalactiae) in Pregnant Women at the HRLMP

Group B streptococcal (GBS) infection carries a high morbidity and mortality rate, especially amongst newborn babies. It has been a leading cause of septicemia and meningitis in neonates and infants <3 months of age since the 1970's [1]. Infants acquire the infection vertically or during labor from colonized mothers.

GBS infected infants have a mortality rate of 5% and those who survive can develop neurological problems such as visual or auditory dysfunction and cognitive dysfunction [2]. Currently, there is no vaccine available to prevent the infection [3]. Fortunately, Intrapartum antibiotic prophylaxis in mothers colonized with GBS has been shown to reduce neonatal infection, and is currently recommended by Centers for Disease Control and Prevention (CDC) [4].

Current guidelines recommend that all pregnant women be screened for vaginal/rectal colonization of S. agalactiae (GBS) at 35-37 weeks of gestation. The CDC recommends the use of enrichment broth regardless of the method used to identify GBS to enhance the detection, based on a number of studies where they compared the sensitivities with and without enrichment in Nucleic Acid Amplification Tests(NAAT). They found sensitivity increased by approximately 10%, to 92.5%- 100%, with the enrichment compared to without [5].The conventional detection of GBS in the anogenital tract of pregnant women involves overnight broth enrichment followed by culture. In our lab, prior to 2016, we used a culture based method for GBS identification. Initially we used LIM enrichment broth overnight and then plate them on the CHROMagar which are left overnight as well. The CHROMagar plates were checked the following day and further testing done for identification, with a total culture turn-around-time of 48-72 hours. Now there are a number of nucleic acid-based commercial tests based on polymerase chain reaction (PCR) and loop-mediated isothermal amplification (LAMP) with much shorter turn-around-times (TAT) and better test performances than culture. However, they are significantly more expensive (approximately $25 to $45 per test) than culture to use in routine screening. Since first reported in 2000 [6] LAMP has been recognized as an isothermal target amplification method that is robust, faster and cheaper than PCR. Unlike PCR, LAMP does not require DNA purification hence reduces TAT and cost of nucleic acid extraction. A simple cell lysis by boiling a liquid specimen for 10 minutes with or without a lysis solution is sufficient to release nucleic acid to obtain a crude preparation of DNA for amplification. LAMP has been used to detect a number of other etiologic agents including bacteria, fungi, parasites and viruses. In our laboratory, LAMP technology is also used to detect toxigenic Clostridium difficile, culture confirmation of vancomycin-resistant enterococci (VRE) and methicillin-resistant Staphylococcus aureus (MRSA), and Streptococcus pyogenes (Group A streptococci) from throat swabs.

With this in mind, the Molecular Microbiology Section of the HRLMP developed a new Loop-Mediated Isothermal Amplification (LAMP) method for the detection of GBS. This novel method targets cfb gene (cAMP protein) of S. agalactiae. Specimens are enriched in LIM broth overnight (≥ 11 hours). The enrichment time was selected based on the laboratory work-flow by comparing it to the CDC recommended time (≥18 hours) for equal test performance. Comparison of the test performance of LAMP with broth enrichment showed a 10% higher sensitivity of GBS detection than direct testing, or conventional culture methods. To optimize accuracy for GBS detection, our final protocol incorporates overnight enrichment followed by LAMP testing. The TAT for the new method is about 12- 24 hours, depending on time of specimen receipt, as compared to 48 to 72 hours for conventional LIM/culture.

The test performance characteristics for LIM/LAMP as compared to LIM/culture were 99.2% sensitivity, 97.5% specificity, 91.5% positive predictive value (PPV), and 99.8% negative predictive value (NPV). The cost for LAMP is similar to culture (<$10) and the limit of detection (LOD) for GBS detection by LAMP was calculated to be 8.8x103 CFU/ml (44 CFU/LAMP reaction). This method has undergone validation and was introduced into the routine laboratory testing at the St. Joseph's Healthcare site of HRLMP in January 2016. Since then the laboratory has been testing approximately 250 to 300 specimens for GBS per month. The Genie® II (OptiGene, UK) real-time fluorescence detection instrument or any other Real-Time PCR instrument such as Rotor-Gene Q (Qiagen Co.) can be used for LAMP amplification and detection.

Our new method is rapid, highly sensitive, and cost-neutral compared with our previous methods. However, without culture, we cannot routinely provide antibiotic susceptibility results. Since GBS is

universally susceptible to penicillin, susceptibility results are only required for patients allergic to penicillin, in whom culture will be performed to determine susceptibilities to other antibiotics.

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References

1. Heath, P.T., Balfour, G., Weisner, A.M., et al. (2004): Group B streptococcal disease in UK and Irish infants younger than 90 days. Lancet, 363, 292–294.
2. Schrag, S., Gorwitz, R., Fulz-Butts, K., et al. (2002): Prevention of perinatal group B streptococcal disease. Morbid. Mortal. Wkly. Rep., 51 (RR-11), 1–22.
3. Edwards, M.S. (2008): Group B streptococcal conjugate vaccine. A timely concept for which the time has come. Hum. Vaccin., 4, 444–448.
4. Boyer KM, Gotoff SP. Prevention of early-onset neonatal group B streptococcal disease with selective intrapartum chemoprophylaxis. N Engl J Med 1986;314:1665–9.
5. Davies HD, Miller MA, Faro S, Gregson D, Kehl SC, Jordan JA. Multicenter study of a rapid molecular-based assay for the diagnosis of group B Streptococcus colonization in pregnant women. Clin Infect Dis 2004;39:1129–35.
6. Notomi T, Loop Mediated Amplification of DNA. Nucleic Acid research, 2000, vol. 28, no. 12.

News from HRLMP

News from Chemistry

An Example of a successful Utilization Initiative: ESR and CRP Testing

Situation:
In keeping with the ***Choosing Wisely*** campaign, the Hamilton Regional Laboratory Medicine Program (HRLMP) has embarked on an effort to reduce inappropriate testing of erythrocyte sedimentation rate (ESR). The aim was to help raise awareness about the clinical utility and best practice guidelines around ESR versus C-reactive protein (CRP) testing.

Background:
ESR and CRP are widely used markers of infection/inflammation. However, CRP has numerous advantages over ESR, including:

- CRP is an automated test with a lower cost and considerably faster turnaround;
- CRP is a more accurate test and is not affected by certain medical conditions, drugs, and smoking;
- CRP is a more sensitive marker for infection/inflammation as it rises and falls more naturally with the course of disease.

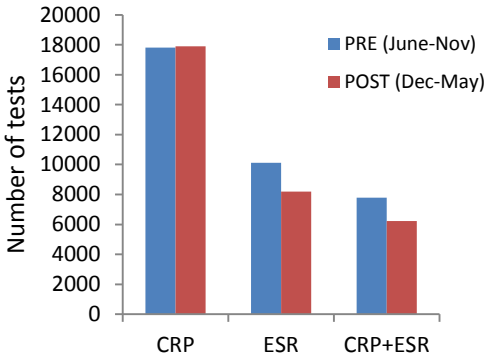
Intervention
The HRLMP informed health care providers within the Hamilton Hospitals of the clinical utility and best practice guidelines around ESR versus CRP testing. The results of both ESR and CRP testing were examined six months before and six months after dissemination of the information (see figure 1).

Analysis:
Six months following dissemination of the information, the following outcomes were noted:

- 19% reduction in overall ESR testing;
- 20% reduction in combined ESR and CRP testing.

Conclusions:

- HRLMP strongly advocates the use of CRP in the majority of clinical situations;
- CRP is a more accurate test, has a faster turnaround time, and lower cost;
- This initiative is an excellent starting point to reinforce appropriate testing of ESR and CRP;
- The HRLMP will continue to monitor the results of this study and maintain good evidence-based ordering of laboratory tests.



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References:

1. CADTH Health Technology Assessment, No. 140: Canadian Agency for Drugs and Technologies in Health; 2015 Nov:1-66.
2. Ontario Association of Medical Laboratories. Guideline for the ordering of erythrocyte Sedimentation Rate (ESR). Revised 2001. [Guidelines for clinical laboratory practice]. Retrieved November 19, 2010 from OAML web site at http://www.oaml.com/PDF/CLP023.pdf
3. Olshaker JS, Jerrard DA. The erythrocyte sedimentation rate. J Emerg Med 1997;15(6):869-874.
4. Lane SK, Gravel JW Jr. Clinical utility of common serum rheumatologic tests. Am Fam Physician 2002;65(6):1073-1080.
5. An HS, Seldomridge JA. Spinal Infections. Diagnostic tests and imaging studies. Clin Orthop Relat Res 2006;444:27-33.
6. Colombet I, Pouchot J, Kronz V, et al. Agreement between erythrocyte sedimentation rate and C-reactive protein in hospital practice. Am J Med. 2010 Sep;123(9):863.e7-13.

Education News

Medical Laboratory Science was recently "*in the news*" …

Click on the link below to read a recent article in the Globe and Mail from the perspective of a MLT working on Prince Edward Island:

http://www.theglobeandmail.com/life/health-and-fitness/health/what-its-like-to-identify-cancer-in-a-childs-blood-sample/article30787647/

Keep up the great work!

The **9th Annual HRLMP Rapid Fire Showcase** will be held on **Saturday, November 26, 2016** from 0815 – 1215 at St. Joseph's Healthcare, Hamilton.

This free half-day CE event provides an excellent opportunity to meet colleagues both internal and external to the HRLMP and to learn about the great work that happens across our program!

Registration details will be available soon.

We look forward to seeing you November 26th, 2016.

News from Genetics

Congratulations to our CCMG fellows!

We would like to congratulate our Canadian College of Medical Genetics (CCMG) fellows **Lauren MacNeil**, **Hanxin Lin** and **Daria Grafodatskaya** on successful completion of their training!

Lauren has accepted the position of Biochemical Geneticist at Hospital for Sick Children.
Hanxin is now working as a molecular geneticist at London Health Sciences Centre, and Daria has accepted a position in our Molecular Oncology laboratory.

These former trainees would like to thank the CCMG program director Dr. Margaret Nowaczyk, training directors and training committee members: Dr. Elizabeth McCready, Dr. Murray Potter, Dr. John Waye and Dr. Mariya Kozenko, as well as all professional, laboratory and clinical staff for making this training program a success!

We also would like to announce the retirement of our medical laboratory technologist, **Maria Harvey**. Maria joined the Molecular Oncology laboratory in 2002 after working for multiple years in research in Pathology at McMaster University. In the molecular oncology lab, Maria performed molecular diagnostic tests for hematological cancers and solid tumors. Her colleagues describe Maria as attentive to detail, kind and always ready to help. We will miss Maria and wish her a happy retirement!

Last but not least, we would also like to announce that our medical laboratory assistant, **Carolyn Carrol** has retired after many years with the HRLMP. We will miss Carolyn and wish her well as she starts this new chapter in her life!

Microbiology News

We're pleased to announce that effective September 1, 2016, **Dr. Daniela Leto** takes over as **Program Director** for McMaster University's Medical Microbiology program. We wish Dr. Leto all the best as she transitions into this role!

CONGRATULATIONS to **Dr. Lei Jiao** and **Dr. Fatimah Al Mutawa.**

Dr. Jiao, a resident in Medical Microbiology, was recently awarded a ***Quality Assurance Award*** through McMaster University. The selection committee was impressed by the quality of work of Dr. Jiao's project, titled "*How many bronchoalveolar lavage specimens should we collect*".

In addition, **Dr. Al Mutawa,** another Medical Microbiology resident, was recently awarded the ***Harry Richardson Quality Management Award for Medical Microbiology*** through McMaster University.

News from Pathology

Congratulations to **Dr. Monalisa Sur**.

Dr. Sur won the Canadian Association of Pathologists prestigious **National Award for Leadership in Education 2016**.

It was presented during the Annual CAP-ACP meeting in Vancouver, BC on July 12, 2016.

We are all very proud to have members of our Anatomical Pathology Program recognized at the National level.

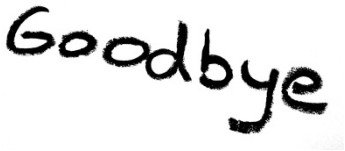
In addition;

Congratulations to our Anatomical Pathology residents, **Dr. Paul Borowy-Borowski** and **Dr. Ian Brain,** and their supervisors Dr. Ross and Dr. Sur on receiving national awards at the recent Canadian Association of Pathologists Annual Meeting in Vancouver!

Awards

Donald W Penner award for best poster presentation – Dr. Paul Borowy-Borowski, PGY5 AP.

Award for best presentation Hematological section – Dr. Ian Brain, PGY3 AP.



Dr. Allison Edgecombe will be departing HRLMP to assume a position in forensic pathology at the University of Ottawa.

Thanks for everything Allison!

We are thrilled to announce that **Dr. Jane Turner** has joined the forensic pathology unit as full-time staff with an academic appointment as Professor.

Dr. Turner has relocated from Saint Louis University, Missouri, where she was Program Director in forensic pathology. Dr. Turner brings many years of

experience in forensics to the Hamilton Forensics Unit.

Dr. Turner started with the HRLMP August 1, 2016 and replaces the retiring Dr. Rao.

Please join us in a warm welcome to Dr. Turner.

Quality News

Thank you to **Teri Johnson** for her work as a HRLMP Quality Specialist. Her commitment to continued quality within the HRLMP is greatly appreciated.

Please welcome **Michael McConnell** on his return to the HRLMP as our new Quality Specialist.

All the best to both Teri and Mike as they transition to their new roles with the HRLMP!

Dr. Cheryl Main and **Allahna Elahie** were recently featured in a ***HHS Share*** article for their continued work in clinical education and the importance of the "order of draw" for our laboratory samples.

Click on the link below to read the full story:

http://hhsshare.ca/2016/08/order-of-draw/