The Delaware Public Health Laboratory (DPHL) hosted its second bi-annual Laboratory Preparedness Advisory Committee (LPAC) on October 16, 2014. There was a full house of representatives from hospitals, state agencies, and the military. Discussions revolved around the impending flu season, an overview of mosquito surveillance by the Delaware Department of Natural Resources and Environmental Control (DNREC) as well as the current outbreak of Ebola in West Africa.

Emily Hanlin, manager of the Molecular Virology section of DPHL, provided information regarding the start of the 2014-15 influenza season. (See Emily’s article on page 3.)

A brief overview of the state’s mosquito control program was presented by Bill Meredith, Program Administrator for the Delaware Mosquito Control Section. Topics included why mosquito control is needed and how it is performed, and some basic aspects of larval and adult mosquito biology and ecology, including a quick survey of problematic breeding habitats. There was also a focus on mosquito-borne diseases and the problems they cause, including discussion of their modes of transmission and how their presence is detected for West Nile virus, eastern equine encephalitis, and chikungunya. Chikungunya is a newly-emerging infectious disease in the Western Hemisphere.

Rick Hong, MD, Medical Director of the Division of Public Health’s (DPH) Office of Preparedness, gave a presentation about Ebola Virus Disease. His presentation encompassed the collaboration between hospital preparedness to quickly identify and isolate a suspected Ebola patient and the public health response that would be initiated. Dr. Hong answered many questions from the audience and provided expertise on managing patient specimens for laboratory analysis.

Timothy R. Cooper, Director of the Office of Preparedness, gave an overview of preparedness activities, which included planning, training, evaluation and logistics. DPHL and the Emergency Medical Services and Preparedness Section work together to provide public health laboratory testing, which is one of the 15 Public Health Emergency Preparedness capabilities set forth by the Centers for Disease Control and Prevention (CDC). Public health laboratory testing includes rapid and conventional detection, characterization, confirmatory testing, data reporting, epidemiological investigative support, and laboratory networking to address actual or potential exposure to all hazards. Mr. Cooper provided updates on other capabilities such as: community preparedness and recovery; surge management; emergency public information and warnings; and distribution of pharmaceuticals.

DPHL asked each of the hospital sentinel laboratories to perform a risk assessment of their testing procedures involving blood and other patient specimens. The emphasis for testing focused on possible splash risks as Ebola is spread by contact, not aerosol. Just as in the past with HIV and Hepatitis, all patient specimens must be considered possibly infectious. An example of donning and doffing personal protective equipment (PPE) was given by Marion Fowler, MT(ASCP), Microbiologist II. Proper doffing of PPE is especially important to prevent contamination from patient bodily fluids which allow the

Continued page 2
LPAC meeting (continued from page 1)

Ebola virus to survive for a longer period of time outside of the body. The Delaware sentinel laboratory microbiology managers discussed how they would handle samples from a suspected or confirmed Ebola patient. Only essential testing will be performed to minimize any biosafety breaches. Malaria testing (another endemic disease in West Africa) will continue but will be set up in a biosafety cabinet. Several hospitals have point-of-care testing available for suspected or confirmed patients. Most of the labs are now using biosafety cabinets for specimen preparation since any body fluid could have the Ebola virus present. DPHL offered training in specimen shipping and handling to hospital labs in November 2014. The training covered how to properly package and ship Ebola-suspect specimens to CDC if needed.

As of November 24, 2014, DPHL was certified to test CDC-approved suspect patient samples for Ebola using the Laboratory Response Network Real Time Polymerase Chain Reaction Method.

Hospital Workflow Sequence for Possible Ebola Case

Suspect cases — contact Delaware Public Health Epidemiology at 888-295-5156 Monday - Friday: 8:30 a.m.- 4:30 p.m. or 302-744-4700 after 4:30 p.m., and on weekends and holidays.

Get approval and PUI number for testing from the CDC. This will be done by the State Public Health Epidemiologist.

If testing has been approved by CDC EOC — collect two purple top (EDTA) tubes. Have a certified shipper package the specimens in two separate Category A packages along with the shipper’s declaration. Send one to the Delaware Public Health Laboratory (DPHL) and hold the second for the CDC, if needed.

Each hospital has trained/certified shippers within their laboratory sections (most are in the microbiology or immunology groups). The DPHL specimen needs to be maintained at 2-8°C or on cold packs. The completed DPHL LIMS Requisition Form for testing needs to be included. The CDC sample should be stored at 2-8°C or frozen (whole EDTA in plastic tube) and shipped on cold packs. DPHL has staff who can assist shippers by phone if necessary at 302-223-1520.

Once the specimens are packaged per Category A guidance (IATA), to expedite transfer and diagnosis, a hospital courier or security staff will transfer the specimen box to the DPHL. At the same time, the hospital needs to contact DPHL at 302-223-1520 to advise the estimated time of arrival. This will allow DPHL to assemble the team that will analyze the specimen.

Isolation of a suspect case should already be in effect along with the use of appropriate Personal Protective Equipment (PPE).

Laboratory test results that are negative can be reported as a final negative. However, repeat testing may need to be done if samples are collected within 72 hours of the initial appearance of signs and symptoms.

If the laboratory results are presumptive positive for Ebola virus, DPH, CDC, and the hospital will coordinate by conference call to transfer the second package containing the purple top tube to the CDC for confirmation. This will be done by World Courier, Inc.
Molecular Diagnosis of Influenza Infection and Live Attenuated Vaccines: A Story

Emily Hanlin, Laboratory Manager

The inception of real-time Polymerase Chain Reaction (real-time PCR) testing for infectious diseases revolutionized modern diagnostic capabilities. Organisms that would normally take several days or weeks to culture can be now detected in as little as one hour. Multiplex real-time PCR assays allow for detection of multiple infectious diseases from one sample, in one well. Viable organisms are no longer needed, since real-time PCR amplifies an organism’s Ribonucleic Acid (RNA) or Deoxyribonucleic Acid (DNA). Real-time PCR is both sensitive (detecting as little as 1-10 copies of nucleic acid) and specific (detects sequences of nucleic acids that are specific to that organism). As described above, one might think that real-time PCR would be the perfect diagnostic method but it has limitations.

At the DPHL, the 2014-15 influenza season started a bit early, with the first positive influenza (flu) A, subtype H3N2, identified in a 2-year-old child from Sussex County on October 7, 2014. By October 20, 2014, three additional positive cases for flu A/H3N2 were found. By October 21, DPHL received a specimen from a local hospital requesting confirmation of the presence of flu B from another two year old child. The following day, DPHL received a second specimen from a 10-year-old child at a different hospital for confirmation of flu B. Both specimens were run for flu by real-time PCR — type A and B detection. The first specimen came up positive for both flu A and B while the second was positive only for flu B. DPHL assumed that there might have been some nucleic acid contamination of the first specimen. For this reason, the test was fully repeated. The same results appeared. Flu B genotyping was done for the second specimen. It came up positive for both lineages — i.e., B/Yamagata and B/Victoria. The first specimen was then reflexed to the flu A subtyping run as well as a flu B genotyping run. It came up positive for flu A/H3N2, influenza A/H1N1 (pandemic) and both flu B/Yamagata and flu B/Victoria. This raised the question, “With such low levels of circulating influenza in October, how did two children acquire co-infections of the flu strains?” The answer: “They didn’t.”

When scientists consulted the CDC’s package insert for the “Human Influenza Virus Real-Time RT-PCR Diagnostic Panel (CDC flu rRT-PCR dx panel)” it stated, “Specimen should be referred to CDC for further characterization; possible co-infection or Live Attenuated Influenza Vaccine (LAIV) detection.” In looking at the ages of the two children, it was theorized that the children had probably received LAIV, better known as “FluMist.” FluMist is the recommended influenza vaccine for healthy children ages 2-8, and comes in the form of a nasal spray. The specimens received from the two children were a nasal swab and a nasopharyngeal wash.

While the Molecular Virology (MV) Laboratory prepared to ship the specimens to CDC for a specific method that targets the LAIV nucleic acids (and is not available at DPHL), the influenza Epidemiologist reached out to the hospitals to determine if FluMist had been administered. Through a medical records search, it was found that the first patient had received FluMist three days prior to specimen collection. The second patient had received FluMist four days prior to specimen collection. CDC testing confirmed the presence of FluMist, which proved that the children were not infected with multiple flu strains. The rRT-PCR methods at both of the hospitals and at DPHL had detected residual live vaccine from the children’s nasopharynx. The flu vaccine (including FluMist) does not cause flu. Following these two specimens, DPHL received two more LAIV positive rRT-PCR specimens from a 9-year-old and an 11-year-old. Both were confirmed by CDC.

So what does this mean for real-time PCR testing? While real-time PCR testing is an excellent method for diagnosing infections, the presence of RNA or DNA may not necessarily be indicative of active infection. In these two cases, clinicians were not made aware of the fact that the children had received FluMist prior to the collection of specimens for testing. Other Live Attenuated Vaccines that may also produce erroneous real-time PCR results include: Chicken Pox, Shingles (Varicella zoster); Measles, Mumps, Rubella (MMR); Vaccinia (smallpox); Rotavirus; Yellow Fever (Flaviviruses); and Typhoid fever (Salmonella Typhi or paratyphi)?

It is important to inform an attending physician and practitioner if a vaccine has been recently received, particularly if some ill effect or discomfort was felt. Often, practitioners who provide service in an emergency setting are not the same as those who provide routine medical care. In effect, there is no “perfect” test for diagnosing infectious diseases. Often it is only by providing the necessary pertinent information, such as recent vaccinations, that prevent inaccurate diagnoses.

References:
On November 12 and 13, 2014, the State Health Operations Center (SHOC) assisted the Delaware Emergency Management Agency (DEMA) with a graded exercise outlining hurricane emergency preparedness. This exercise simulated a seven to eight day post-recovery to a Category 2 Hurricane (Hurricane Merlin). The SHOC planning section took advantage of the opportunity to exercise a drinking water perspective of an adverse natural event.

The SHOC planning section coordinated communications to planning section staff that traveled to various locations throughout Delaware to collect water samples. The water samples represented both public and private drinking water systems “impacted” during hurricane landfall and aftermath. Also, water samples were collected from the emergency water purification system that the SHOC logistics section set up to obtain filtered water for those that may need potable drinking water during an emergency. All water samples were collected and transported to the Delaware Public Health Laboratory (DPHL). The lab received the samples, verified the chain-of-custody, and performed total coliforms and Escherichia coli (E. coli) analyses.

All water samples collected, including those collected from the emergency water purification system, were on an 18-24 hour turnaround-time. All samples reached the lab by 1430-hours (2:30pm), on November 12, 2014. All samples were analyzed and results reported out by 0910-hours (9:10am), on November 13, 2014. All results reported negative for total coliforms and E. coli. Utilization of the Office of Drinking Water, Community Environmental Health Services, and DPHL staff helped to facilitate a timely, efficient, and communicable response to ensure that the public would know whether their water would be safe to drink during a natural disaster.
Preparedness Suites: A Retrospective
Margaret Zimmerman, ASQ CQA, Laboratory Manager

In an unassuming brick building, on the campus of the Delaware Hospital for the Chronically Ill outside of Smyrna, DE, are two laboratories that stand at the ready to serve the citizens of Delaware at a moment’s notice. These laboratories, based within the Delaware Public Health Laboratory (DPHL), process biological or chemical testing when the need arises during an outbreak, chemical incident, or national disaster. The laboratories are members of the Laboratory Response Network (LRN) and serve as reference laboratories within that network. Within the building at 30 Sunnyside Road, these laboratories are known by the monikers Biological Preparedness Laboratory (BPL) and the Chemical Preparedness Laboratory (CPL).

In 2004, construction was started on a suite to house the newly ordained CPL and BPL. These new suites were designed to house state-of-the-art instrumentation for chemical analyses; a clean room, an anteroom, multiple biosafety hoods, glove boxes, chemical safety hoods, a pass-through autoclave, sub-zero freezers, and refrigerators. Over the next decade, extraction equipment and additional analytical instrumentation would be added as the complex testing methods were updated or changed.

On the CPL side, the laboratory was rated as a “Level 2” laboratory within the LRN. As a Level 2 laboratory, the CPL would respond to events by performing analyses on patient specimens for a variety of chemical constituents; or packaging and shipping specimens to the CDC and their partners for chemical analysis that could not be performed on site. On the BPL side, the laboratory was designated as a “Level 3” laboratory. As a BSL Level 3 laboratory, the BPL handles agents that could be indigenous or exotic and could be passed from one source to another by inhalation. These agents are harmful to human health to the extent of causing serious or lethal disease.

The BPL actively participates in the Food Emergency Response Network which “integrates the nation’s food-testing laboratories at the local, state, and federal levels into a network that is able to respond to emergencies involving biological, chemical, or radiological contamination of food.”

Many projects and events have occurred over the last decade where one or both of these labs were “in the thick of it,” so to speak. The BPL has assisted law enforcement agencies from local and state police to the Federal Bureau of Investigation.

They have also provided their expertise in testing food items that were suspected of being contaminated and causing serious illness.

Similar to the BPL, the CPL, too, has assisted law enforcement agencies preparing for chemical events and exercises. The CPL has actively trained with the Delaware National Guard’s Civil Support Team and the Delaware Department of Natural Resources and Environmental Control’s Environmental Response Team.

One notable exercise occurred in 2011. This exercise went by the code name “Operation Loose Package.” This exercise took place on November 8-10, 2011. Its premise was that a letter potentially containing anthrax was processed at the Hares Corner Postal Distribution Center in New Castle, DE. This activated the Bio Detection System (BDS) contained within the mail sorting system. The BDS was extracted and sent to the lab for analysis. In another part of the exercise, a call came in to the DPH Office of Drinking Water about someone seen dumping a powder into water storage tanks near the Postal Distribution Center. Samples were collected from that tank and sent to the lab. In addition to the laboratory analyses, decontamination exercises and simulated treatment were performed on volunteers. Various agencies participated in this exercise to test and hone their processes and procedures. Overall, this exercise was a huge success and DPHL was successful in detecting Bacillus anthracis and chromium from the tainted water supply.

Throughout the last decade, both of these laboratories have seen many changes, from adding new equipment and tests, to funding cuts and turnover of personnel. The latest change took place on October 13, 2014. It was decided that, due to funding cuts, the CPL would downgrade to a Level 3 laboratory within the LRN. This change in level means that the CPL will no longer maintain the ability to test patient specimens for chemical agents. In the case of an actual event or exercise the CPL will ship all patient specimens directly to the CDC laboratory or other designated laboratory within the network.

www.dhss.delaware.gov/dhss/dph/lab/labs.html
The DPHL had several staff participate in this race.

Nick Rapp placed 2nd in age group of men 29 and under, and placed 9th overall. Emily Hanlin placed 2nd in age group of women 30-39, and placed 18th overall. Yan Choi placed 2nd in age group of women 29 and under, and 46th overall.

Pictured above are the top three runners in the Male age 29 and Under Category (in no special order): Brian Clarke, Nick Rapp, and Claudy Joinville.

Pictured above are the top three runners in the Female age 29 and Under Category (in no special order): Misty Seemans, Yan Choi, and Valorie Luke.

Pictured above are the top three runners in the Female age 30 to 39 Category (in no special order): Lauren Brueckner, Emily Hanlin and Eliza Hirst.
New Year, Many Changes

Debra Rutledge, MBA MT(ASCP) Infectious Disease Laboratory Manager II

It is with mixed emotions that we inform everyone that Tara Lydick has left employment with the Delaware Public Health Laboratory (DPHL) as of January 9, 2015. Tara has accepted employment with the National Security Agency (NSA) as a chemist in Washington, DC. The NSA core missions are to protect U.S. national security systems and to produce foreign signals intelligence information.

Tara began working for the State of Delaware in 2004. Tara was hired as a Chemist IV and started the Chemical Terrorism (CT) program for the laboratory. Biological and Chemical Preparedness were two new major programs funded by the Centers for Disease Control (CDC) as a result of the 9-11 terrorist attacks on the World Trade Centers and the anthrax mailing in 2001. Tara was very much involved in getting the CT laboratory and methods up and running and getting the laboratory approved by CDC’s Select Agent Program. Tara spent numerous hours writing, training, and implementing many laboratory processes and plans. Tara was involved in outreach with sentinel laboratories and first responders by providing training for sample collection, chain of custody, and infectious and hazardous materials packaging and shipping. Tara most recently expanded her role to involve the Environmental Protection Agency’s Drinking Water Program and overseeing the Environmental Chemistry section at DPHL.

We wish Tara and her family well in her new position. We also want to congratulate Tara and her husband on the birth of their daughter in July of 2014. Best of luck and we appreciate all the hard work and accomplishments that she has done over the past years for the laboratory.

DPHL recently had the opportunity to work with Mr. Malik White, who assisted with LIMS administration. Malik worked on troubleshooting, running queries, updating reports, data entry, and helping in the various sections where he was needed.

Malik grew up in North East Philadelphia and relocated to Bear, DE for the duration of his middle school and high school years. He attended Virginia State University in Petersburg, VA from 2010 to 2013, where he played football and graduated with a degree in Biology.

In his career, Malik has conducted research with the Food and Drug Administration as an intern. The research consisted of testing the effects silver and titanium nano-particles have on DNA repair systems using E. Coli bacterium. He has worked as a laboratory technician for Alere Toxicology in Richmond, VA, testing urine samples for various illegal drugs through chemical extraction. His career also included a year of professional level arena football as a quarterback, and he currently coaches the Delaware Gamecocks club football team, a team of college kids attending local community colleges.

He has recently been accepted into the United States Army Reserves. He will attend basic training in Fort Jackson, SC, and then move on to Officer Candidate School in Fort Benning, GA, after completion of which he will be commissioned as an officer. He hopes to eventually attend medical school.

We wish Malik the best of luck on his new life adventure.
The Newborn Screening section of the Delaware Public Health Laboratory is very happy to welcome Microbiologist II Veronica Aleman, a great addition to our team. Veronica previously worked in an accredited forensic laboratory processing improvised explosive devices (IED's) to discover latent prints in Washington, DC. She has a keen sense of detail in her work. Prior to that, she attended Notre Dame of Maryland University in Baltimore, MD, studying Biology and Philosophy. Veronica self-describes herself as an Air Force 'brat' who was born in Austin, TX but has lived in Dover, DE since she was three. Done with loud, city living, she is looking forward to quiet, country living. Her favorite sport to play is field hockey and favorite sport to watch is college football. Go Texas Longhorns!

She is a huge animal lover who will never be a vegetarian because she hates vegetables. She is a welcome addition to our team and we feel fortunate to have culled her out of a large group of applicants.

Shemeeakah Powell resigned her position as contract Molecular Biologist on January 30, 2015. Shemeeakah joined the lab in late 2012 and successfully took over the Laboratory Response Network (LRN) testing and active involvement in the Select Agent and Toxin Program. During her tenure, she also participated in numerous validation studies for newer methods and implemented an inventory system now used throughout the lab.

We wish Shemeeakah the best of luck in her new position.

Veronica Aleman

If you have questions regarding these articles or would like to receive a hard copy of this newsletter, contact the DPHL at 302-223-1520.

To receive this newsletter by email, contact Anita Kettlehake, Editor, at Anita.Kettlehake@state.de.us.