PULSENET
A CRITICAL FOOD SAFETY SURVEILLANCE SYSTEM

Public health laboratorians are critical to the detection and prevention of foodborne illnesses. Through a national laboratory-based foodborne disease surveillance network known as PulseNet, public health and agricultural laboratories have detected high-profile outbreaks such as those from imported produce, peanut butter and peanut butter-containing products and raw cookie dough.\textsuperscript{1-3}

In 2008 alone, PulseNet laboratorians detected more than 1,500 local clusters of foodborne illnesses and increased the number of foodborne bacterial isolates tested. Additional resources and support are needed for public health laboratories to test and investigate all cases of foodborne illness. It is critical for the nation to recognize the impact that public health and agricultural laboratories have on the overall foodborne disease surveillance system to ensure a safer food supply.

The PulseNet network links public health laboratories nationwide, monitoring pathogens such as Shigatoxin producing Escherichia coli or STEC (including E. coli O157:H7), Listeria monocytogenes, Salmonella species (sp.), Shigella species and Campylobacter species using a molecular subtyping method called pulsed-field gel electrophoresis (PFGE) or DNA fingerprinting. Established in 1996 by the Centers for Disease Control and Prevention (CDC), four public health laboratories, the US Department of Agriculture (USDA) and the Association of Public Health Laboratories (APHL), PulseNet has since grown to more than 70 laboratories nationwide, including state and local public health laboratories, state agricultural laboratories and regulatory laboratories within the US Food and Drug Administration (FDA) and USDA. Molecular subtyping and computer analysis are performed to generate and analyze the DNA fingerprint patterns. These patterns generated from isolates of ill persons and/or food and environmental samples are then compared to national databases at CDC, allowing for the early identification of foodborne disease clusters. Such information assists epidemiologists with their investigations and may potentially
lead to the identification of the source of an outbreak.

APHL, which represents state and local public health and agricultural laboratories, supports its members by providing various training opportunities, disseminating information relevant to the public health laboratory community, facilitating the transfer of new technology nationwide, and conducting assessments of laboratory capacity and capabilities. In 2009, APHL administered a survey to assess the capability and capacity of the PulseNet network and to determine the challenges that face public health laboratories in achieving the goals and objectives of the network. This issue brief presents the findings of that survey.

METHODS
In April 2009, APHL conducted a survey to assess participating laboratories’ capacities and capabilities for conducting molecular subtyping through the PulseNet network for the 2008 calendar year. The survey was sent to 64 public health laboratories and agricultural laboratories—which included 50 state and territorial public health laboratories, 10 local public health laboratories and 4 state agricultural laboratories. Of those surveyed, APHL received 57 responses—comprised of 46 state and territorial public health laboratories, 9 local public health laboratories and 2 state agricultural laboratories—for an overall response rate of 89%. A similar survey was conducted in 2006 to assess activities for the 2005 calendar year. Some comparisons are included in this report for questions that were largely unchanged between 2005 and 2008.

The survey was administered through MR Interview, a web-based repository and survey tool. Descriptive analyses were conducted, and responses were grouped into three main categories: surveillance, communication, and laboratory management.

SURVEILLANCE
The PulseNet network has proven to be an essential component to the national foodborne disease surveillance system. Through active participation by local and state laboratories, this network has ensured improvements to the nation’s food safety system by detecting numerous outbreaks and preventing many illnesses. In recent years, laboratories have continued to increase the number of isolates tested by this molecular subtyping method.

In 2008, the PulseNet network collectively subtyped a total of 48,194 isolates of STEC, Salmonella, Shigella, L. monocytogenes and Campylobacter. This is a 47% increase from 2005 in which 32,830 isolates of the same pathogens were subtyped. In 2008, PulseNet laboratories were able to subtype 70% of the aforementioned foodborne pathogens received from clinical laboratory partners, as compared to 61% in 2005.

Although PulseNet has increased the percentage of foodborne isolates subtyped, all pathogens under PulseNet surveillance that are submitted to public health laboratories should ideally be subtyped. A significant delay or lack
of outbreak recognition may occur when laboratories do not subtype all foodborne isolates. In 2008, only 61% of laboratories were able to subtype all STEC, Shigella, L. monocytogenes and Salmonella isolates received in their laboratories. Laboratories did not subtype all isolates due to inadequate funding for supplies, staff shortages and increased workload in all areas of public health laboratory testing.

Currently, the bulk of isolates received and subtyped in PulseNet laboratories are of clinical origin; however, environmental samples and food samples are also important in potentially identifying the source of foodborne outbreaks. Sixty-seven percent (67%) of PulseNet laboratories subtyped isolates from environmental and food origins. This is an increase from 2005, where 45% of PulseNet laboratories subtyped food and environmental isolates. The importance of subtyping isolates of food and environmental origins was demonstrated in the 2008-2009 Salmonella Typhimurium multi-state outbreak associated with peanut butter and peanut butter-containing products. A match in DNA fingerprint profiles from the food source to the human outbreak strain prompted public health officials to issue consumer warnings and product recalls.

PFGE has proven to be a powerful tool for the detection of foodborne disease clusters. Additionally, newer molecular subtyping methods, such as multiple-locus variable number tandem repeats analysis (MLVA), can greatly contribute to some foodborne outbreak investigations. In some cases, MLVA can further discriminate between bacterial strains that have identical or very similar PFGE patterns. Currently, E. coli O157:H7 isolates that are part of a multi-state outbreak are sent to the CDC for MLVA testing and analysis. In 2008, only 12% of PulseNet laboratories performed supplemental or additional subtyping methods (a 3% increase from 2005). With additional funding and personnel, PulseNet laboratories would have the capability to perform these new subtyping techniques in their own laboratories, thus eliminating the additional time needed for specimen transport to CDC and potentially reducing the amount of time it takes to identify an outbreak.

Among the more than 1,500 foodborne disease clusters that were identified through PulseNet in 2008, three of every four local clusters detected were followed-up by an epidemiologist. The detection of small clusters by local public health laboratories can aid in the identification of larger multi-state outbreaks. This demonstrates the important role that state and local agricultural and public health laboratories contribute to the overall foodborne investigation process.

Figure 1. Comparison of Isolates PFGE Subtyped by PulseNet Laboratories in 2005 and 2008

Note: Comparison of isolates PFGE subtyped as share of the total amount of isolates received in PulseNet laboratories in 2005 and 2008.
COMMUNICATION
Communication and data sharing among PulseNet laboratories is critical to detecting and identifying multi-state outbreaks. One important component of the PulseNet surveillance system is the secure online databases that allow for the comparison of DNA fingerprints from regulatory agencies, public health and agricultural laboratories across the nation. The rapid comparison of DNA fingerprints allows laboratories to determine whether a small, local cluster is part of a larger multi-state outbreak. These databases, along with a secure online web-board available to PulseNet participants, allow for timely notification of new clusters or “matches” to an existing national cluster. In 2008, available resources allowed for 72% of laboratories to respond to web-board postings within two working days. This is an increase from 2005, in which 49% of laboratories were able to respond within two working days.

Communication between laboratory and epidemiology partners is another key factor in promptly identifying foodborne outbreaks. Seventy-five percent (75%) of PulseNet laboratories in 2008 reported communicating with their state or local foodborne epidemiologists on a weekly basis or more. This is a significant improvement over 2005, in which 59% of laboratories reported communicating with their epidemiologists in the same timeframe.

Additional tools, such as an electronic interface linking laboratory and epidemiology data, could allow outbreak investigations to be conducted more efficiently. In 2008, only 11 of 57 laboratories (19%) reported having a mechanism capable of data exchange between the laboratory and the epidemiology programs. If more laboratories can electronically exchange information with epidemiologists, case interviews could readily be linked to isolate information allowing for rapid response to outbreak investigations.

Figure 2. Comparison of Isolates PFGE Subtyped by PulseNet Laboratories in 2008

Note: Comparison of isolates PFGE subtyped by pathogens as share of total isolates received in 2008.


In 2008, only 11 of 57 laboratories (19%) reported having a mechanism capable of data exchange between the laboratory and the epidemiology programs.
LABORATORY MANAGEMENT

Many laboratories are struggling to keep up with the demand of foodborne pathogen testing due to budget decreases at the state and local government levels. PulseNet laboratories need the capabilities, capacity and resources to promptly subtype all foodborne pathogens received in their laboratories. Findings from our survey suggest that lack of funding in 2008 has impacted laboratory capacity at the local and state levels.

- 46% encountered personnel shortages
- 41% reported inadequate funding for supplies
- 27% reported a lack of certified laboratory staff to perform DNA subtyping

TURNAROUND TIME

Since 2005, PulseNet laboratories have decreased the amount of time it takes to subtype an isolate. Turnaround time is defined as the time (in working days) from the receipt of an isolate in the PulseNet laboratory to the time the isolate’s image is uploaded to the national PulseNet database. The PulseNet surveillance system uses turnaround time as a measure to determine its efficiency. CDC guidelines suggest a turnaround time of four working days for all PulseNet organisms. In 2008, the median turnaround time for STEC and L. monocytogenes was four days, a decrease from 2005 by one day. The median turnaround time in 2008 for Salmonella and Shigella was five days, a significant decrease of three days from 2005.

Although turnaround time has decreased over the years, there is always room for improvement. As demonstrated by recent large, multi-state outbreaks—such as the Salmonella Saintpaul outbreak associated with fresh produce and the E. coli O157:H7 outbreak associated with raw cookie dough products—turnaround time affects the timeliness of identifying related ill cases and the source of the outbreak. Approximately 33% of laboratories subtyped Salmonella and Shigella isolates within CDC’s suggested turnaround time of four days, as compared to approximately 75% of laboratories that subtyped STEC and L. monocytogenes. The large amount of Salmonella and Shigella isolates received in laboratories may account for the five-day turnaround time. In 2008, PulseNet laboratories received 59,249 isolates of Salmonella and Shigella compared to 5,238 isolates of STEC and L. monocytogenes. Due to the sheer number of Salmonella and Shigella isolates received in PulseNet laboratories, some laboratories were unable to perform testing within the four-day period.

Another factor that affects turnaround time is the point at which molecular subtyping is performed. Some pathogens require an additional testing method known as serotyping.

### Figure 3. Turnaround Time (in Median Working Days) of Foodborne Pathogens under PulseNet Surveillance in 2005 and 2008

<table>
<thead>
<tr>
<th>Pathogen</th>
<th>2005</th>
<th>2008</th>
</tr>
</thead>
<tbody>
<tr>
<td>STEC</td>
<td>5</td>
<td>4</td>
</tr>
<tr>
<td>L. monocytogenes</td>
<td>5</td>
<td>4</td>
</tr>
<tr>
<td>Salmonella</td>
<td>8</td>
<td>5</td>
</tr>
<tr>
<td>Shigella</td>
<td>8</td>
<td>5</td>
</tr>
<tr>
<td>Campylobacter</td>
<td>13</td>
<td>5</td>
</tr>
</tbody>
</table>

which is a traditional method to characterize bacterial isolates. In general, once serotyping is completed, the laboratory then proceeds with molecular subtyping. Serotyping may take a few days to perform and, thus, increases the overall turnaround time. On average, 42% of PulseNet laboratories simultaneously performed serotyping and molecular subtyping of STEC, L. monocytogenes, Salmonella sp., Shigella sp. and Campylobacter sp. in 2008. This is an improvement from 2005, where 30% of laboratories reported simultaneous testing. Thus, as expected, laboratories that reported simultaneous serotyping and molecular subtyping had a lower turnaround time than those that completed serotyping first then continued with molecular subtyping.

While it appears that the best practice to decrease turnaround time in PulseNet laboratories is to simultaneously perform serotyping and molecular subtyping on all isolates, some laboratories do not find this application feasible. With workforce shortages, decreasing funds and lack of trained staff, some laboratories do not have the capability to simultaneously serotype and subtype PulseNet pathogens, nor do they have the resources to subtype all these pathogens once they have been serotyped.

**Staff Shortage**

The impact of inadequately staffed laboratories on the foodborne surveillance system is tremendous. In 2005, one in three PulseNet laboratories had a vacancy rate of 25% or greater. In 2008, one in two PulseNet laboratories had a vacancy rate of 25% or greater. As funds continue to decrease, and layoffs of public health personnel and work furloughs continue, many public health laboratory staff are faced with multiple responsibilities and priorities that negatively affect their ability to perform molecular subtyping on foodborne pathogens in a timely fashion. Additionally, with emerging diseases such as the novel 2009 strain of Influenza A/H1N1, it is very possible that laboratories will encounter staff shortages due to the need to redirect efforts to new public health threats.

**CONCLUSION**

PulseNet laboratories at the state and local levels have been and will continue to be an integral part of the nation’s foodborne disease surveillance system. The 2009 APHL survey demonstrated

---

**Figure 4. General PulseNet Algorithm for Laboratory Testing of Foodborne Pathogens: Serotyping and Subtyping**

- Confirm identification of foodborne pathogen isolated from ill patient, food or environmental sample
- Serotyping performed to characterize the pathogen
- Molecular subtyping and analysis performed
- Simultaneous or non-simultaneous testing
- Check for other isolates that match this DNA fingerprint (clusters) in the local databases
- Upload the DNA fingerprint to national PulseNet databases
- Check the national databases for multi-state clusters
- Notify epidemiologists of DNA fingerprint matches or clusters
In 2005, one in three PulseNet laboratories had a vacancy rate of 25% or greater. In 2008, one in two PulseNet laboratories had a vacancy rate of 25% or greater.

that public health and agricultural laboratories overall increased the percentage of isolates subtyped, identified an increasing amount of foodborne disease clusters and have decreased the turnaround time for PFGE testing. However, challenges to the network include the ability to timely subtype all foodborne pathogens, hire additional certified staff and provide tools that will enhance communication between laboratorians and epidemiologists.

The challenges faced by PulseNet laboratories can be addressed with additional resources and commitment from local, state and federal governments. In 2009, the Council to Improve Foodborne Outbreak Response (CIFOR) released guidelines aimed at standardizing practices among all food safety disciplines responsible for foodborne disease surveillance and outbreak response.7 Hopefully, with these guidelines, PulseNet laboratories will continue to improve surveillance and response to foodborne outbreaks at all levels of government.

REFERENCES


The Association of Public Health Laboratories is a national non-profit located in Silver Spring, MD, that is dedicated to working with members to strengthen governmental laboratories with a public health mandate. By promoting effective programs and public policy, APHL strives to provide public health laboratories with the resources and infrastructure needed to protect the health of US residents and to prevent and control disease globally.

Funders
This issue brief was supported in part by Cooperative Agreement Number U60/CCU33019 from CDC. Its contents are solely the responsibility of the authors and do not necessarily represent the official views of CDC.