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Laboratory-Based Surveillance for Influenza: Role of the Wisconsin State Laboratory of Hygiene

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ABSTRACT

Influenza poses a significant threat to public health worldwide. In the United States alone, mortality attributed to annual epidemics of influenza is estimated at 36,000 deaths per year. Influenza viruses also cause pandemics, during which the rates of illness and death can be expected to be much higher. In order to optimize prevention and control strategies for influenza, a variety of surveillance activities are carried out year-round internationally, nationally, and at the state level. This article summarizes relevant features of influenza and the surveillance activities carried out at each of these levels to monitor influenza activity. Particular emphasis is given to the state's laboratory-based surveillance network, developed and coordinated by the Wisconsin State Laboratory of Hygiene, that features strong and productive partnerships with private sector clinicians and laboratories and that provides a solid foundation for surveillance directed at other public health threats such as Severe Acute Respiratory Syndrome and bioterrorism.

INTRODUCTION

It is ironic that, in light of the emergence and media coverage of newly recognized disease threats such as West Nile virus, Severe Acute Respiratory Syndrome (SARS), and monkeypox, one of our oldest and most well-recognized diseases, influenza, remains among the greatest national and international threats to public health. More than 2400 years after influenza-like epidemics were first documented by Hippocrates and Thucydides,[1] influenza viruses continue to plague us with annual epidemics and sporadic pandemics. An estimated 36,000 deaths[2] are attributed to influenza epidemics annually in the United States despite the availability of an effective vaccine. An estimated 500,000 deaths in the United States and 20-50 million deaths worldwide were attributed to the "Spanish Flu" pandemic of 1918-1919. The impact of annual influenza epidemics and the threat of future pandemics provide the impetus for comprehensive influenza surveillance activities at international, national, and state levels. In Wisconsin, influenza surveillance is dependent on the contributions of many partners in both the public health and private health care sectors. A vital component of this surveillance system, the Wisconsin State Laboratory of Hygiene (WSLH) has played a central role in surveillance efforts since it first began influenza testing as part of a "respiratory virus watch program" instituted in 1957 in response to clinician concerns about a predicted pandemic.

WHAT IS INFLUENZA?

Influenza is a respiratory illness characterized by the abrupt onset of fever and chills, typically followed by severe myalgia, sore throat, and cough that ensue over the next several days and often leave the patient bed-ridden.[3,4] Short-term gastrointestinal illnesses, often referred to as "stomach flu," are not caused by the viral agents that produce the acute respiratory disease of true influenza. Although the acute symptoms of influenza usually last only a few days, recovery can take weeks. Uncomplicated influenza in otherwise healthy individuals is responsible for significant morbidity each year, often leading to high rates of school and work absenteeism.[2] Of even greater public health concern is the impact that influenza and its complications, most notably pneumonia, have on certain

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high-risk groups such as the elderly and individuals of any age with chronic underlying disease, often resulting in high rates of hospitalization and mortality.[2] More recently, influenza has been recognized as a significant pathogen among infants and young children, leading to increased rates of hospitalization within these age groups during influenza epidemics.[5,6]

Influenza Nomenclature

The influenza viruses are classified into three types: A, B, and C. Influenza types A and B pose the greatest threats to human health; influenza type C viruses appear to be responsible for only mild respiratory illness.[3,4] Influenza type A viruses are further classified into subtypes, based on the surface glycoproteins hemagglutinin (H) and neuraminidase (N). The H and N are the viral components to which the protective host immune response is directed and by which strains are selected for inclusion in the vaccine each year. Only two subtypes, H1N1, and H3N2, currently circulate in humans, with new strains of one or both of these subtypes emerging each year. Recently, several novel avian subtypes of influenza, H5N1, H9N2 and H7N7, have been isolated from humans in the Far East and Europe, causing illness of varying severity, including death (H5N1).[7] Thus far, none of these viruses has demonstrated the capability for widespread human-to-human transmission. Influenza type B viruses do not have subtypes; however, new strains of type B influenza emerge routinely to cause epidemics, usually of milder illness than that of influenza A. Influenza viruses are named by the geographic origin of the first isolate, followed by the specimen number of that isolate and the year it was first isolated, for example, influenza B/Hong Kong/330/01. The names of influenza A viruses also include the subtype of the virus, for example, influenza A/New Caledonia/20/99 (H1N1).

Key Characteristics of Influenza Viruses

Two key characteristics help define the epidemiology of influenza and need to be accounted for in the public health response to influenza. The first of these is the ability of these viruses to readily change antigenically and thus continually re-infect human populations.[8] This antigenic change is accomplished by two distinct mechanisms.

The most dramatic mechanism of change, antigenic shift, occurs following reassortment between an animal (usually avian) and a circulating human subtype of influenza, giving rise to a novel subtype (a new H and/or N) of influenza to which the human population will have no existing immunity.[9] This event is a necessary, but not sole, prerequisite for development of an influenza pandemic such as occurred in 1918-1919 and more recently in 1957-1958 (influenza A/Japan (H2N2), "Asian Flu") and 1968-1969 (influenza A/Hong Kong (H3N2), "Hong Kong Flu"). Preparation for the next influenza pandemic, which most experts consider an inevitability, currently is a public health priority.[10,11]

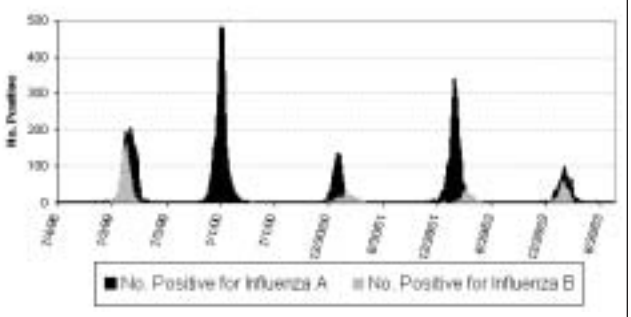

Figure 1. Seasonality of influenza detections by Wisconsin Vinology Laboratories, 1998-2003.

A less dramatic change, antigenic drift, occurs following frequent, but relatively minor point mutations that arise in the H or N year-round.[4] Antigenic drift gives rise to new strains of influenza A and B, resulting in the widespread winter influenza epidemics that we see nearly every year. Worldwide surveillance is carried out year-round to detect these antigenic changes and to assess the need for modification of the existing vaccine.

The second key characteristic of influenza is its striking seasonality in temperate climates. In Wisconsin, the first influenza viruses of the season are typically detected in November or December with peak epidemic activity occurring in January and February. Sporadic cases and minor outbreaks are occasionally detected during the early autumn, but human to human transmission of "out-of-season" cases is usually very limited. This pattern is seen every year, in virtually every community (Figure 1).

Both the annual winter epidemics and the sporadic pandemics pose significant threats to public health and require aggressive laboratory diagnostic and public health surveillance activities to detect and ultimately prevent or contain them.

LABORATORY DIAGNOSTIC METHODS

Clinical diagnosis of influenza is extremely difficult due to the overlapping seasonality and the symptomatology it shares with many other respiratory viruses. Laboratory testing is, therefore, essential to accurate diagnosis. A number of diagnostic methods are available, however, virus isolation in cell culture remains the

"gold standard."[1] Methodology improvements and the increased commercial availability of cell cultures and reagents have moved virus isolation beyond the traditional purview of the public health laboratory to the clinical laboratory. Other methods, such as direct specimen immunofluorescence have also found relatively widespread use, but with varying degrees of success. Although molecular methods are available, they are not widely used in the clinical setting.

Laboratory diagnosis of influenza has been further revolutionized with the advent of handheld direct antigen detection kits that employ enzyme immunoassay and similar technologies. Seven such test kits have been marketed since 1990. Instead of the days required for viral culture results, these rapid influenza tests provide results in minutes, allowing their use in patient management decisions. The relative simplicity of the tests allows testing to be performed outside traditional virology laboratories, e.g., in physician offices and long-term care facilities, further improving the availability of results for patient management. These tests are not without problems, however, including the lack of adequate validation studies and limitations in sensitivity, specificity, and result interpretation.[12,13] In addition to patient management, laboratory testing is also critical to public health surveillance for influenza.

INFLUENZA SURVEILLANCE

Comprehensive influenza surveillance is conducted at international, national, and state levels. The data obtained are used to provide guidance in the formulation of each season's influenza vaccine, in the use of antivirals for treatment and/or prophylaxis of influenza, and to provide early warning of the emergence of novel influenza subtypes or strains.

International influenza surveillance is coordinated by the World Health Organization (WHO) and serves as a global monitor for the emergence of influenza viruses with pandemic potential. One of the four WHO collaborating centers is the Centers for Disease Control and Prevention (CDC) in the United States, which also coordinates national surveillance in the United States.

National influenza surveillance in the United States utilizes a multi-faceted approach to track circulating strains of influenza and antigenic changes that occur, to track influenza-related illness, and to measure the impact influenza has on mortality rates. The first element of surveillance is provided by the vital statistics offices of selected US cities and monitors the total number of death certificates filed that list pneumonia or influenza as the underlying or contributing cause of death. The second element is carried out by state epidemiologists, who provide the CDC with weekly reports of the estimated level of influenza activity in their states based on analyses of laboratory and clinician data and reports of outbreaks in long-term care facilities, schools, and other settings.

The third element is provided by sentinel clinicians in each state, who provide weekly reports to the CDC on the proportion of patients they see with influenza-like illness (ILI), defined as fever of >100°F plus either a cough or a sore throat. In Wisconsin, these clinicians are strategically located around the state to provide representative population data.

The final element in virologic surveillance is provided by clinical and public health laboratories in the United States. Nationally, laboratories report the total number of respiratory specimens tested and the number positive for influenza types A and B each week. Moreover, certain laboratories, including the WSLH and the Milwaukee Health Department Laboratory (MHDL), provide data on the antigenic characteristics of circulating influenza strains and submit representative influenza isolates to the CDC. These efforts are vital for detection of novel strains or subtypes of influenza and for monitoring the relatedness of circulating virus strains to those strains present in the vaccine. In Wisconsin, the WSLH coordinates all laboratory-based surveillance.

THE WSLH'S LABORATORY-BASED INFLUENZA SURVEILLANCE NETWORK

Laboratory-based surveillance for influenza in Wisconsin is a collaborative effort between the Wisconsin Division of Public Health (WDPH) and the WSLH. Surveillance data, including the number of specimens tested and the number and type of viruses isolated, was once only available from the WSLH and the MHDL. These data are now provided by networks of clinician specimen submitters, virology laboratories, and rapid influenza test sites, and compiled by the WSLH for analysis by WDPH (Figure 2). In addition, patient specimens for virus isolation and viral isolates are submitted to the WSLH for further characterization. Each of these networks is distinct and has developed independently.

The network of clinician specimen submitters evolved from a collaboration with the University of Wisconsin-Madison Department of Family Medicine, and currently includes approximately 30 sites, including 2 University of Wisconsin student health centers, 1 rural hospital-clinic combination, and a variety of clinic sites strategically located around the state.

The virology laboratory network consists of Wisconsin's 10 clinical and public health virology laboratories and has been operational since 1995. Testing data from these laboratories are submitted to the WSLH and compiled on a weekly basis, with summary comments and graphics reported back to the participants and to the WDPH. Data are also made available on the WSLH website and the Health Alert Network for interested members of the clinical community.

In response to the growing number and increasing use of rapid influenza tests, the WSLH instituted seasonal reporting by rapid test sites during the 2001-2002 season as a potential enhancement to influenza surveillance. This system was initiated after an intensive campaign to educate users on methods to optimize use and interpretation of these tests. Testing sites provide the WSLH with weekly reports of the total number of specimens tested and the number of specimens positive for influenza. They also provide samples of their initial influenza-positive specimens for virus culture confirmation.

The WSLH has taken the lead role in the development of the state's laboratory network for influenza surveillance. This role represents a significant evolution from that of one of the state's only primary diagnostic virology laboratories to that of a reference and research laboratory, consultant, trainer, data manager, and information resource. These diverse activities are consistent with the core functions of a state public health laboratory, as prescribed by the Association of Public Health Laboratories (APHL), to more effectively support public health programs (Table 1).[14]

Specifically, the WSLH currently provides only limited primary diagnostic testing in support of patient management and influenza surveillance. However, as the state's public health laboratory, the WSLH provides reference testing, including subtyping and strain characterization of influenza isolates submitted from other laboratories and serves as a link to the CDC for further characterization. The WSLH also provides diagnostic testing in support of research into the epidemiology of influenza and strategies for prevention and control in select populations (i.e., long-term care facilities).[15-17] Beyond testing, the WSLH collates and analyzes information provided by Wisconsin's virology laboratories, rapid testing sites, and clinician submitters and disseminates these data to clinicians, state, and local health departments and others. The WSLH also provides guidance and consultation in the use and interpretation of antigen detection tests to clinicians, laboratorians, and public health personnel through audioconferences, workshops, on-site presentations, and mailings, and provides bench-training to Wisconsin virology laboratories on request.

Figure 2. Geographic distribution of laboratory-based influenza surveillance sites in Wisconsin, 2002-2003.

CONCLUSION

According to a 1998 prediction, "Submissions of specimens for virus isolation are expected to decrease as rapid antigen test kits are improved and become more widely available. Having fewer isolates for characterization is a potential public health problem."[18] One need not accept this outcome, however. The challenge to the public health laboratories is to adapt to the realities of today's health care and laboratory environments and develop plans to access and incorporate data available from other laboratory sources into their laboratory-based influenza surveillance systems.

This inclusionary approach in fact can provide many benefits. For example, the sensitivity of the surveillance network was enhanced by inclusion of the rapid test sites since the first Wisconsin influenza isolates in each of the last 2 seasons have been recovered from samples submitted to virology laboratories for culture confirmation of rapid test positives. Another benefit is the enhanced regional surveillance within the state demonstrated during the 2001-2002 season. In response to the detection of a potentially significant influenza A (H1N2) reassortant virus in Wisconsin, the WSLH requested additional samples from virology laboratories, a rapid test

Table 1. Core Functions of State Public Health Laboratories*

Disease prevention, control, and surveillance
Integrated data management
Reference and specialized testing
Environmental health and protection
Food safety
Laboratory improvement and regulation
Policy development
Emergency response
Public health-related research
Training and education
Partnerships and communication

* *Source*: Adapted from reference 14.

site, and clinician specimen submitters in the region of the initial virus detection to monitor for additional reassortants, which, indeed, led to the detection of 8 additional such viruses.

Finally, this network serves as a model and provides the foundation for laboratory-based surveillance and emergency response to public health threats other than influenza (e.g., SARS, monkeypox, and agents of bioterrorism).

The WSLH recognizes that it cannot accomplish its public health mission alone. More importantly, just as the WSLH recognizes the need for partners to achieve its public health mission, so the individual members of the surveillance network will recognize the key role they play in public health, especially in surveillance.

Developing and sustaining partnerships requires commitment and recognition of the quality, capabilities, and roles of private and public sector laboratories. In a recent statement, the president of the APHL stated "From local to national, the laboratories of our nation are becoming less independent and more interdependent…"[19] In Wisconsin, we believe that the development of interdependence and relationships between laboratories is not only beneficial, but is necessary to provide the best possible services to the citizens of the state. The institution and the staff of the WSLH remain as committed to serving the citizens and caregivers of Wisconsin as they were in 1957 when the WSLH responded to clinician needs and concerns by expanding its testing capability for influenza. We look forward to the challenges and opportunities that the next 100 years of the WSLH are certain to provide.

REFERENCES

1. Lennette EH, Lennette DA, Lennette ET, eds. Diagnostic Procedures for Viral, Rickettsial, and Chlamydial Infections. 7th edition. Washington, D.C.: American Public Health Association; 1995.
2. Centers for Disease Control and Prevention. Prevention and Control of Influenza: Recommendations of the Advisory Committee on Immunization Practices (ACIP). *MMWR-Morbidity and Mortality Weekly Report.* April 25, 2003; 52(RR08):1-36.
3. Cox N, Subbarao K. Influenza. *Lancet.* 1999;354:1277-1282.
4. Treanor JJ. Influenza Virus. In: Mandell GL, Bennett JE, Dolin R, eds. Mandell, Douglas and Bennett's Principles and Practice of Infectious Diseases. Philadelphia, PA: Churchill Livingstone; 2000:1823-1849.
5. Neuzil KM, Mellen BG, Wright PF, Mitchel EF, Griffin MR. The Effect of Influenza on Hospitalizations, Outpatient Visits, and Courses of Antibiotics in Children. *N Engl J Med.* 2000;342:225-231.
6. Neuzil K, Zhu Y, Griffin M, et al. Burden of Interpandemic Influenza in Children Younger than 5 Years: A 25-Year Prospective Study. *J Infect Dis.* 2002:185:147-152.
7. Centers for Disease Control and Prevention. The Influenza (Flu) Viruses page. Available at: http://www.cdc.gov/ncidod/diseases/flu/viruses.htm. Accessed August 14, 2003.
8. Webster R. Influenza: An Emerging Disease. *Emerging Infect Dis.* July-September 1998;4(3). Available at: http://www.cdc.gov/ncidod/EID/vol8no7/02-0157.htm. Accessed August 14, 2003.
9. Horimoto T, Kawaoka Y. Pandemic Threat Posed by Avian Influenza A Viruses. *Clin Microbiol Rev.* January 2001;129-149.
10. Patriarca P, Cox N. Influenza Pandemic Preparedness Plan for the United States. *J Infect Dis.* 1997;176(Suppl I):S4-7.
11. Snacken R, Kendal A, Haaheim L, Wood J. The Next Influenza Pandemic: Lessons from Hong Kong, 1997. *Emerging Inf Dis.* March-April 1999;5(2). Available at: http://www.cdc.gov/ncidod/eid/vol5no2/snacken.htm. Accessed August 14, 2003.
12. Kirk C. Recommendations for the use of rapid influenza tests. Results - The WSLH Newsletter. Fall 2000;26.
13. Kirk C. Influenza and so much more - it's winter in Wisconsin. Results - The WSLH Newsletter. Winter 2002;30.
14. Centers for Disease Control and Prevention. Core Functions and Capabilities of State Public Health Laboratories. *MMWR-Morbidity and Mortality Weekly Report.* [serial online]. September 20, 2002; 51(RR14); 1-8. Available at: http://www.cdc.gov/mmwr/PDF/RR/RR5114.pdf. Accessed August 14, 2003.
15. Drinka P, Gravenstein S, Krause P, Nest L, Dissing M and Shult P. Management of influenza in a nursing home. *Ann LTC: Clin Care Aging.* 2000;8:23-30.
16. Drinka P, Gravenstein S, Krause P, Nest L, Dissing M, and Shult P. Re-introduction of influenza A to a nursing building. *Infect Control & Hosp Epidemiol.* 2000;21:732-735.
17. Vij S, Gravenstein S, Schilling M, Drinka P, Shult P, Miller B. Influenza B presentation differs in elderly patients. *J Amer Geriat Soc.* 2001;49(118):339.
18. Gensheimer K, Fukuda K, Brammer L, Cox N, Patriarca P, Strikas R. Preparing for Pandemic Influenza: The Need for Enhanced Surveillance. Emerging Infect Dis. March-April 1999;5(2). Available at: http://www.cdc.gov/ncidod/eid/vol5no2/gensheimer.htm. Accessed August 14, 2003.
19. Crouch N. President's Thoughts. The APHL Minute. July-August 2003:4:2.

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