1996-1997 INFLUENZA SEASON: CANADIAN LABORATORY DIAGNOSES AND STRAIN CHARACTERIZATION

Introduction

In collaboration with the World Health Organization (WHO) international collaborating laboratories, provincial laboratories, and other Canadian hospital and university-based virus laboratories, the Laboratory Centre for Disease Control (LCDC) conducts national surveillance on human influenza viruses. This surveillance monitors influenza activity, detects and describes antigenic changes in the circulating strains of influenza virus in Canada, and estimates, through periodic serosurveys, susceptibility to currently circulating and emerging strains. Canadian influenza surveillance information and actual representative strains are then shared with the WHO’s collaborating centres for influenza to contribute to global influenza monitoring.

Influenza Activity

Figure 1 shows the number and month of laboratory-confirmed influenza virus isolations, detections, and serodiagnoses reported from laboratories that contribute to the Canadian Virus Reporting (CVR) program, a surveillance program covering all laboratory-diagnosed viral infections. Since 1989, the number of laboratories participating in CVR has increased gradually (30, 30, 32, 33, 37, 39, 42, respectively), which should be taken into consideration when comparisons are made over different years. The 1996-1997 influenza season in Canada began in late November 1996 and continued to May 1997. During this period, there were 1,173 reports of influenza B viruses, with the largest number (492) occurring in March. There were 1,953 laboratory reports of influenza A and influenza A subtypes in the same period; the peak was reached in January.

During influenza seasons, influenza A and influenza B viruses have usually predominated alternatively during a particular season. However, in the 1996-1997 season, both A and B types prevailed in Canada, although the type A viruses peaked earlier whereas the type B viruses occurred late during the season. Furthermore, the activity level of both type A and type B viruses in the 1996-1997 season was relatively higher than those of the previous 4 years, which could not be explained solely by the increasing number of reporting laboratories.

Strain Characterization

During the 1996-1997 influenza season, isolates submitted from provinces to LCDC for strain typing turned out to be highly homogenous. All type A isolates except one were A/Wuhan/359/95-like(H3N2) (223) and all of the 62 type B isolates were B/Beijing/184/93-like (Figure 2 and Table 1). No subtype H1N1 isolates were submitted to LCDC. A large number of influenza A isolates were still arriving at LCDC in March and most of the type B isolates were received in February and March. Table 1 indicates the provincial source and identity of submitted isolates.

A quick genetic approach has been developed at LCDC to facilitate the screening for influenza virus variants. Preliminary testing of the approach on influenza isolates received during the 1996-1997 season indicated that some strains showed variation from the vaccine strains, although the standard hemagglutination inhibition assay did not identify significant antigenic changes. Work is under way at LCDC to correlate identified genetic variations with antigenic changes and, consequently, to develop strategies for integrated antigenic and genetic characterization of influenza virus variants.

Discussion

The past 1996-1997 influenza season in Canada was more severe than the previous ones; both type A and type B infections reached high levels in the season according to laboratory reports from provinces. However, all of the characterized isolates were homogenous and antigenically similar to the vaccine component strains.
Figure 1
Laboratory evidence of human influenza virus infections in Canada
Infections par le virus grippal humain d’après les données de laboratoire, Canada

![Graph showing influenza virus infection reports by month and season](image)

- Influenza A (total)*
- Grippe A (total)*
- Influenza A(H1N1)
- Grippe A(H1N1)
- Influenza A(H3N2)
- Grippe A(H3N2)
- Influenza B
- Grippe B
* Includes A(H1N1), A(H3N2), and non-subtyped.
* Inclut A(H1N1), A(H3N2), et les souches non-typées.

Figure 2
LCDC antigenic characterization completed on influenza virus isolates in the 1996-1997 season, by month of submission
Caractérisation antigénique par le LLCM des isolats de virus grippal de la saison 1996-1997, selon le mois de leur soumission

![Bar chart showing influenza virus isolates by month](image)

- A/Johannesburg/33/94(H3N2)
- A/Wuhan/359/65(H3N2)
- B/Beijing/184/93
Globally, influenza A(H3N2), A(H1N1), and B viruses continued to circulate worldwide (1). Most antigenically characterized influenza A(H3N2) viruses were similar to the reference strain A/Wuhan/359/95 and the antigenically equivalent vaccine strain A/Nanchang/933/95. Although influenza A(H1N1) viruses were isolated only sporadically during the 1996-1997 influenza season, an increasing number of antigenically characterized isolates showed variation from the vaccine strain A/Texas/36/91 but were close to A/Bayern/07/95. Most influenza B viruses were similar to the reference strains B/Beijing/184/93 and B/Harbin/07/94. A small number of type B viruses that were related to the antigenically distinct B/Victoria/02/87 were isolated in Asia (1).

Vaccines containing A/Nanchang/933/95 and B/Harbin/07/94 induced antibodies with similar frequency and titre to the vaccine viruses and to recently isolated H3N2 and B strains. However, the H1N1 component A/Texas/36/91 induced a good antibody response to the vaccine strain but less frequent and reduced antibody responses to recent H1N1 isolates such as A/Bayern/07/95(1). Therefore, WHO recommended the following strains as vaccine components for the 1997-1998 season (1):

- A/Wuhan/359/95-like(H3N2),
- A/Bayern/07/95-like(H3N2), and
- A/Bayern/184/93-like.

Basically, the H1N1 component is changed and the antigenically equivalent strain that will be used by American vaccine manufacturers is A/Johannesburg/82/96(1).

**Acknowledgements**

The collaboration of laboratories in the CVR program and of provincial and hospital laboratories who forwarded early and representative isolates of influenza virus is a vital part of influenza surveillance in Canada.

Influenza virus isolates were submitted from the following centres:

- British Columbia Centre for Disease Control, Virology Services, Vancouver, BC;
- Virology and Reference Laboratory, U.B.C., Vancouver, BC;
- Provincial Laboratory of Public Health for Southern Alberta, Calgary, AB;
- Provincial Laboratory of Public Health for Northern Alberta, Edmonton, AB;
- Saskatchewan Public Health Laboratory, Laboratory and Disease Control Services Branch, Regina, SK;
- Cadham Provincial Laboratory, Winnipeg, MB;
- Regional Public Health Laboratory, Laboratory Services Branch, Virus Laboratory, Toronto, ON;
- Regional Public Health Laboratory, Peterborough, ON;
- Regional Public Health Laboratory, Kingston, ON;
- Regional Public Health Laboratory, Ottawa, ON;
- Children’s Hospital of Eastern Ontario, Ottawa, ON;
- Regional Public Health Laboratory, Timmins, ON;
- Regional Public Health Laboratory, Thunder Bay, ON;

### Table 1
LCDC strain characterization completed on influenza isolates, Canada*

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* Submitted from 20 November 1996 to 15 May 1997, by province.
**Case Report**

**NEISSERIA MENINGITIDIS SEROGROUP A IN ONTARIO**

Epidemics of meningococcal meningitis caused by *Neisseria meningitidis* serogroup A virtually disappeared from industrialized nations after World War II\(^1\). Since 1990, the Laboratory Centre for Disease Control (LCDC) in Ottawa has confirmed five cases of *N. meningitidis* serogroup A in Canada, with no laboratory-confirmed cases in the past 3 years. In Ontario, *N. meningitidis* serogroup A has not been confirmed since 1990. In early 1997, we identified *N. meningitidis* serogroup A isolated from a case of meningitis in the greater Toronto area.

A 30-year-old male returning to Canada in January 1997 from the northern state of Punjab, India, became ill with symptoms of a high fever and headache during the flight back to Toronto. The patient was admitted to hospital where cultures of both blood and cerebrospinal fluid were positive for *N. meningitidis*. The patient was treated with a course of ampicillin and ceftriaxone, and recovered uneventfully. The isolates were referred to the Central Public Health Laboratory, Toronto, for confirmation and typing.

The isolates were grown overnight at 35°C in 5% CO\(_2\) on sheep blood agar for biochemical confirmation and on Mueller-Hinton agar with 5% lysed horse blood for typing and were serogrouped by slide agglutination\(^2\). Serology revealed strong agglutination in serogroup A antiserum with no visible reaction in antiserum for groups B, C, X, Y, Z, W135, 29E and wheat germ agglutinin. The isolates were susceptible to penicillin (minimum inhibitory concentration = 0.06 mg/L), ceftriaxone, chloramphenicol, rifampin, and resistant to sulfadiazine by agar dilution method\(^3\). The cultures were forwarded to the Neisseria Reference Centre, LCDC, Ottawa, where they were characterized as NM A:4:P1.9/clone III-1.

**Discussion**

Meningococcal disease is a serious public-health concern worldwide. In Europe and North America, the annual incidence of meningococcal disease is one to three per 100,000 population and epidemics have virtually disappeared, except for limited outbreaks and clusters of cases. In developing countries and specific areas such as the "meningitis belt" of sub-Saharan African, epidemics of meningococcal disease occur approximately every 8 to 10 years and can affect 1% of the population. The annual incidence of disease is about 10 to 50 cases per 100,000 in these hyper-epidemic areas, and can rise to more than 500 cases per 100,000 during epidemics\(^4-7\). In the early 1980s, serogroup A:4:P1.9/clone III-1 caused epidemics in China and Nepal before spreading to the north of India in 1985. In the summer of 1987, the same strain of *N. meningitidis* was carried by Muslim pilgrims from southern Asia to Mecca and Medina, Saudi Arabia, resulting in 7,000 cases\(^8-10\). The relatively small number of cases associated with this epidemic that were detected in Europe and North America were linked to pilgrims returning home. Continued spread of this strain to Africa following the return of Muslim pilgrims to "meningitis-belt" countries has continued into 1996\(^11,12\).

In Ontario, the majority of cases of meningococcal disease are due to *N. meningitidis* serogroups B, C, Y, and occasionally W135. We have described an isolated case in Ontario of meningococcal disease associated with the same strain of *N. meningitidis* A:4P1.9/clone III-1 that has been linked to the major outbreaks in Africa, China, Nepal, the Middle East, and North India. Although only sporadic cases of meningococcal serogroup A may occur in Canada, laboratory staff dealing with such specimens should be aware of the possible occurrence of *N. meningitidis* serogroup A, particularly in patients with a recent travel history to pandemic or epidemic regions.

**References**


Since April 1997, the Centers for Disease Control and Prevention (CDC) has received reports of outbreaks of cyclosporiasis in the United States and Canada. As of 11 June, there have been 21 clusters of cases of cyclosporiasis reported from eight states (California, Florida, Maryland, Nebraska, Nevada, New York, Rhode Island, and Texas) and one province in Canada (Ontario). These clusters were associated with events (e.g. receptions, banquets, or time-place related exposures [meals in the same restaurant on the same day]) that occurred during 19 March to 25 May, and comprise approximately 140 laboratory-confirmed and 370 clinically defined cases of cyclosporiasis. In addition, four laboratory-confirmed and approximately 220 clinically defined cases have been reported among persons who, during 29 March to 5 April, were on a cruise ship that departed from Florida. Approximately 70 laboratory-confirmed sporadic cases (i.e. cases not associated with events, the cruise, or recent overseas travel) have been reported in the United States and Canada. The most recent laboratory-confirmed sporadic case occurred in a person who had onset of symptoms on 3 June.

Fresh raspberries were served at 19 of the 21 events and were the only food in common to all 19 events, which occurred in April and May. At six of the 19 events, raspberries were the only type of berry served or were served separately from other berries; at 13 events, raspberries were included in mixtures of various types of berries. Eating the food item that included raspberries was associated with illness in seven of the 15 events for which epidemiologic data are currently available (including for three of the events at which raspberries were not served with other types of berries), and was associated with illness but not significantly for six events (i.e. all or nearly all ill persons ate the berry item that was served). The raspberries reportedly had been rinsed in water at 10 (71%) of the 14 events for which such information is available. Guatemala has been identified as one of the possible sources of raspberries for all eight events for which traceback data are currently available (i.e. Guatemala was the source of at least one of the shipments of raspberries that could have been used) and as the only possible source for at least one of these events and perhaps for two others for which the traceback investigations are ongoing.

Fresh raspberries were not served at two events in restaurants in Florida that have been associated with clusters of cases of cyclosporiasis (persons were exposed on 19 March and 10 April, respectively, in two different cities). The first cluster was associated with eating mesclun (also known as spring mix, field greens, or baby greens, a mixture of various types of baby leaves of lettuce); the specific source of the implicated mesclun has not been determined. Mesclun also is suspected as the vehicle for the second cluster.

**MMWR Editorial Note**

The investigations described in this report indicate that fresh raspberries imported from Guatemala are the probable vehicle of infection for most of the outbreaks of cyclosporiasis identified in 1997. There is no evidence of ongoing transmission of Cyclospora in association with mesclun, which was the vehicle for one, and possibly two, early outbreaks in March and April. In the spring and summer of 1996, an outbreak of cyclosporiasis in the United States and Canada was linked to eating raspberries imported from Guatemala. However, the mode of contamination of the raspberries implicated in that outbreak was not determined in part because the methods for testing produce and other environmental samples for this emerging pathogen are insensitive and nonstandardized. No outbreaks of cyclosporiasis were reported in the United States in association with importation of raspberries from Guatemala during the fall and winter of 1996; however, cyclosporiasis is highly seasonal in some countries.

After the outbreak in 1996, the berry industry in Guatemala, in consultation with the Food and Drug Administration (FDA) and CDC, voluntarily implemented a Hazard Analysis and Critical Control Point system and improved water quality and sanitary conditions on individual farms. The occurrence of outbreaks in 1997 suggests either that some farms did not fully implement the control measures or that the contamination is associated with a source against which these measures were not directed.

At FDA’s request, on 30 May, 1997, the government of Guatemala and the Guatemalan Berries Commission announced their decision to voluntarily suspend exports of fresh raspberries to the United States (the last shipment was 28 May). FDA is working with CDC, the government of Guatemala, and the Guatemalan Berries Commission to determine when exports can resume.