Laboratory-supported influenza surveillance in Victorian sentinel general practices

Heath Kelly,[1] Anne Murphy,[2] Wendy Leong,[1] Jennie Leydon,[1] Penny Tresise,[1] Marie Gerrard,[1] Doris Chibo,[1] Chris Birch,[1] Ross Andrews,[2] Mike Catton[1]

Abstract

Laboratory-supported influenza surveillance is important as part of pandemic preparedness, for identifying and isolating candidate vaccine strains, for supporting trials of anti-influenza drugs and for refining the influenza surveillance case definition in practice. This study describes the implementation of laboratory-supported influenza surveillance in Victorian sentinel general practices and provides an estimate of the proportion of patients with an influenza-like illness proven to have influenza. During 1998 and 1999, 25 sentinel general practices contributed clinical surveillance data and 16 metropolitan practices participated in laboratory surveillance. Serological, virus-antigen detection, virus culture and multiplex polymerase chain reaction procedures were used to establish the diagnosis of influenza. Two laboratories at major teaching hospitals in Melbourne provided additional data on influenza virus identification. General practice sentinel surveillance and laboratory identification of influenza provided similar data on the pattern of influenza in the community between May and September. The clinical suspicion of influenza was confirmed in 49 to 54 per cent of cases seen in general practice. *Commun Dis Intell* 2000:24;379-383.

Keywords: influenza, diagnosis, surveillance, community medicine

Introduction

Laboratory confirmation of the diagnosis of influenza in sentinel general practices was introduced into Victoria in 1998 as a joint initiative of the Victorian Infectious Diseases Reference Laboratory (VIDRL)* and the Department of Human Services (DHS). This complemented the clinical influenza surveillance that had been coordinated by DHS through a sentinel general practitioner (GP) network in previous years.

Surveillance with laboratory support has been recognised as an important part of the pandemic response in Australia's recently published pandemic plan.[1] In late 1997, during the planning of this initiative, the significance of a small number of human cases of influenza virus type A (H5N1) in Hong Kong was unclear and the possibility of the emergence of a new pandemic strain remained very real.[2]

Moreover, in recent years, laboratory surveillance has also been critical in supporting general practice trials of anti-influenza drugs, specifically neuraminidase inhibitors, used in the treatment and prevention of influenza.[3] However, not all influenza-like illness (ILI) seen in general practice will be confirmed as influenza. Published estimates of the proportion of patients with ILI confirmed as having influenza by laboratory testing vary from as low as 6 to14 per cent at the introduction of surveillance networks[4] up to 70 per cent as part of a trial of influenza antiviral medication.[3] The present study describes the implementation of laboratory supported influenza surveillance in Victorian sentinel practice sites in 1998-9 and provides an estimate of the proportion of patients with an ILI proven to have influenza.

Methods

Recruitment of GPs to the sentinel surveillance network was restricted to those who were anticipated to have an interest in surveillance. Random selection of general practice sites was not attempted. Medical officers of health in local government authorities and general practitioners who had participated in influenza surveillance in previous years, or those known to have an interest in immunisation, were invited to become part of the sentinel surveillance network. At the end of the recruitment process, in an attempt to include sentinel practices throughout Victoria, practices in regions that were not represented in the network were contacted. In all 17 practices participated in 1998 and 26 in 1999. GPs in 14 practices participated in both years.

General practice surveillance and specimen collection

Two types of surveillance were undertaken in sentinel general practices. The first required practices only to notify the proportion of patients with an ILI as a proportion of total patients seen each week. As in previous years, ILI was defined using the Australian Sentinel Practice Research Network (ASPREN) criteria for the diagnosis of influenza, criteria also used for the International Classification of Health Care Problems in Primary Care (ICHPP-2).[5] These are:

an influenza epidemic and 4 of the criteria below, or 6 of the criteria below in the absence of an epidemic;

1. Victorian Infectious Diseases Reference Laboratory, North Melbourne, Victoria
2. Department of Human Services Victoria, Melbourne, Victoria

Corresponding author: Dr H Kelly, Victorian Infectious Diseases Reference Laboratory, Locked bag 815, Carlton South, Vic, Australia 3053. Telephone: +61 3 9342 2608. Fax: +61 3 9342 2665. E-mail: heath.kelly@mh.org.au

1. onset within 12 hours
2. cough
3. rigours or chills
4. fever
5. prostration or weakness
6. myalgia, widespread aches and pains
7. no significant respiratory signs other than redness of the throat and nasal mucous membranes
8. influenza in close contacts (modified for this study to 'history of influenza-like illness' to allow inclusion of illness in non laboratory-confirmed close contacts).

The second type of surveillance used laboratory support for the confirmation of the diagnosis of influenza; this was restricted to 16 metropolitan practices for logistical reasons. Eight practices participated in both years; 2 practices participated only in 1998 and 6 new practices were recruited in 1999. At each site the GP determined which patients were to be asked to provide a clinical specimen, gave these patients an information sheet and discussed the project with them. Following consultation with the Victorian DHS, since this study was considered an extension of normal patient care, formal ethics approval was not sought. Patients were, however, asked to sign a consent form indicating they understood this process and consenting to the collection of specimens, including a convalescent serum sample.

Participating patients provided details of which ASPREN criteria they met, their influenza vaccination history, and any history of recent travel or contact with travellers. From those fulfilling the ASPREN criteria, a throat swab and nose swabs from each nostril were pooled in a single vial of viral transport medium. Acute phase blood was collected. In 1998 attempts to collect a convalescent blood sample were made only for patients for whom a laboratory diagnosis could not be made by detection of viral antigen using an immunofluorescence assay, viral isolation or serology (detection of influenza virus specific IgM, or IgG titre³ 160, see below). In 1999 all patients were asked to provide a convalescent blood sample. Where possible, following collection, specimens were stored at 4°C before transport to VIDRL on the same day.

Laboratory testing

In both 1998 and 1999 epithelial cells drawn from nose and throat swabs were examined for the presence of influenza antigen using an indirect immunofluorescence method. Viral isolation was also attempted using standard methods.[6] Acute and convalescent sera, where available, were screened in parallel using a complement fixation test.[7] A 4-fold rise in titre between acute and convalescent samples or a titre 160 in the acute phase serum was taken to indicate recent infection with influenza virus. An immuno-fluorescence based assay to detect antibodies to influenza A and influenza B virus was used when the complement fixation test gave indeterminate results (2-4 fold rise in antibody titre or acute phase titre = 80). In 1999 a multiplex polymerase chain reaction (PCR) assay that detected and differentiated influenza virus type A (H1N1), influenza virus type A (H3N2) and influenza virus type B was used as an additional test of the nose and throat swabs.[8,9] All tests were performed on all available specimens in both years. A patient was considered to have influenza virus infection if one or more of the laboratory tests were positive.

Results and data analysis

Patient results were sent to the referring GPs in the usual way. A progress report of the influenza season was sent to participating general practices and laboratories each fortnight. Data were analysed in Epi Info (version 6).[10] Fisher's exact test and the Chi-squared distribution were used to test for association, assuming that patients who were part of surveillance were representative of all patients from the sentinel practices.

Surveillance at other laboratories

Virology laboratory staff at the Royal Children's Hospital and the Monash Medical Centre provided information on the number of identifications of influenza virus types A and B made each week. Data on age, sex and vaccination status were included when available. In addition VIDRL provided data on patients identified with influenza A or B who were not part of the surveillance project.

Results

In both 1998 and 1999, an influenza virus type A (H3N2) Sydney/5/97-like strain was the predominant circulating strain. This strain was a component of the influenza vaccine distributed in Australia in both years.

General practice clinical surveillance

Between April and September 1998, 463 patients (73 per cent from metropolitan practices) satisfying the ASPREN criteria for the clinical diagnosis of influenza were notified from 10 metropolitan and 7 regional practices. Over a similar period in 1999, 351 patients were notified, 30 per cent from 12 metropolitan practices and the remainder from 14 regional practices. There was no significant difference between the sex distribution of notified cases in 1998 (46% male) and 1999 (42% male). However, there was a significant difference in age (Figure 1).

Vaccination data were available for 807 of the 814 patients notified over both years. Only 8 per cent of all notified patients were vaccinated. Those aged 65 years and over were more likely to be vaccinated than younger people (46.5 per cent compared with 5.1 per cent respectively; $p<0.0001$).

General practice laboratory surveillance

A diagnosis of influenza was considered laboratory-confirmed if at least one of the laboratory tests for influenza was positive. The diagnosis was excluded if all tests, including convalescent serology, were negative. When a patient provided an incomplete set of specimen types and the results of all available tests were negative, the diagnosis was classified as not determined.

In the 2 years of this study, 152 patients provided clinical specimens for laboratory surveillance. The median age of patients was 40 years in 1998 (inter-quartile range 25 to 53 years) and 38 years (28 to 45 years) in 1999. There were fewer males (47%) than females. In 1998 the clinical diagnosis of influenza was confirmed by laboratory testing in 45/110 (41%) of all patients tested and in 45/92 (49%) of patients for whom a definite laboratory diagnosis or exclusion was made. In 1999 influenza was confirmed in 20/42 (48%) of all patients who provided clinical specimens and 20/37 (54%) for whom a definite diagnosis could be made (Table).

The proportion of patients with ILI confirmed by laboratory testing to have influenza virus infection was not significantly different in the 2 years during which the same type A (H3N2) Sydney/5/97-like influenza strain was circulating, so further analysis combined data for both years. Of all patients notified with an ILI, 46/85 (53%) were confirmed as having influenza during the peak weeks of the season, compared with 19/44 (43%) in the lower incidence weeks at the start and end of the seasons (p = 0.50). The proportion of patients for whom the clinical diagnosis of influenza was confirmed was not significantly different between the under 20 years, 20 to 59 years and 60 years-and-over age groups (71, 49 and 53 per cent respectively; p = 0.45). Nor was there any significant difference in the confirmation rate by practice, although this analysis is based on small numbers.

Vaccination and travel history

Of all patients indicating whether they had received vaccination against influenza prior to April in either year, influenza infection was either diagnosed or excluded in 107 of them. Of these, 23 (21%) were vaccinated. Influenza infection was confirmed in 9/23 (39%) vaccinated patients and 48/84 (57%) unvaccinated patients (p = 0.12).

Only 12 patients gave a history of recent travel in 1998 or 1999; there was no relationship between travel and a positive diagnosis for influenza (p = 0.27). Fifty patients gave a history of close contact with at least one person recently travelling outside Australia; there was no significant relationship between such contact and the confirmation of influenza (p = 0.16).

Combined surveillance data from all testing laboratories

Over the 2 years there were 730 notifications from 3 laboratories. Data contributed by VIDRL consisted of specimens referred from hospital in-patients who were not part of sentinel surveillance. Data from the Royal Children's Hospital and the Monash Medical Centre consisted of influenza virus identification in hospital in-patients. Except for 3 type B identifications in 1998 and 17 in 1999, all were influenza virus type A. Because 2 of the contributing laboratories service a predominantly paediatric population, 70 per cent of laboratory notifications were from patients aged less than 10 years. Despite the differences in the age of hospital in-patients and general practice sentinel surveillance patients, the seasonal pattern of influenza and ILI was similar in both years (Figure 2a and Figure 2b).

Discussion

Neither the circulating strains of seasonal influenza virus nor the severity of epidemics is readily predictable. Since 1981, influenza virus type A has been the predominant circulating strain in one season, with a mixture of influenza A and B virus types in alternate seasons.[11,12] Based on this pattern, an influenza season involving both influenza A and B had been expected for, but did not eventuate in, 1999 in Victoria or nationally.[13]

Influenza surveillance with laboratory confirmation of an influenza diagnosis allows the differentiation of ILI from influenza. In 2 years of surveillance in Victoria the proportion of patients confirmed to have influenza varied from 49 to 54 per cent, which is higher than the proportion reported from most other surveillance studies but lower than that reported from therapeutic trials[3,14] when a more specific clinical diagnosis may be required. A study from France during the 1995-6 influenza season examined the positive predictive

Figure 1. Patients with influenza-like illness notified from Victorian sentinel general practices, 1998 to 1999, by age-group

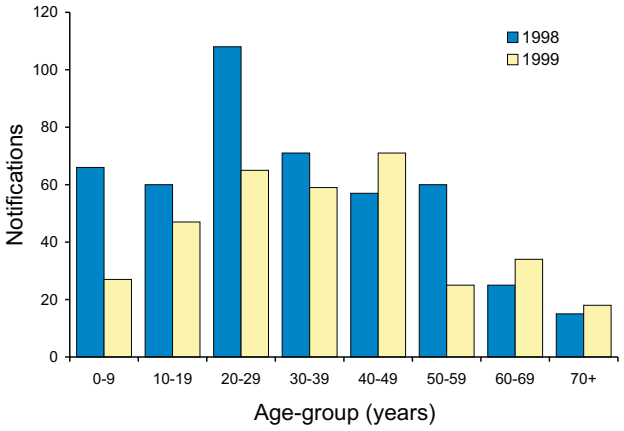


Table. Laboratory diagnosis of patients with influenza-like illness from sentinel general practice surveillance, Victoria, 1998 to 1999

	1998[1]		1999[2]	
Laboratory diagnosis	Patients	Per cent	Patients	Per cent
Influenza virus type A[3]	44	40	18	43
Influenza virus type B	1	1	2	5
Other viruses[4]	3	3	0	0
No virus detected[5]	44	40	17	40
Not determined[6]	18	16	5	12
Total	110	100	42	100

1 Tests used, 1998: serology, influenza virus antigen detection by immunofluorescence and virus isolation

2 Tests used, 1999: serology, influenza virus antigen detection by immunofluorescence, virus isolation and PCR

3 Coxsackie type B4 also cultured in one patient who seroconverted to influenza A

4 Rhinovirus, adenovirus, respiratory syncytial virus

5 Virus not detected by any method with complete set of specimens available

6 Virus not detected by any method with incomplete set of specimens available

value of 12 different surveillance case definitions for influenza and found predictive values of 12 to 40 per cent.[15] These values were higher than those obtained in the first 2 years of laboratory surveillance in England and Wales when influenza was confirmed using viral isolation and direct IF in only 6 per cent of patients in 1993-4 and 14 per cent in 1994-5.[4] More recent virological surveillance of community influenza in 6 Scottish general practices using PCR, culture and serology confirmed the diagnosis of influenza in 67 per cent of notified cases.[16] The positive predictive value of any test rises with the prevalence. In the 2 years of this study we were able to demonstrate a lower positive predictive value for influenza diagnosis in the early and later weeks of the season (43%) compared with the higher prevalence weeks (53%); this difference was not significant.

There are many different definitions of influenza for surveillance purposes[15] and the utility of these definitions is related both to the clinical symptoms included in the definition and the sensitivity of the tests against which the definitions are evaluated. The ASPREN criteria have recently been evaluated as the ICHPP-2 in people aged 65 years and over.[5] The use of all the ASPREN/ICHPP-2 criteria proved more likely to predict an ILI than influenza, and the evaluation concluded that the 3 criteria of sudden onset, fever and cough could be used as a reliable surveillance definition of influenza. Using these criteria, influenza was confirmed in 30 per cent of patients in the ICHPP-2 criteria evaluation study, with serological testing as the only test for laboratory confirmation of influenza infection. In our patients, using a combination of laboratory tests, the symptoms identified in the ICHPP-2 evaluation study predicted influenza in 54 per cent of patients with an ILI. This is similar to the proportion predicted by all the ASPREN criteria but has the distinct advantage of being a much simpler surveillance definition. We are currently reviewing various influenza case definitions using surveillance data from Victoria and Western Australia.

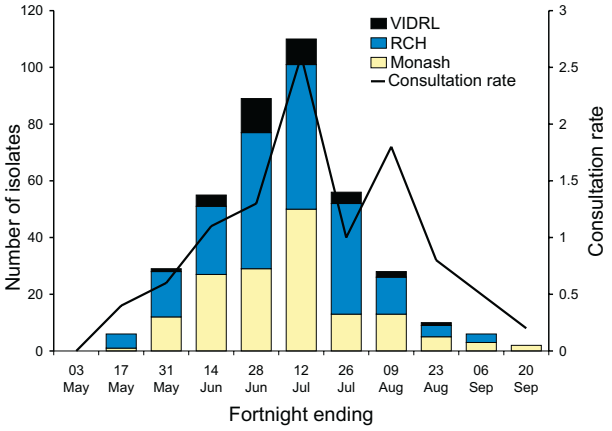
Although travel may play an important part in the global spread of the influenza virus, our data do not show a significantly increased risk of infection in people who had travelled recently or who had been in contact with travellers. Both the destination and the timing of international travel will impact on the likelihood of any association between travel and infection.

Laboratory confirmation of influenza infection from patients in sentinel general practices has been established in Victoria and continues to be refined. Because a number of laboratory tests, including convalescent serology, were used in the present study, the confirmation of influenza infection amongst patients notified with ILI was relatively high (49-54%). Pattern differences in influenza surveillance from laboratories in teaching hospitals and ILI in general practice may relate to the proportion of patients with ILI in general practice proven to have influenza and the different age groups and disease severity in the two surveillance systems.

***Abbreviations**

ASPREN, Australian Sentinel Practice Research Network; DHS, Department of Human Services; GP, general practitioner; ICHPP-2, International Classification of Health Care Problems in Primary Care; ILI, influenza-like illness; PCR, polymerase chain reaction; VIDRL, Victorian Infectious Diseases Reference Laboratory.

Figure 2a. Hospital admission based influenza surveillance compared with influenza-like illness surveillance in sentinel general practices, 1998, by fortnight*



*Monash Monash Medical Centre, Clayton, Victoria

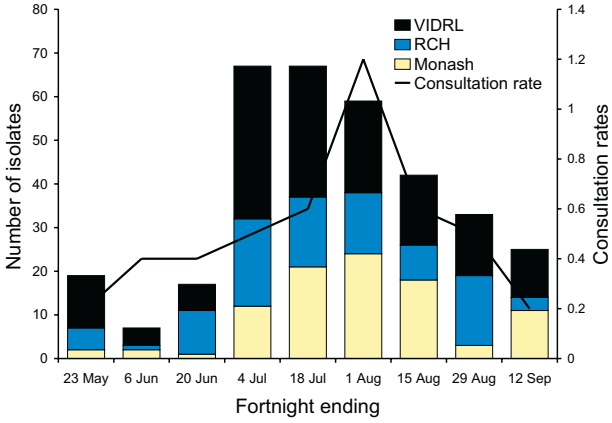
RCH Royal Children's Hospital, Parkville, Victoria

VIDRL Victorian Infectious Diseases Reference Laboratory, North Melbourne, Victoria

Consultation rate the number of patients with influenza-like illness as a proportion of all patients in sentinel general practice sites

Influenza notifications from all laboratories, including VIDRL, are from hospital in-patients. Cases of influenza confirmed from general practice ILI surveillance are not included in the surveillance from VIDRL.

Figure 2b. Hospital admission based influenza surveillance compared with influenza-like illness surveillance in sentinel general practices, 1999, by fortnight*



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