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Original Article

Epidemiological study of influenza virus infections in young adult outpatients from Buenos Aires, Argentina

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Background Influenza virus is the most common cause of influenza-like illness (ILI) in adults. In Argentina, studies on influenza and other respiratory viruses were performed mostly in pediatric populations.

Objectives To determine: (1) the frequency of influenza virus and other common respiratory viruses in adult outpatients with ILI, (2) whether the signs and symptoms predict viral etiology, (3) whether viral diagnosis changes clinical management or infection control measures and (4) to characterize the influenza strains circulating in the community.

Population and methods Nasal and pharyngeal swabs from adult outpatients with ILI attending the emergency room during the winter seasons of 2004 and 2005 in Argentina were evaluated by immunofluorescence and RT-PCR.

Results Of 151 samples analyzed, 39 (26%) were influenza A positive, 5 (3·3%) influenza B positive and 4 (2·6%) respiratory syncytial virus positive by immunofluorescence. Two samples (1·3%) were human metapneumovirus positive by RT PCR. Cell culture detected six additional influenza viruses and one adenovirus positive sample. The sensitivity of immunofluorescence for influenza compared with culture was 70%. Symptoms did not predict etiology.

Conclusions In this study, 40% of the patients with ILI had a specific viral infection and 83% were influenza viruses. Viral detection was necessary to determine the etiology as signs and symptoms were not different between patients with or without viral infection. Viral diagnosis was important to implement infectious control measures. Circulating influenza strains in this study were similar to the correspondent vaccine strains selected for the Southern hemisphere.

Keywords Adult outpatients, infection control, influenza virus, influenza-like illness, upper respiratory tract infections.

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Introduction

Viral respiratory infections are the most common acute illness worldwide. Influenza (Flu) virus is the most frequent cause of influenza-like illness (ILI) in adults. They usually present as abrupt, self-limited febrile infections that occur in outbreaks almost every winter. The attack rates during such periods have been reported to be as high as 10–40%.[1] Other viruses implicated are respiratory syncycial virus (RSV), adenovirus, parainfluenza, rhinovirus and in a lower frequency, coronavirus and enterovirus.[2] New viruses such as human metapneumovirus (hMPV), bocavirus, the new coronaviruses (NL 63) and polyomavirus (KI, WU) have been recently identified. Limited information is available about their significance and frequency in non-hospitalized adult patients although they can also cause severe respiratory illness.[3–5]

All age groups are affected by Flu infection but serious complications occur more frequently in young children, immunocompromised patients and the elderly.[6] These infections can lead to inadequate antibiotic use, numerous outpatient visits, and increased mortality in elderly nursing home patients.[7] Several studies have reported that other viruses such as RSV[8] and even bacteria can cause similar signs and symptoms. Therefore, rapid viral diagnosis is important to install antiviral treatment and to detect Flu outbreaks.

Surveillance studies have been done mainly in developed countries, and data from Argentina come mainly from pediatric populations.[9]

The purpose of this study was to determine the frequency of influenza virus in young adult outpatients with ILI and to compare with the frequency of the other common respiratory viruses (RSV, adenovirus, parainfluenza viruses as well as hMPV). In addition, we evaluated whether the signs and symptoms predicted viral etiology and whether the use of indirect immunofluorescence (IIF) changed the clinical management and infection control measures. Flu strains recovered from this study were compared with those recommended for the annual Flu vaccine.

Population and methods

Adult outpatients with ILI attending the emergency room of a University Hospital in Buenos Aires, Argentina (Centro de Educación Médica e Investigacion Clínica, CEMIC) during two consecutive winters (May–September 2004 and June–September 2005), were included. These periods corresponded to the 18–39 and 24–34 epidemiological weeks, respectively. Influenza-like illness was defined as fever (>38°C) at least once during the last 72 hours of symptoms and at least one upper respiratory sign/symptom (cough, nasal discharge, throat sore) and at least one of the following systemic sign/symptom: headache, myalgias, malaise, chills, prostration. The patients who gathered the inclusion criteria were invited to read and sign the informed consent and those who accepted were included in the study.

Throat and nasal swabs were collected at the emergency room and sent in viral transport media to the Virology Laboratory. Clinical data including age, symptoms, Flu vaccination, presence of immunosupression and clinical management at the end of the visit were recorded in a form specially designed for this study. The attending physician at the emergency room decided whether the patient required symptomatic and/or antiviral or antibiotic treatment. This study was approved by the local IRB.

Virological studies

Samples were processed by IIF with monoclonal antibodies (Chemicon, Temecula, CA, USA) for Flu A, Flu B, parainfluenza, adenovirus and RSV. All samples (n = 151) were tested by IIF and RT-PCR for hMPV detection. In addition, specimens from year 2004 were retrospectively cultured for Flu and adenovirus detection by inoculation in MDCK and HEp-2 cell lines.

Follow up

Patients with a positive viral result by IIF were telephoned within 24 hours by the principal investigator to evaluate any complications, evaluate the utility of viral treatment, and suggest infection control measures. When a complication was suspected, the patient was reexamined.

Flu subtyping

Flu positive samples were sent to the National Center of Influenza (INES-ANLIS Carlos G. Malbrán) for characterization of the strains by hemoagglutination-inhibition test.

Statistical analysis

Student's t-test was used to analyze continuous variables and chi-square test for categorical ones. Relationship between variables was evaluated using Spearman's correlation coefficient and Pearson's correlation coefficient for categorical and continuous variables respectively. Statistical significance was assumed for P values of less than 0·05.

Logistic regression models were used to calculate odds ratios and its respective confidence intervals.

Results

A total of 152 specimens were evaluated (48 from 2004 and 104 from 2005).

Virological studies

Of the 152 specimens received at the laboratory, 151 (99%) were adequate in cells for diagnosis by IIF. For year 2004, 11 samples were Flu A positive and 3 were Flu B positive by IIF. Cell culture allowed to detect three more Flu A cases; three more Flu B cases and one adenovirus case (Table 1). The sensitivity of IIF compared to cell culture for Flu was 70%.

Epidemiology

During both years, circulation of Flu A was detected up to week 30, while few cases of Flu B were observed during the whole studied period. RSV was only detected in 2005.

All Flu A strains were subtyped as H3. For 2004, four Flu A isolates were similar to the 2004 vaccine strain A/Fujian/411/02 (H3N2) and four were antigenically different from vaccine virus, but closely related to A/Wellington/1/2004 (H3N2). The rest could not be characterized. For 2005, characterization was achieved in 13 samples only, 11 Flu A and 2 Flu B. All Flu A strains were characterized as A/California/7/04 (H3N2) and were closely related to 2005 vaccine strain A/Wellington/1/2004 (H3N2). For Flu B, one strain was similar to B/Hong Kong/330/01 (Victoria lineage) vaccine strain for 2004, and the other corresponded to B/Shangai/361/02 (Yamagata lineage), vaccine strain for the Southern hemisphere for 2005.

Clinical data

The mean age of the patients was 36·6 years (SD 13·6), predominantly female (60%). There were no statistical differences between the positive and negative groups regarding sex or age (Table 2).

Table 1. Detection of respiratory viruses from adults with ILI in the emergency room in Argentina

Detected virus	Year						
	2004 (*n* = 48) Positive by IIF	Positive by cell culture	RT-PCR	2005 (*n* = 103) Positive by IIF	RT-PCR	Total (*n* = 151) Positive by IIF	RT-PCR
Flu A*	11 (23)	14 (29)	nd	28 (27)	nd	39 (26)	nd
Flu B**	3 (6)	6 (13)	nd	2 (2)	nd	5 (3·3)	nd
Adenovirus	0	1 (2)	na	0	na	1 (0·6)	na
Parainfluenza	0	0	nd	0	nd	0	nd
RSV	0	0	nd	4 (4)	nd	4 (2·6)	nd
hMPV	na	na	1 (2)	na	1 (1)	na	2 (1·3)

Values are given as *n* (%). na, not applicable; nd, not done.
*Flu A circulating strains in year 2004, A/Fujian/411/02 and A/Wellington/1/2004; in year 2005, A/California/7/04.
**Flu B circulating strains in year 2004, B/Hong Kong/330/01; in year 2005, B/Shangai/361/02.

Table 2. Epidemiological and clinical characteristics of adult outpatients with ILI in Argentina

Characteristics	Population (*n* = 151)	Virus negative (*n* = 96)	Virus positive (*n* = 55)
Male sex	60 (40%)	43 (45%)	18 (33%)
Age (years)	36·6 (SD 13·6)		
Time to consult (hours)	43 (SD 20)	46 (SD 20)	40 (SD 21)
Immunocompromised individuals	14 (9%)	11 (7%)	2 (1%)
Vaccination for influenza	12 (8%)	10 (7%)	2 (4%)
Signs and symptoms			
Fever	151 (100%)	96 (100%)	55 (100%)
Pharyngeal pain	99 (65%)	62 (64%)	37 (67%)
Cough	137 (90%)	82 (85%)	55 (100%)
Nasal discharge	127 (84%)	77 (80%)	50 (90%)
Headache	87 (58%)	53 (55%)	32 (58%)
Myalgias	102 (68%)	64 (67%)	38 (69%)
Postration	112 (74%)	71 (73%)	41 (75%)
Malaise	124 (82%)	77 (80%)	47 (85%)
Chills	82 (54%)	46 (48%)	36 (65%)

Of 151 patients, only 12 (10%) had received the annual Flu vaccine. One of them was an immunocompromised HIV-infected patient who was Flu A positive.

None of the signs and symptoms were significantly associated with positive or negative results (Table 2). No complications were reported and none of the patients required hospitalization.

Clinical management

Only one Flu A positive patient received empirical oseltamivir at the emergency room because she had a renal transplant and was under immunosuppressive treatment. The rest of the patients consulted in an average of 43 hours after the symptoms started. In addition, viral diagnosis by IFI was not always available in the same day. As oseltamivir is effective within 48 hours of the onset of symptoms, we could not implement treatment in our patients.

Inadequate use of antibiotics was not observed in this study. Only two Flu negative patients were given empirical antibiotics, as they developed bacterial pneumonia and had to be hospitalized.

Discussion

In this study, 38% of the young adult outpatients with ILI had a viral etiology, 33% corresponded to Flu virus followed by RSV (3%). Other studies have shown similar or even lower viral detection in young adults.[10–12] In this study we have only investigated the common respiratory viruses, which are routinely detected by conventional

immunofluorescence assays with monoclonal antibodies or by cell culture. Therefore, if molecular methods would have been used, viral detection would have possibly been higher.[13] This concept is especially true for viruses such as RSV for which IIF is not recommended for diagnosis in adults.[14] Nevertheless, in all studies a significant proportion of patients with ILI remain without etiological diagnosis. This leads us to the inevitable question: what other agents cause the majority of the ILI in our adult population? Other viruses such as rhinovirus, enteroviruses, the new coronavirus and bocavirus could be implicated, but it would not be surprising if new viruses causing ILI were discovered in the near future.

Moreover, the application of molecular methods had revealed the existence of coinfections with two or more respiratory viruses. The clinical significance of these findings has yet to be determined.

No correlation was observed between specific signs and symptoms and etiology. This reinforces the need for a rapid viral diagnosis in order to define etiology, prevent indiscriminate use of antibiotics, reduce unnecessary interventions and implement infection-control measures including isolation of infected individuals. Lina and Valette. also observed that viral etiology in patients with ILI disease cannot be predicted on the basis of their clinical picture only.[15]

Flu virus only responds to the use of antivirals if the treatment is started within the first 2 days of the onset of fever. In our study, most of the patients consulted after 48 hours of the onset of symptoms. In addition, viral diagnosis by IFI was not available in the same day. Therefore, our results were not useful in a timely manner for antiviral treatment. Rapid antigen devices for Flu which can give a result within the hour are useful in these cases. On the other side, viral etiology permitted infection control measures, like patient isolation. This is particularly important in the high-risk population such as immunocompromised patients and their close contacts. In addition, viral etiology contributed to epidemiological data and permitted identification of circulating strains.

Most of Flu strains from our patients were similar to the corresponding vaccine strains for the Southern hemisphere for the correspondent year. Diagnosis and specific characterization of Flu strains is important in order to provide early warning on a potential Flu pandemic or epidemic.

In conclusion, viral diagnosis was achieved in almost 40% of the patients with ILI. The most frequent virus was Flu A followed by Flu B and RSV. Human metapneumovirus was detected in only 1·3% of the cases, similar to what was described previously.[16]

This study represents the first one evaluating viral etiology in young adult outpatients with ILI in Argentina.

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References

1 Mandell G, Bennett J, Dolin R. Principles of Infectious Diseases 5th edn. Philadelphia, Pennsylvania: Churchill Livingstone, 2000.
2 Lin T-Y, Huang Y-C, Ning H-C, Tsao K-C. Surveillance of respiratory viral infections among pediatric outpatients in Northen Taiwan. J Clin Virol 2004; 30:81–85.
3 Van den Hoogen B, de Jong J, Groen J *et al.* A newly discovered human metapneumovirus isolated from young children with respiratory tract disease. Nat Med 2001; 6:719–724.
4 Drosten C, Günther S, Preiser W *et al.* Identification of a novel coronavirus in patients with severe acute respiratory syndrome. N Engl J Med 2003; 348:1967–1976.
5 Ksiazek T, Erdman D, Goldsmith C *et al.* A novel coronavirus associated with severe acute respiratory syndrome. N Engl J Med 2003; 348:1953–1966.
6 Ayora-Talavera G. Detection of human influenza virus in Yucatán, Mexico. Rev Invest Clin 2002; 54:410–414.
7 Drinka PJ, Gravenstein S, Langer E, Krause P, Shult P. Mortality following isolation of various respiratory viruses in nursing home residents. Infect Control Hosp Epidemiol 1999; 20:812–815.
8 Zambon MC, Stockton JD, Clewley JP, Fleming DM. Contribution of influenza and respiratory syncytial virus to community cases of influenza-like illness: an observational study. Lancet 2001; 358:1410–1416.
9 Carballal G, Videla C, Espinosa M *et al.* Multicentered study of viral acute lower respiratory infections in children from four cities of Argentina 1993–1994. J Med Virol 2001; 64:167–174.
10 Govaert TM, Dinant GJ, Aretz K, Knottherus JA. The predictive value of influenza symptomathology in elderly people. Fam Pract 1998; 15:16–22.
11 Brydak LB, Lietzau G, Machała M. Diagnostics of viral respiratory infections in hospitalized patients and ambulatory patients from SENTINEL program during 2004/05 season in Poland. Pol Arch Med Wewn 2005; 114:958–967.
12 Smith KJ, Roberts MS. Cost-effectiveness of newer treatment strategies for influenza. Am J Med 2002; 113:300–307.
13 Kuypers J, Wright N, Ferrenberg J, Huang M, Cent A, Corey L. Comparison of real time PCR assays with fluorescent-antibody assays for diagnosis of respiratory viruses in children. J Clin Microbiol 2006; 44:2382–2388.
14 Falsey AR, Wash E. Respiratory syncytial virus infection in adults. Clin Microbiol Rev 2000; 13:371–384.
15 Lina B, Valette M. Surveillance of community-acquired viral infections due to respiratory viruses in Rhone-Alpes (France) during winter 1994 to 1995. J Clin Microbiol 1996; 34:3007–3011.
16 Stockton J, Stephenson I, Fleming D, Zambon M. Human metapneumovirus as a cause of community-adquired respiratory illness. Emerg Infect Dis 2002; 8:897–901.

