Epidemiology of viral respiratory infections in a tertiary care centre in the era of molecular diagnosis, Geneva, Switzerland, 2011–2012

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Abstract

Few studies have examined the epidemiology of respiratory viral infections in large tertiary centres over more than one season in the era of molecular diagnosis. Respiratory clinical specimens received between 1 January 2011 and 31 December 2012 were analysed. Respiratory virus testing was performed using a large panel of real-time PCR or RT-PCR. Results were analysed according to sample type (upper versus lower respiratory tract) and age group. In all, 2996 (2469 (82.4%) upper; 527 (17.6%) lower) specimens were analysed. Overall positivity rate was 47.4% and 23.7% for upper and lower respiratory samples, respectively. The highest positivity rate was observed in patients under 18 years old ($p < 0.001$); picornaviruses were the most frequent viruses detected over the year. Influenza virus, respiratory syncytial virus, human metapneumovirus and coronaviruses showed a seasonal peak during the winter season, while picornaviruses and adenoviruses were less frequently detected in these periods. Multiple viral infections were identified in 12% of positive cases and were significantly more frequent in children ($p < 0.001$). In conclusion, we observed significant differences in viral infection rates and virus types among age groups, clinical sample types and seasons. Follow-up of viral detection over several seasons allows a better understanding of respiratory viral epidemiology.

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Introduction

Respiratory viral infections are a leading cause of morbidity and mortality, particularly in children, the elderly and immunocompromised persons. Rapid identification of viral aetiology is critical to avoid unnecessary antibiotics, to initiate antiviral treatment when available and to limit the spread of the infection [1].

Nucleic acid-based amplification tests (NATs) allow sensitive detection of a broad panel of both conventional and emerging viruses in respiratory tract specimens. NATs are more sensitive than any other diagnostic method, including virus isolation in cell culture and antigen detection, and now form the backbone of clinical virology laboratory testing around the world. This advance has changed the landscape of virus detection and highlights the need to better understand the epidemiology of viruses ranging from rhinoviruses to influenza virus [2,3].

Most of the available literature describing the epidemiology of respiratory viruses is focused on the paediatric population, other particular populations, or specific viral agents; studies are frequently limited to one season. A longitudinal examination of the epidemiology of viral respiratory agents among patients frequenting a large university centre is lacking; prevalence patterns in this group may differ from those in the community and may be subject to seasonal variation.

Our study aims to describe the general molecular epidemiology of viral respiratory infections in paediatric, adult and elderly populations admitted to or screened in a tertiary care centre over a 2-year period, and, more specifically, to

compare the epidemiological patterns of upper and lower respiratory specimens.

Methods

The University Hospital of Geneva is a tertiary care teaching hospital with 1600 acute-care beds and >40 000 admissions each year. It is comprised of surgical and internal medicine services, as well as bone-marrow and solid-organ transplant units. The virology laboratory of the University Hospital of Geneva processes all clinical specimens from adult and paediatric inpatient and outpatient departments.

Clinical specimens

All respiratory specimens (nasopharyngeal swabs and aspirates, bronchial and/or tracheal aspirates and bronchoalveolar lavage (BAL) specimens) from adult and paediatric inpatients and outpatients received between 1 January 2011 and 31 December 2012 were included in the study. Nasopharyngeal swabs and aspirates were grouped as 'upper respiratory samples', while bronchial and tracheal aspirates, as well as BAL specimens, were considered 'lower respiratory samples'. Further, specimens were considered 'paediatric' if they were from patients under 18 years of age, 'adult' if from patients between 18 and 65 years old, and 'elderly' if from patients over age 65. Collection of samples for analysis was not systematic but was based on clinical judgement according to local practice and guidelines that recommend viral screening in patients at risk of lower respiratory complications and systematically in transplant recipients, in hospitalized patients with an acute respiratory disease during the influenza season, and in those that do no respond to the usual empirical antibiotic treatment. However, detection of viral pathogens was systematic in BAL obtained from immunocompromised patients such as transplant recipients.

Viral real-time PCR detection

Each specimen was screened by nucleic acid detection for the presence of influenza A (both seasonal and 2009 pandemic H1N1 strains) or B virus, respiratory syncytial virus (RSV) A and B, parainfluenza virus 1–3, human metapneumovirus, rhinovirus A, B and C, enterovirus, adenovirus and, beginning on 23 August 2011, coronaviruses 229E, OC43, HKU1 and NL63. Screening was performed using individual one-step real-time Taqman©-based PCR or RT-PCR as described previously. Respiratory specimens were extracted using Easymag© (bioMérieux, Geneva, Switzerland) according to the manufacturer's recommendations. The viral real-time PCR detection was performed as described for parainfluenza viruses 1 and 3 [4], influenza viruses, RSV A and B, coronaviruses, parainfluenza virus 2, human metapneumovirus, coronaviruses, adenoviruses [5, 6] enteroviruses and rhinoviruses [7].

Extraction, presence of PCR inhibitors, and reverse transcription were controlled by spiking each specimen with a quantified standard of canine distemper virus; experiments were validated only if the resulting cycling threshold value was within the expected ranges. PCR detection was considered positive if the cycling threshold value was ≤39.

In addition to molecular tests, rapid tests for antigen detection (data not shown) are routinely used for influenza A and B viruses (Bionexia; bioMérieux, Lyon, France) and RSV (Quickvue; Quidel, San Diego, CA, USA) detection in the paediatric emergency wards.

Definition of clinical episodes

For patients with specimens that were repeatedly positive for the same viral agent in a period shorter than 3 weeks, only the first positive result was considered. If the interval between positive specimens was 3 weeks or longer, a different episode was assumed. A separate clinical episode was also assumed for patients with different viruses detected throughout their clinical course, independent of the time elapsed between sampling. If two upper or two lower respiratory samples were sent at the same time or within 3 days of one another, these were considered a single clinical episode if both were positive. Simultaneous detection of more than one virus in the same sample was considered as a single clinical episode with a multiple viral infection (MVI).

Statistical analysis

The independent *t*-test was used to compare continuous data, while the chi-squared test was used to compare categorical data. Associations with a p value <0.05 were considered significant. All data were analysed within IBM SPSS Statistics for Windows, Version 20.0 (IBM Corp, Armonk, NY, USA).

Results

During the study period, 2996 clinical respiratory specimens were analysed, with a total of 32 387 real-time PCRs or RT-PCRs performed. Upper respiratory samples predominated, representing 82.4% of all cases (2469); 17.6% (527) were lower respiratory samples. Among the 527 lower respiratory samples, 451 (85.6%) were collected via BAL. The overall positivity rate (PR) for any respiratory virus was 43.2%, while for upper and lower respiratory samples it was 47.4% and 23.7%, respectively (Fig. 1). Most (89%) nasopha-