Respiratory Virus Shedding in a Cohort of On-Duty Healthcare Workers Undergoing Prospective Surveillance

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Abstract

Background—Healthcare-associated transmission of respiratory viruses is a concerning patient safety issue.

Design—Surveillance for influenza virus among a cohort of healthcare workers (HCWs) was conducted in a tertiary care children’s hospital from November 2009 until April 2010, using biweekly nasal swab collection. If a subject reported respiratory symptoms, an additional specimen was collected. Specimens from ill HCWs and a randomly selected sample from asymptomatic subjects were tested for additional respiratory viruses by multiplex PCR.

Results—From 170 enrolled subjects, 1404 nasal swabs were collected. Influenza circulated at very low levels during the surveillance period and 74.2% of subjects received influenza vaccination. Influenza was not detected in any specimen. Multiplex respiratory virus PCR analysis of all 119 samples from symptomatic subjects and 200 specimens from asymptomatic subjects yielded a total of 42 positive specimens; 7 (16.7%) in asymptomatic subjects. Viral shedding was associated with report of any symptom (OR 13.06, p<0.0001, 95% CI 5.45-31.28) and younger age (OR 0.96, p=0.023, 95% CI 0.92-0.99) when controlled for gender and occupation of physician or nurse. After the surveillance period, 46% of subjects reported working while ill with an influenza-like illness during the previous influenza season.

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All other authors declare no conflicts of interest.
Conclusions—In this cohort, HCWs working while ill was common, as was viral shedding among those with symptoms. Asymptomatic viral shedding was infrequent, but did occur. HCWs should refrain from patient care duties while ill, and staffing contingencies should accommodate them.

Introduction

Healthcare-associated spread of respiratory viruses has been known to occur, and acquisition of viral respiratory tract infection can result in severe disease in immunocompromised and other high-risk patients. Transmission of respiratory viruses arises most frequently from contact with symptomatic individuals, although subclinical infection has been documented and may have a role in transmission. Therefore, guidelines for preventing pathogen transmission in healthcare settings have emphasized restriction of ill healthcare workers (HCWs) and visitors from patient care areas. However, HCWs are known to work while ill rather than take sick leave (a.k.a. “presenteeism”), thereby potentially exposing vulnerable patients to contagious pathogens. In previous reports, younger HCWs commonly reported working despite being ill, in part due to a concern about placing a burden on their colleagues. Community surveillance for respiratory viruses, including HCW cohorts, has detected shedding of rhinovirus and influenza in the outpatient setting, but published studies of surveillance for respiratory viruses among HCWs who are actively working in inpatient care areas are lacking. This study was initially designed as surveillance for influenza virus in on-duty HCWs; however, because of low circulation of 2009 H1N1 pandemic influenza (pdmH1N1) in Nashville after initiating surveillance for the study, other respiratory viral pathogens in both symptomatic and asymptomatic on-duty HCWs were assessed.

Study Design

Surveillance for influenza virus took place among a cohort of HCWs serving inpatient areas in Monroe Carell Jr. Children’s Hospital at Vanderbilt (MCJCHV) in Nashville, Tennessee during the 2009-2010 pandemic influenza season from November 16, 2009 through April 16, 2010, a 20-week period during which seasonal influenza was expected to circulate. The study was originally designed as surveillance for seasonal influenza, prior to emergence of the pandemic strain.

Methods

After approval of the study protocol by the Vanderbilt University Institutional Review Board, HCWs provided informed consent and were recruited from two floors of MCJCHV, a 238-bed free-standing children’s tertiary care hospital. These units included pediatric critical care, hematology/oncology, cardiology, and general pediatrics units. The study was announced as the “SWIS Study” (Surveillance of healthcare Workers for Influenza Shedding) through educational flyers, mass e-mail advertisements, and visits to staff meetings. Volunteers were eligible for study if they were at least 18 years old and anticipated employment throughout the study period. Since sample collection was offered during daytime hours for logistical reasons; HCWs were excluded if they worked nights.
only. Individuals who anticipated being away from work responsibilities for greater than 1 month during surveillance were also excluded, with the exception of pregnant staff members who were expected to return to work before April 30, 2010. An enrollment questionnaire captured demographic information, comorbid conditions, medication use including immunosuppressants, smoking, pregnancy status, type of patient care responsibility, length of employment at Vanderbilt University Medical Center (VUMC), and children <18 years of age in the household. Volunteers consented to review of vaccination records in the Occupational Health Clinic at Vanderbilt to confirm self-report of influenza vaccination.

Study volunteers presented to specimen collection sites in a central location for collection of nasal swabs for influenza PCR testing and simultaneous documentation of symptoms of illness, if present. Specimens were collected every 2 weeks, and subjects were instructed to return for an additional sample collection if they felt ill while working before the next routine collection was due; after this collection, they were instructed to return for their next routine sample collection 2 weeks later. Investigators were blinded to results of influenza PCR testing until after the surveillance period, and individual test results were not reported to study volunteers. Serum samples were collected pre- and post-surveillance dates for influenza hemagglutination-inhibition testing using a reference strain of pdmH1N1, A/California/04/200919; virus and serial dilutions of patient sera were incubated with turkey erythrocytes, and the hemagglutination-inhibition assay titer was defined as the highest serum dilution that completely inhibited hemagglutination. Following study completion, nasal specimens from all symptomatic HCWs collected at the time of symptom report and a randomly selected sample of specimens collected from asymptomatic HCWs underwent additional testing with multiplex PCR targeted against additional respiratory viruses. Further analysis was performed using multivariable logistic regression.

Nasal swabs were collected by a trained, non-physician staff member every 2 weeks within each of the enrolling units during the study period. The study did not use nasopharyngeal swabs that can yield higher viral detection rates than nasal swabs, due to the concern that recruiting and retaining HCWs for repeated specimens would be difficult with the nasopharyngeal swab technique, which can be uncomfortable. Polyester-tipped applicators were used to collect nasal specimens. Volunteers were given a gift card with each sample collection as an incentive to continue participating. RNA was extracted from nasal specimens by MagNA Pure LC (Roche Diagnostics) as has been described previously. To detect influenza virus, a real time PCR (RT-PCR) assay based on CDC-designed primers was performed on all specimens. Positive results were reported as influenza A unsubtyped, H1, H3, pdmH1N1, or influenza B. As an internal control, β-actin mRNA was targeted to confirm specimen adequacy and absence of endogenous PCR inhibitors. To test for additional respiratory viruses other than influenza, the MultiCode-PLx-RVP assay was performed for the following viruses: respiratory syncytial virus (RSV), human metapneumovirus (HMPV), rhinovirus (HRV), parainfluenza (PIV), coronavirus, adenovirus, human bocavirus, and enterovirus; this assay utilizes multiplex PCR chemistry (EraGen MultiCode-PLx technology) coupled with high-throughput microsphere flow cytometry (Luminex). Only specimens deemed an adequate specimen by detection of β-actin mRNA were included in the analysis.
We aimed to enroll a sample size of 150 subjects to follow as a prospective cohort to detect symptomatic and asymptomatic influenza infection in 1-5% of vaccinated study subjects and 5-20% of unvaccinated study subjects, assuming a 5-10% annual influenza attack rate, 50% influenza vaccination rate, and 75% influenza vaccine efficacy in healthy adults. In addition, 33% of infections were expected to be asymptomatic based on prior investigations. For univariate analyses, nominal data were compared using chi-square, and continuous data compared using the Wilcoxon rank-sum test. Comparison of multiple categories was done using Kruskal-Wallis one-way analysis of variance. A redundancy analysis was done to ensure that the effect of each variable was not explained by another variable. Study data were collected and managed using REDCap electronic data capture tools hosted at Vanderbilt University. Variables of interest for multivariable logistic regression were determined a priori, and included report of any symptom, age, occupation of physician or nurse vs. other occupation, and gender in the model that encompassed all specimens that underwent multiplex PCR testing, and variables for the model restricted to specimens from ill HCWs included age, occupation of physician or nurse vs. other occupation, and gender.

Results

The study enrolled 170 HCWs. The median age of enrollees was 31 years, with 81.8% women. The racial identification of subjects was as follows: 145 (85.3%) White, 21 (12.4%) African American, 3 (1.8%) Asian, and 1 (0.6%) American Indian or Alaska Native; 2 (1.4%) enrollees were of Hispanic ethnicity. The areas from which volunteers were recruited were staffed by a pool of approximately 240 nurses, 60 attending physicians and fellows, 80 resident physicians, and ancillary staff. In the cohort, there were 39 (22.9%) physicians, 64 (37.6%) nurses, and 67 other HCWs whose job involved work in patient care areas, including 13 (7.6%) social workers, 13 (7.6%) environmental services workers, 12 (7.1%) radiology technicians, 10 (5.9%) medical receptionists, 8 (4.7%) respiratory therapists, 3 (1.8%) hospitality workers, 3 (1.8%) food services workers, 2 (1.2%) clinical pharmacists, 1 (0.6%) dietitian, and 1 (0.6%) administrator, and 1 (0.6%) designated as “Other.” About one-third of individuals had a child less than 18 years of age in their household (36.1%), and 81.8% had worked more than one year at the institution. Eleven HCWs enrolled but never returned for surveillance activities. A total of 1404 nasal swab specimens were collected from 159 HCWs (median 10 specimens per volunteer, interquartile range [IQR] 8-11), of which 119 were collected from HCWs reporting concurrent illness. The 119 specimens from ill HCWs were obtained from 83 HCWs, as 29 of these individuals (24.3%) returned multiple times for sample collection during distinct illnesses. The 200 randomly selected specimens corresponded to 106 volunteers. As a control, β-actin mRNA was detected in all 119 specimens from subjects reporting symptoms and in 1270 (98.8%) specimens from subjects who were asymptomatic.

The US 2009-2010 influenza season was atypical, with the emergence of pdmH1N1 and subsequent lack of widespread seasonal influenza virus circulation in the US. According to active, prospective, population-based influenza surveillance among residents of Davidson County, TN, pdmH1N1 virus circulation diminished after November, and remained low for the duration of the study period. In addition, 74.2% of the cohort was vaccinated against...
this strain (compared to a 48.9% institutional and 37.1% national vaccination rates)\textsuperscript{25}. Influenza infection was not detected in any subject by PCR or serologic assays.

Multiplex PCR of samples from symptomatic HCWs (n=119) and 200 randomly selected specimens from asymptomatic HCWs revealed 42 positive specimens, with HRV identified in 33 specimens (78.6%), PIV in 4 (9.5%), coronavirus in 4 (9.5%), and HMPV in 1 (2.4%, Table 1). Of the 42 positive specimens, 35 (29.4%) were from ill HCWs (n=119), and 7 (3.5%) from asymptomatic HCWs (n=200). Univariate analysis was performed on all specimens that underwent multiplex PCR testing; a positive test was associated with the presence of cough (p<0.001), rhinorrhea (p<0.001), sore throat (p<0.001), myalgias (p<0.001), headache (p<0.001), and fatigue (p=0.014, Table 2). In multivariable logistic regression, identification of a virus was strongly associated with a report of feeling ill (OR 13.06, p<0.0001, 95% CI 5.45-31.28) and younger age (OR 0.96, p=0.023, 95% CI 0.92-0.99), when controlled for gender and occupation as physician or nurse vs. all other job types (Table 3). When regression was restricted to specimens collected from symptomatic subjects, younger age was positively associated with viral shedding (OR 0.94, p=0.014, CI 0.90-0.99), and physician or nurse occupation was negatively associated with viral shedding (OR 0.34, p=0.018, 95% CI 0.14-0.83), when controlled for gender. In a web-based questionnaire after the surveillance period, there were 141 respondents (82.9% of subjects), including 54 nurses and 36 physicians (38.3% and 25.5% of post-surveillance questionnaire respondents, respectively). Of all respondents, 39 (27.6%) reported having an influenza-like illness (ILI), using the CDC definition of ILI (fever with cough and/or sore throat), during the previous influenza season. Of the 39 respondents who reported an ILI, 17 (43.4%) were nurses and 5 (12.8%) were physicians. Overall, 18 (46.1%) of all respondents who recalled ILI continued to work despite being ill. Of the 17 nurses who recalled ILI, 8 (47.1%) continued to work through the illness, as did 4 (80%) of the 5 physicians.

**Discussion**

Respiratory viruses are important causes of morbidity and mortality among high-risk patients, with RSV, influenza, PIV, HMPV, coronavirus, and HRV known to cause serious and potentially fatal lower respiratory tract infections in the immunocompromised population\textsuperscript{1,4,26,27}. This is the first published prospective study examining respiratory viral shedding among on-duty HCWs in a hospital setting. In this cohort of HCWs, many of whom served high-risk inpatients, continuing patient care duties despite being ill was a common occurrence, and a respiratory virus was identified in 29.4% of symptomatic HCWs. These findings suggest that ill HCWs may subject patients to unnecessary risk. In our cohort, HRV was detected in the majority of positive specimens; while HRV transmits less efficiently than influenza or RSV, and is usually associated with mild illness in the typical host, this pathogen can cause severe disease in high-risk groups, such as those with chronic respiratory disease and immunosuppression, and can even cause severe illness in previously healthy individuals without chronic conditions\textsuperscript{26-29}.

In the subset of specimens that underwent multiplex PCR testing, a positive test result was associated with cough, rhinorrhea, sore throat, myalgias, headache, or fatigue. Multivariable analysis showed a strong association between having any symptom and identifying a
respiratory virus. When multivariable analysis was restricted to specimens from ill HCWs, younger age was significantly associated with having a positive specimen. We hypothesize that younger age of a HCW may indicate a decreased willingness or ability to refrain from professional duties despite illness due to concern about negative social or financial consequences of taking sick leave. In effect, younger HCWs may be working with more severe symptoms, which could explain an increased odds of viral shedding. Additionally, shedding may be associated with higher viral titers, owing to decreased immunologic experience of younger adults. Occupation as a physician or nurse (vs. other occupations) was included as a single variable a priori due to the hypothesis of greater risk of acquiring infections among physicians and nurses because of close proximity to patients, as has been described previously. The finding of a lower odds of having a positive test in those with a physician or nurse occupation compared to other occupations seems paradoxical, however, this observation may be explained by a greater proportion of negative test results among symptomatic nurses compared to physicians in our cohort, leading to apparent lower virus recovery in physicians and nurses who presented for specimen collection while ill. Combining physicians and nurses together as a single variable may have been inappropriate due to differences in the reasons or frequency with which they presented for specimen collection. For example, nurses in the cohort may have been more likely to present for specimen collection with mild symptoms since they tended to have scheduled breaks and closer proximity to nasal swab collection sites, compared to physicians in the study. It does not appear that an increased willingness among nurses to work during an illness accounts for the observed difference, as the post-surveillance questionnaire data suggest nurses in the cohort were more likely than physicians to refrain from patient care duties while ill (52.9% of nurses who recalled ILI took sick leave, compared to 20.0% of physicians).

Initially designed as an assessment of influenza virus shedding in a cohort of HCWs, the study had limitations that affected the likelihood of detecting influenza virus in this group. First, the second wave of pdmH1N1 occurred exceptionally early from August - October 2009, which was prior to the anticipated seasonal influenza season. The decline in local pdmH1N1 activity prior to the study specimen collection period and absence of significant seasonal non-pdmH1N1 influenza circulation in the community during the remainder of the influenza season impacted our findings. In addition, higher vaccination rates in the study cohort may reflect volunteer bias. Further, consent during enrollment for study staff to verify vaccination records at the end of the study may have introduced a Hawthorne effect, leading to a greater willingness to be vaccinated. Another such effect may have occurred if volunteers who may have commonly reported to work during an illness demonstrated increased adherence to the institutional policy to refrain from work until fever has resolved for 24 hours, or to wear a surgical mask during a mild illness with nasal secretions or cough while working to prevent spread of contagion to patients and other HCWs, as these were underscored at enrollment. We observed a low rate of virus identification in specimens submitted for multiplex PCR testing, however the presence of symptoms was based on self-report, and symptomatic individuals did not need to meet any particular threshold or score to be considered ill. Another reason for low viral RNA detection in symptomatic individuals may have been because the sampling site was the nasal cavity rather than the nasopharynx.
Lastly, detecting virus by RT-PCR of nasal swab specimens only suggests the potential for transmission, as studying transmission dynamics was beyond the scope of the investigation.

**Conclusions**

Our data suggest that symptomatic HCWs with detected viral pathogens work while ill and that these individuals may have the potential to transmit respiratory viruses to vulnerable patients in the hospital setting. Detection of virus in asymptomatic participants was infrequent, but did occur. HCWs should be encouraged to avoid patient care duties when ill to prevent exposing patients and others. Updating the institutional illness policy to include indications and parameters for sick leave, staffing contingencies, and removal of financial disincentives may mitigate perceived negative social or financial barriers to taking leave while ill, and therefore should be considered to promote patient safety.

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**References**


### Table 1

**Symptoms Reported for Nasal Swab Specimens that Underwent Multiplex PCR Testing, by Virus Detected**

<table>
<thead>
<tr>
<th>Virus Detected</th>
<th>Asymptomatic</th>
<th>Symptomatic</th>
<th>Cough</th>
<th>Rhinorrhea</th>
<th>Headache</th>
<th>Sore Throat</th>
<th>Myalgias</th>
<th>Fatigue</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rhinovirus (n=33)</td>
<td>5 (15.2%)</td>
<td>28 (84.8%)</td>
<td>15 (45.5%)</td>
<td>25 (75.8%)</td>
<td>16 (48.5%)</td>
<td>20 (60.6%)</td>
<td>9 (27.3%)</td>
<td>9 (27.3%)</td>
</tr>
<tr>
<td>Coronavirus (n=4)</td>
<td>1 (25.0%)</td>
<td>3 (75.0%)</td>
<td>2 (50.0%)</td>
<td>3 (75.0%)</td>
<td>0</td>
<td>1 (25.0%)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>NL63, n=2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>OC43, n=2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Parainfluenza (n=4)</td>
<td>1 (25.0%)</td>
<td>3 (75.0%)</td>
<td>2 (66.7%)</td>
<td>1 (25.0%)</td>
<td>0</td>
<td>1 (25.0%)</td>
<td>1 (25.0%)</td>
<td>0</td>
</tr>
<tr>
<td>PIV 1, n=2</td>
<td></td>
<td></td>
<td></td>
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<td></td>
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<td></td>
<td></td>
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<tr>
<td>PIV 3, n=1</td>
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<td></td>
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<td></td>
<td></td>
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<tr>
<td>PIV 4b, n=1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Metapneumovirus (n=1)</td>
<td>0</td>
<td>1 (100.0%)</td>
<td>1 (100.0%)</td>
<td>1 (100.0%)</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>None detected (n=277)</td>
<td>193 (96.5%)</td>
<td>84 (70.6%)</td>
<td>44 (15.9%)</td>
<td>73 (26.4%)</td>
<td>32 (11.6%)</td>
<td>55 (19.9%)</td>
<td>17 (6.1%)</td>
<td>28 (10.1%)</td>
</tr>
</tbody>
</table>

Analysis of n=319 specimens, with 42 specimens positive for a respiratory virus, and no specimen with >1 virus isolated; Asymptomatic n=200, Symptomatic n=119
### Table 2
Univariate Analysis to Determine Associations between HCW Variables and Outcome of Viral Shedding Detected, using Multiplex PCR

<table>
<thead>
<tr>
<th>Variable</th>
<th>Positive Test (Any Virus)</th>
<th>Negative Test</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cough</td>
<td>20 (30.8%)</td>
<td>22 (8.7%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Runny Nose</td>
<td>30 (29.1%)</td>
<td>12 (5.6%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Headache</td>
<td>16 (34.8%)</td>
<td>26 (9.5%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Sore Throat</td>
<td>22 (29.7%)</td>
<td>20 (8.2%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Myalgias</td>
<td>10 (37.0%)</td>
<td>32 (11.0%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Fatigue</td>
<td>10 (25.6%)</td>
<td>32 (11.4%)</td>
<td>0.014</td>
</tr>
<tr>
<td>Fever &gt;24h Prior</td>
<td>3 (12.7%)</td>
<td>39 (12.7%)</td>
<td>0.217</td>
</tr>
<tr>
<td>Female Gender</td>
<td>36 (13.3%)</td>
<td>6 (12.2%)</td>
<td>0.836</td>
</tr>
<tr>
<td>Age, years, Median (IQR)</td>
<td>29 (27, 33)</td>
<td>30 (27, 41)</td>
<td>0.086</td>
</tr>
<tr>
<td>Occupation as Physician or Nurse</td>
<td>23 (12.4%)</td>
<td>19 (14.2%)</td>
<td>0.649</td>
</tr>
<tr>
<td>Child &lt;18yo at Home</td>
<td>14 (12.4%)</td>
<td>28 (13.6%)</td>
<td>0.761</td>
</tr>
<tr>
<td>Employed &gt;1 year at VUMC</td>
<td>30 (30.3%)</td>
<td>5 (25.0%)</td>
<td>0.635</td>
</tr>
<tr>
<td>Month specimen collected</td>
<td>n/a</td>
<td>n/a</td>
<td>0.446</td>
</tr>
</tbody>
</table>

Analysis of n=319 specimens, with 42 specimens positive for a respiratory virus and 279 specimens negative; VUMC = Vanderbilt University Medical Center
### Table 3
Multivariable Analysis to Determine Associations between HCW Variables and Outcome as Viral Shedding Detected, using Multiplex PCR

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Odds Ratio</th>
<th>95% CI</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Subject symptomatic</td>
<td>13.06</td>
<td>(5.45, 31.28)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Age</td>
<td>0.96</td>
<td>(0.92, 0.99)</td>
<td>0.023</td>
</tr>
<tr>
<td>Occupation as physician or nurse</td>
<td>0.52</td>
<td>(0.24, 1.11)</td>
<td>0.092</td>
</tr>
<tr>
<td>Gender</td>
<td>0.78</td>
<td>(0.28, 2.17)</td>
<td>0.629</td>
</tr>
</tbody>
</table>

n=319 specimens, with 42 specimens positive for a respiratory virus

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