CONCISE COMMUNICATION

Effectiveness of Common Healthcare Disinfectants against H1N1 Influenza Virus on Reusable Elastomeric Respirators

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This study evaluated the efficacy of 3 common hospital disinfectants to inactivate influenza virus on elastomeric respirators. Quaternary ammonium/isopropyl alcohol and bleach detergent wipes eliminated live virus, whereas 70% isopropyl alcohol alone was ineffective.

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The US Centers for Disease Control and Prevention recommend N95 or other respiratory protection devices to shield healthcare personnel against airborne pathogens. Healthcare facilities typically maintain small stocks of N95 respirators, relying on just-in-time deliveries when usage increases. In recent years, manufacturers have been unable to meet demand during public health emergencies.[1,2]

Most N95 respirators are intended for a single use and are not to be reused except in severe shortages.[3,4] Reusable elastomeric respirators may substitute for N95 respirators, but it is not yet clear how or when they should be cleaned between uses. We aimed to determine whether widely available healthcare disinfectant wipes would effectively eliminate influenza virus from the surfaces of elastomeric respirators.

METHODS

Influenza virus (A/H1N1/California 2009) was grown in cell culture in Mink Lung shell vials (Quidel/Diagnostics Hybrids) using standard techniques. The approximate titer of virus was identified by serial dilution and rapid centrifugation culture, and the most suitable dilution was determined in preliminary experiments.

Filter cartridges and straps were removed from 40 elastomeric respirators (model 7500; 3M), and a 2 × 2-cm square test area was marked on the front portion of each respirator. After sterilization by standard methods, the marked section of 32 respirators was inoculated with 50 μL of influenza virus, and the respirators were allowed to air dry for 5–10 minutes. The remaining 8 respirators were inoculated with sterile medium (negative controls).

The influenza virus inoculated respirators were divided into 4 equal groups. Respirators in the first group (positive controls) were not disinfected. Respirators in the remaining 3 groups were treated with 1 of 3 different disinfectant wipes (8 per disinfectant). The first was 70% isopropyl alcohol (Webcol; Kendall), the second was 0.28% 2–2-pdiisobutylphenoxyethoxyethyldimethyl ammonium chloride (a quaternary ammonium chloride [QAC]) plus 17.2% isopropyl alcohol (Caviwipe; Metrex Research Corporation), and the third was a 1 : 10 bleach dilution plus detergent (Dispatch; Caltech Industries). The respirator group used as negative controls was also treated with the 3 wipes. After application of the wipe, the elastomeric material was allowed to air dry for 15 minutes. Investigators were blinded to the study groups.

Influenza virus was recovered with a swab premoistened with 50 μL of viral transport medium (Remel M4). The area was brushed 10 times with the moistened swab using a back and forth motion (1 wipe was moving the swab from one side of the square to the opposite side and back), rotating one-quarter turn and moving closer to the other end of the square on each successive wipe. The swab was then placed in 1 mL of viral transport media and vortexed for 30 seconds. A 50-μL aliquot was used to inoculate a centrifugation culture of mink lung cells, incubated for 18–24 hours, and processed for monoclonal direct fluorescent antibody staining of influenza A nucleoprotein (Simulfluor Flu A/Flu B; Millipore). Each infected cell or contiguous cluster of infected cells was counted as a single plaque-forming unit (pfu). Nucleic acids were extracted from the remaining sample and were tested for influenza using real-time polymerase chain reaction (PCR; GenProbe Proflu+; Prodesse). The PCR was considered positive if the threshold for detection was 35 cycles or less. Cultures and PCR assays were performed in duplicate for each specimen.

For culture of virus, the pfu count distributions were subjected to analysis by Kruskal-Wallis nonparametric 1-way analysis of variance followed by a subanalysis of the pairwise group differences using the Wilcoxon rank-sum test. The PCR data were analyzed using the χ^2 test for homogeneity followed by a pairwise comparison of the groups using the Fisher exact test (with the Bonferroni inequality adjustment).

RESULTS

The culture and PCR results are represented in Figures 1 and 2. All of the positive controls were positive by cell culture: 14–73 pfu of influenza, and PCR positive at 28–29 PCR cycles. All negative controls were negative by both culture and PCR. Influenza virus was not recovered by culture from any respirator treated with a quaternary ammonium chloride/isopropyl alcohol or bleach detergent–impregnated wipe, but 12.5% and 62.5%, respectively, were positive by PCR. Influenza virus was recovered by culture from 75% of respirators

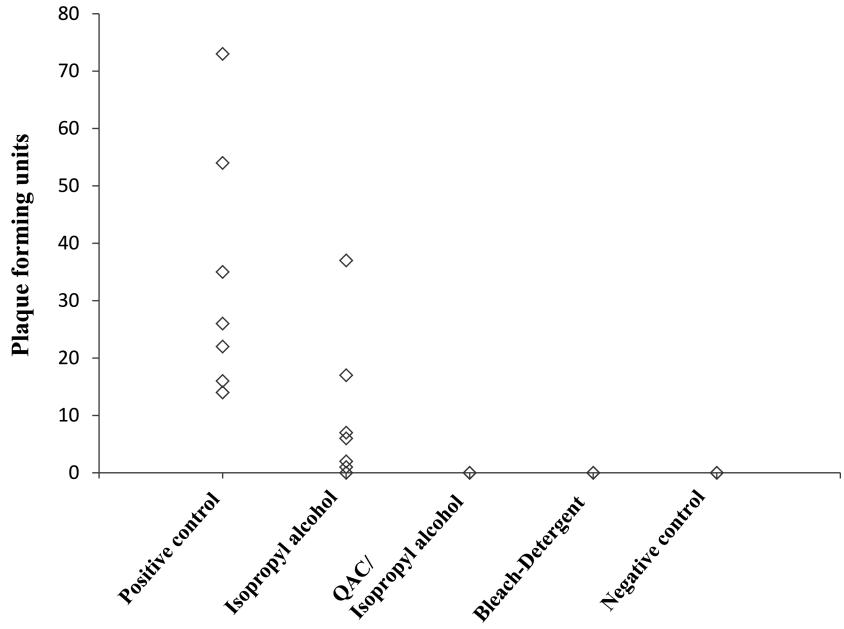

FIGURE 1. Shell vial culture results. Viable influenza virus was recovered from respirators treated with isopropyl alcohol but not from respirators treated with quaternary ammonium chloride (QAC)/isopropyl alcohol and bleach detergent wipes.

treated with isopropyl alcohol, and 83% were also positive by PCR.

There were significant differences between the groups ($P = .000002$, by Kruskal-Wallis test). The subanalysis using the Dunn test showed that the median number of pfu for the positive controls and isopropyl wipe were significantly higher than for the other 3 groups.

The second analysis, which compared the proportion of positive PCR results between the 5 groups, was also very significant ($P = .00004$). The pairwise comparison of the groups using Fisher exact test showed that positive controls had a significantly higher rate of positive results than did quaternary ammonium/isopropyl alcohol and the negative controls, and the rate of positive results associated with isopropyl alcohol wipe was significantly higher than that associated with the negative control. There were no other significant results.

DISCUSSION

These data demonstrate that, of the methods tested, quaternary ammonium/isopropyl alcohol wipes are the most efficacious against influenza virus on the surface of elastomeric respirators, followed by bleach detergent wipes. Application of 70% isopropyl alcohol led to some reduction in influenza virus but was not completely effective, because influenza virus remained viable by culture. Although 70% alcohol wipes are widely used, these data indicate that they may be inferior to other common wipes for disinfecting elastomeric respirators. The results of our study differ somewhat from those of previous studies regarding the activity of alcohol against influenza. This may be due to differences in growth surfaces (elastomeric material versus epithelial cells or other material) and/or differences in alcohol concentrations or the delivery medium used (wipes vs solutions and gels).[5,6]

Our PCR results were discordant with our culture results. Although PCR-based methods are typically more sensitive than culture for diagnoses of influenza disease, the presence of viral RNA is insufficient to determine whether infectious virus is present.[7] Persistent RNA may simply represent noninfectious viral nucleic acid. None of the disinfectants tested destroys nucleic acid as a primary mode of action.[8] In general, positive PCR results should not be considered to be a specific surrogate marker for detecting viable virus.

Elastomeric respirators are typically designed to be repeatedly disinfected according to the manufacturers' instructions. Previous N95 disinfection studies have been conducted with ultraviolet germicidal irradiation, ethylene oxide, and vaporized hydrogen peroxide.[9,10] However, these techniques require central processing and could pose an operational challenge during an emergency. Advantages of using disinfecting wipes include simplicity of use and wide availability in health care settings, such that healthcare workers may disinfect their own respirators.

We focused on decontaminating a small, flat portion of the respirator in a laboratory setting. Additional studies are necessary to determine how best to disinfect straps, nose clips, and other irregular surfaces between uses by healthcare workers. Our study was limited to an evaluation of the influenza A/California/07/2009 virus. Studies involving other respiratory pathogens would be informative. Finally, these experi-

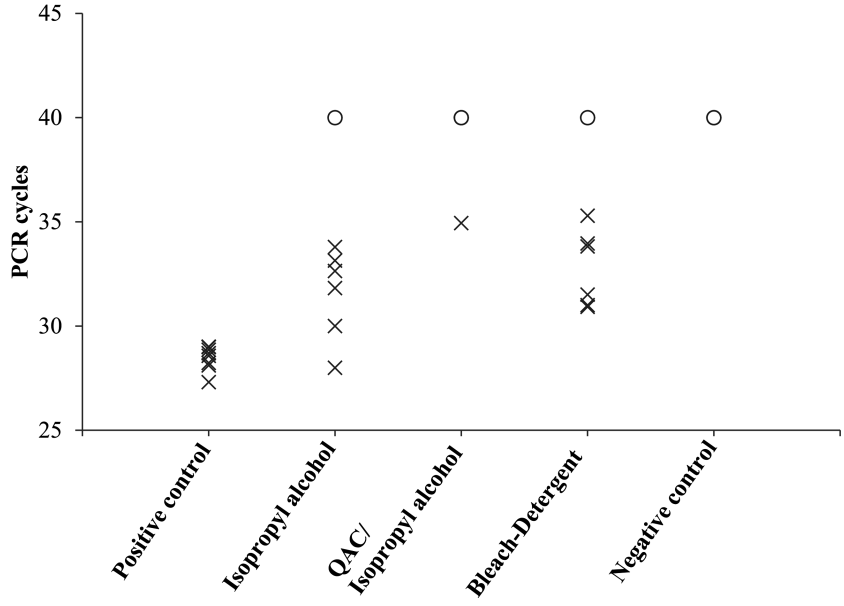

FIGURE 2. Polymerase chain reaction (PCR) results. Higher cycle numbers represent smaller amounts of viral RNA. PCR was run for 40 cycles. O, negative samples for which RNA was not detected; QAC, quaternary ammonium chloride; X, cycles at which samples exceeded the threshold for positivity.

ments were performed in a controlled setting. Results may differ because of variations in environmental conditions and cleaning procedures and may also differ with other disinfectants.

In conclusion, this study demonstrates that quaternary ammonium/isopropyl and bleach detergent disinfectant wipes are efficacious in disinfecting influenza H1N1 virus from reusable elastomeric respirator material. Additional research is needed to demonstrate their effectiveness and feasibility in healthcare settings.

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