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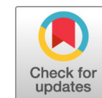
VIRUSES



Drifted Influenza A and B Viruses Collected by a Water-Based Condensation Growth Air Sampler in a Student Health Care Center during an Influenza Outbreak

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ABSTRACT A viable virus aerosol sampler (VIVAS) effectively collected viable influenza A and B viruses from air inside a student health care center during an influenza outbreak. The viruses had "drifted" genes, showcasing the usefulness of the VIVAS for air sampling and noninvasive surveillance of viruses in circulation.

The dynamics and significance of aerosol transmission of human influenza viruses remain controversial (1, 2). A major obstacle has been that virus aerosols are not easily collected by commonly used air samplers, and the viruses collected by such devices are often inactivated by the collection method (3). Infection risk analyses lack accuracy when they are based on virus detection alone, as breathing air always contains viruses that have been inactivated through exposure to ultraviolet light, drying, or other means, and thus pose no inhalation hazards. We have been evaluating and refining the viable virus aerosol sampler (VIVAS), a novel air sampler that operates on a water vapor condensation process to enlarge aerosolized virus particles to facilitate their capture (3, 4), and we tested it thrice a day on three different days during an influenza outbreak. Air samplings were performed in March and April 2016 at the Student Health Care Center (SHC) of the University of Florida; the study was exempt from institutional review board (IRB) approval and was approved by the SHC's director.

The quantity of viruses collected in air samplers is typically low and below the detection limit of PCR-based methods. To improve the chances of virus detection, virus isolation was attempted by inoculation of aliquots of the collection medium onto a variety of cell lines. Remarkably, mixed virus-induced cytopathic effects (CPE) were observed in various cell lines, indicating the isolation of different types of viruses on two of the test days (only human metapneumovirus was isolated on the third attempt).

Typical influenza virus (IFV) CPE, including the formation of focal enlarged granular cells, followed by nonspecific cell deterioration and detachment of the swollen cells from the growth surface, were observed in Madin-Darby canine kidney (MDCK) cell monolayer cultures. To quickly screen for the presence of influenza A and B virions, the spent medium of MDCK cell cultures that exhibited typical IFV-induced CPE was tested using a commercial solid-phase enzyme-linked immunosorbent assay (ELISA) (QuickVue influenza A and B kit; Quidel Corp., San Diego, CA, USA) (1, 5). Viral RNA was purified

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from the virus particles in the medium, and real-time PCR (RT-PCR) analyses were performed to establish virus type and subtype. Both pdmH1N1 and H3N2 IFV A viruses were recovered, as was Victoria lineage IFV B virus. Sanger sequencing of influenza A virus genomic segments 4 (hemagglutinin [HA] gene), 6 (neuraminidase [NA] gene), and 7 (matrix [M2 and M1] genes) and influenza B virus segments 4 (HA gene), 6 (NB glycoprotein [NB] and NA genes), and 7 (matrix protein 1 [M1] and BM2 genes) was accomplished according to previously described methods (1, 6, 7).

Sequencing revealed that the pdmH1N1 viruses belonged to hemagglutinin (HA) subclade 6B.1 (8–10), the H3N2 viruses belonged to HA clade subclade 3C.2a (7, 8), and the IFV B virus was a Victoria lineage clade 1A virus (11). Of interest, the viruses we isolated differ from those of the trivalent Northern Hemisphere influenza virus vaccines for 2015 to 2016, which contained a clade 1 pdmH1N1, A/California/7/2009 (H1N1); a clade 3C.3a H3N2, A/Switzerland/9715293/2013 (H3N2); and a clade 3 Yamagata-lineage IFV B (a B/Phuket/3073/2013) virus.

We conclude that the VIVAS could be useful for passive noninvasive virus surveillance applications.

Accession number(s). The sequences of this study have been deposited in the GenBank database under the following accession numbers: IFV pdmH1N1 HA gene, KX398060, KX398061, KX398062, and KX398063; IFV pdmH1N1 NA gene, KX398064, KX398065, KX398066, and KX398067; IFV pdmH1N1 M2 and M1 genes, KX398068, KX398069, KX398070, and KX398071; IFV H3N2 HA gene, KX398081, KX398082, and KX398083; IFV H3N2 NA gene, KX398084, KX398085, and KX398086; IFV H3N2 M2 and M1 genes, KX398087, KX398088, and KX398089; IFV B HA gene, KX398072, KX398073, and KX398074; IFV B NB and NA genes, KX398075, KX398076, and KX398077; and IFV B M1 and BM2 genes, KX398078, KX398079, and KX398080.

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