

Ionizing air affects influenza virus infectivity and prevents airborne-transmission

Marie Hagbom^{#1}, Johan Nordgren^{#1}, Rolf Nybom², Kjell-Olof Hedlund³, Hans Wigzell² & Lennart Svensson¹

¹Div of Molecular Virology, Department of Clinical and Experimental Medicine, University of Linköping, 581 85, Linköping; ²Department of Microbiology and Tumor Biology (MTC), Karolinska Institute, Stockholm, Sweden; ³Department of Diagnostics and Vaccine, Swedish Institute for Communicable Diseases Control, Stockholm, Sweden.

Contributed equally.

This study was published on 23 June 2015 in Scientific Reports by Nature Publishing Group.

BACKGROUND

- Each year, infectious diseases cause millions of deaths around the world and many of the most common infectious pathogens are spread by droplets or aerosols caused by cough, sneeze, vomiting etc.
- There is an urgent need for simple, portable and sensitive devices to collect, eliminate and identify viruses from air, to rapidly detect and prevent outbreaks and spread of infectious diseases.

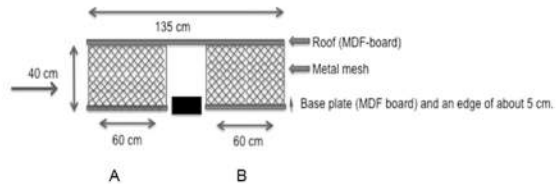
OBJECTIVE

- Investigate how effective ionization is in collecting, eliminating and identifying viruses from the air, using an ionizing device developed based on the IonFlow technology by Lightair.

METHODS

- The experimental room had grounded metal walls, with a volume of 19 m³ (B250°L330°H235cm). A particle counter (PortaCount Plus, TSI Incorporated, USA) was used before and during the experiment.
- The ionizing device used in this study was developed on the basis of the ion-flow ionizing technology from LightAir AB, Solna, Sweden (www.lightair.com). This device generates approximately 35 000 billion electrons per second (www.lightair.com) with a steady-state ozone concentration below the detection limit (0,002 ppm) as tested by VTT Technical Research Center of Finland, Tampere, Finland. It has also been ozone tested and certified by ARB (Air Resources Board) in the US.

Figure 3: Set-up design of influenza virus (H3N2, Pan/99) aerosol-transmission experiments between guinea pigs. Guinea pigs (n = 4) were intranasally infected with 5 × 10³ pfu of Pan/99 virus in 100 µL (50 µL in each nostril).



All four infected animals were placed into an experimental cage "A". At 30 h p.i. four naïve uninfected guinea pigs were placed in cage "B". Air-flow from left to right. Air exchanged 17x/day. Filled rectangle = ionizer.

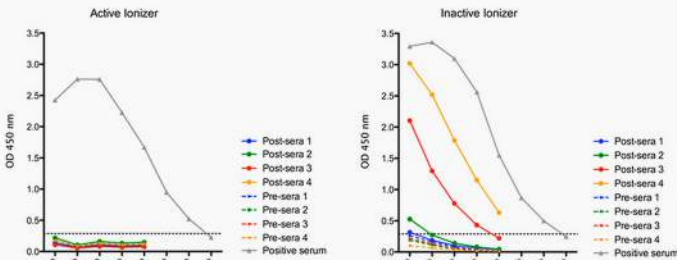
RESULTS

- 3 of 4 animals were infected when the inactive ionizer was used. In contrast, none of the 4 animals in cage "B" developed an immune response to influenza virus when the ionizer was active.
- Significantly higher numbers of rotavirus and CaCV particles were detected on the active ionizer compared to the inactive ionizer (~1500–3000 times), led to the conclusion that this technique can actively and efficiently collect viral particles from air.
- Testing revealed that 40–60 min was required to eliminate >90% of free latex particles in the air. The particle counter can detect particles with size greater than 0.02 µm.
- The infectivity of aerosolized viruses was significantly reduced by >97%, indicating that ionization of the aerosol accounts for the vast majority of infectivity reduction, and not the exposure to the charged collector plate.

Table 1: Collection efficiency of aerosolized CaCV, rhesus rotavirus (RRV) and Influenza A virus in various concentrations as determined by RT-qPCR.

Assess virus aerosolized (genus/PCR reaction)	Assess virus on collector (genus/PCR reaction) ±SE	Assess virus on collector OFF (genus/PCR reaction)	Recovery (%) ON	Recovery (%) OFF	Ratio (ON/OFF)
CaCV					
1.88 × 10 ³	1.18 × 10 ³ ± 0.4 × 10 ³	73	0.03%	0.000079%	1620
1.99 × 10 ³	7.36 × 10 ³ ± 2.36 × 10 ³	~5	0.37%	~0.00024%	~1520
9.93 × 10 ³	1.66 × 10 ³ ± 6.5 × 10 ²	not detected	0.17%	NA	NA
3.20 × 10 ³	8.1 × 10 ³	not detected	0.37%	NA	NA
1.56 × 10 ³	1.85 × 10 ³ ± 9.07 × 10 ²	not detected	10.60%	NA	NA
1.87 × 10 ³	3.88 × 10 ³ ± 1.27 × 10 ³	not detected	21%	NA	NA
RRV					
2.23 × 10 ³	7.54 × 10 ³ ± 6.7 × 10 ³	~2-3	0.34%	~0.00011%	~3800
4.85 × 10 ³	6.48 × 10 ³ ± 86	Not detected	0.13%	NA	NA
9.13 × 10 ³	41 ± 31	Not detected	0.05%	NA	NA
Influenza virus					
4.30 × 10 ³	3.33 × 10 ³ ± 7.22 × 10 ²	Not detected	0.08%	NA	NA

Figure 4: Active ionizer prevents aerosol transmitted influenza virus (H3N2, Pan/99) infection between guinea pigs.



While the active ionizer prevented 4 of 4 exposed guinea pigs from developing an immune response to influenza virus, 3 of 4 animals were infected when the inactive ionizer was used. Graph shows antibody titers by ELISA before infection (pre-serum 1, 2, 3 and 4) and at day 21 post-exposure to influenza virus (post-serum 1, 2, 3 and 4).

Table 2: Reduction of infectivity of Canine Calicivirus (CaCV) and Rhesus Rotavirus (RRV).

Ratio of infectious virus particles to virus genes per PCR-reaction as quantified by RT-qPCR					
Exposed to charged collector	Exposed to uncharged collector	Reduction of infectivity	Aerosolized virus	Aerosolized virus captured	Reduction of infectivity
CaCV 0.74 × 10 ⁻⁴	1.24 × 10 ⁻⁴	40.1%	2.96 × 10 ⁻²	<7.83 × 10 ⁻⁴⁶	>97.4% ^a
RRV n.d.	n.d.	n.d.	4.86 × 10 ⁻¹	<7.66 × 10 ⁻³⁸	>98.4% ^a

^aUnder detection limit (10 peroxidase forming units/mL) on the infectivity assay.

DISCUSSION

- Important advantages with this novel ionizing device is the simple handling, high robustness as well as the wide applicability to airborne pathogens.
- The observation that significantly higher numbers of rotavirus and CaCV particles were detected on the active ionizer compared to the inactive ionizer (~1500–3000 times), led to the conclusion that this technique can actively and efficiently collect viral particles from air.
- It is interesting to note that a broad range of particles sizes, from 35 nm to 10 µm was concentrated, suggesting a wide application range of the technology.
- Most interesting, and of great clinical significance of this study was the novel finding that the ionizing device could detect and prevent influenza virus infection in a controlled setting, mimicking "authentic" conditions.
- The easy handling, low cost, free of ozone production, robustness, high efficiency and low-voltage (12 volt) operation enables large-scale use. Locations critical for infectious spread, such as airplanes, hospitals, day-care centres, school environments and other public places could thus be monitored and controlled by the collection and analysis of airborne viruses and other pathogens on the collector plate.
- We conclude that this innovative technology hold great potential to collect and identify viruses in environmental air.

CONCLUSIONS BY LIGHTAIR

- Lightair is highly efficient in inactivating viruses in the air, and capturing viruses even when there are only small amounts of viruses in the air.
- Lightair's IonFlow technology, the ionizing of air and the electrostatic attraction, offers a new solution to prevent aerosol-transmitted influenza infections by preventing the spread of airborne transmitted viruses. The viruses used in the study are of great clinical and economic importance since they represent and was used as surrogates for viruses that cause among other diseases, the "winter vomiting disease", diarrhea and gastroenteritis.
- They study shows that the IonFlow technology can be of use against viruses and other pathogens in the air and with wide environmental and clinical applications suitable for distribution in airplanes, hospitals, day-care centers, school environments and other public places as well as homes.
- Lightair's IonFlow technology is highly efficient in capturing small particles.

DISCLAIMER: This summary has been prepared by Lightair AB and without the cooperation of the Nature Publishing Group.

The full study can be viewed at www.nature.com