

Efficacy of face masks to prevent respiratory virus transmission: abridged secondary publication

BJ Cowling *, DK Ip, HL Yen

KEY MESSAGES

1. Various respiratory viruses can be detected in exhaled breath of patients with acute respiratory infections.
2. Viral loads are greatest in those with influenza virus infection.
3. We did not identify a significant effect of surgical face masks in reducing the amount of respiratory virus detected in coarse or fine particles in

exhaled breath.

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BJ Cowling, DK Ip, HL Yen

School of Public Health, The University of Hong Kong, Hong Kong

* Principal applicant and corresponding author: bcowling@hku.hk

Introduction

Respiratory viruses cause infections and hence economic losses through sick leave and doctor consultations as well as hospitalisations and deaths. The most severe acute upper respiratory tract infections are usually due to respiratory syncytial virus (RSV) in infants and influenza in all ages. The burden of other common respiratory viruses (parainfluenza, adenovirus, metapneumovirus, coronavirus, and rhinovirus) is also considerable. These viruses often result in a broad and overlapping spectrum of symptoms collectively referred to as common cold.

Modes of respiratory virus transmission include contact, large droplets, and aerosols. Although hand hygiene and use of face masks, primarily targeting contact and large droplet transmission, have been suggested as mitigation strategies against influenza virus, little is known about the relative importance of these modes to transmission for other common respiratory viruses apart from influenza. It has been suggested that contact transmission predominates for RSV, whereas both contact and aerosol transmissions are possible for rhinovirus.

There are a few studies reporting recovery of non-influenza respiratory viruses from human exhaled breath or aerosol samples from clinics. During the SARS epidemic in 2003, most Hong Kong people wore face masks. Although one study suggested that public health interventions during the SARS epidemic were effective in preventing other respiratory virus transmissions, little is known about the efficacy of face masks in filtering respiratory virus from an individual with respiratory infections. Most of the existing evidence on the filtering efficacy of face masks and respirators come from in vitro experiments that mainly use non-biological

particles that may not be generalisable to infectious respiratory virus droplets. There are few in vivo studies investigating the efficacy of face masks and quantifying viral titres and virus generation rates in human exhaled breath aerosols.

This study aimed to examine exhaled breath virus generation rate of different respiratory viruses (with implications for modes of transmission) and to determine the potential benefits of face masks to prevent respiratory virus transmission.

Methods

This study was approved by the Institutional Review Board of the University of Hong Kong. Written informed consent was obtained from all patients and from parents or legal guardians of patients aged 11 to 17 years.

From April 2014 to March 2016, we recruited local residents aged ≥ 11 years who reported at least two signs or symptoms of acute respiratory illness within 72 hours of illness onset from a local outpatient clinic in Hong Kong outside influenza seasons. A set of nose and throat swab sample was collected. Subjects were invited to do a short questionnaire to record demographic information including age, sex, clinical illness symptoms, medication used, and medical, allergy, and smoking history.

Subjects were asked to tidally breathe to the G-II bioaerosol collective device for at least 30 minutes while wearing a surgical face mask (Kimberley Clark). The G-II would collect their exhaled breath aerosol particles into two different size fractions, either with aerodynamic diameter $\geq 5 \mu\text{m}$ or $< 5 \mu\text{m}$, by the sample impactor and collection fluid (viral transport reservoir buffer), respectively. Then, the sample impactor and collection fluid from the G-II were collected and

changed. Finally, the subjects were asked to tidally breathe again to the G-II bioaerosol collective device for at least 30 minutes, without wearing a face mask.

The nasal and throat swabs were placed in viral transport media refrigerated at 2°C to 8°C immediately after collection, stored at -20°C for up to 7 days, and then stored at -80°C until testing qualitatively for the presence of respiratory viruses by the xTAG Respiratory Viral Panel, and subsequently quantitatively by RT-PCR. The exhaled droplets from subjects captured by bioaerosol collector G-II in $\geq 5\text{-}\mu\text{m}$ fraction collected on the impactor plate were stored in a 50 mL tube, and exhaled droplets in $< 5\text{-}\mu\text{m}$ fractions collected in 150 mL viral transport reservoir buffer, were refrigerated at 2°C to 8°C immediately after collection and stored at 4°C during transport. Once in the laboratory, the impactor plate was swabbed and the virus transferred to 2 mL new viral transport buffer, and the 150 mL viral transport reservoir buffer was concentrated to 2 mL using Centricon Plus-70 (Millipore, USA) the same day. After addition of antibiotics, both samples were stored at -80°C until processing with qPCR or viral culture for determination of respiratory virus concentration and infectivity.

Nose and throat swab samples were subjected to in vitro diagnostic-use viral panel, xTAG Respiratory Viral Panel (Abbott Molecular, Illinois, USA), to detect qualitatively 12 common respiratory viruses and subtypes including influenza A (non-specific, H1, and H3) and B, respiratory syncytial virus (RSV, subtypes A and B), parainfluenza (types 1-3), adenovirus, metapneumovirus, coronavirus, and rhinovirus. The xTAG Respiratory Viral Panel has been shown to detect common respiratory viral targets with high sensitivity (100%) and specificity (91%) than other similar panels in over 200 respiratory specimens from adult patients with signs of respiratory infection. After one or more of the candidate respiratory viruses was detected in the nose and throat swab by the Viral Panel, the nose and throat swab, and the exhaled droplets from subjects captured on the impactor plate or the reservoir buffer ($\geq 5\text{ }\mu\text{m}$ or $< 5\text{ }\mu\text{m}$) would be processed with qPCR (and viral culture when a susceptible cell line was available) specific to the candidate virus for determination of virus concentration in different size fractions of aerosols.

The primary outcome of the study was the virus generation rate in the tidal breathing of patients infected by different respiratory viruses, and the efficacy of face mask in preventing virus dissemination. The secondary outcome was the correlation between viral loads in nose/throat swabs and exhaled breath particles. We stratified all analyses by type of respiratory virus infection as determined in the viral panel on the nose swab. We tabulated the respiratory virus positive proportion as determined

by PCR in nasal swabs, throat swabs, exhaled breath coarse particles, and exhaled breath fine particles for the corresponding respiratory virus as identified by the viral panel, among participants who have provided an exhaled breath sample without wearing a face mask stratified by two age groups. For the three groups of respiratory viruses with highest frequency of infection, we plotted log viral load of nasal swab, throat swab, fine/coarse fraction of exhaled breath against age, days since acute respiratory illness onset and mask intervention, number of coughs/sneezes. We investigated the correlations between viral shedding in nose swab, throat swab, fine and coarse fractions of exhaled breath by scatterplot between any two types of samples. We then compared the number of exhaled breath samples containing detectable viral load between patients wearing face mask or not, and tested for significant difference whenever possible by Fisher's exact tests.

Results

From April 2014 to March 2016, of 1746 patients screened, 703 (40%) were eligible and 219 (31%) were recruited. Seven (3%) patients withdrew and the remaining 212 (97%) patients were proceeded to the randomisation of mask intervention for the first exhaled breath collection, and voluntarily provided a second exhaled breath sample of alternate mask intervention. A total of 105 patients were randomised to not wearing a face mask during the first exhaled breath collection and 107 patients randomised to wearing a face mask. To increase the power of the analysis for individual respiratory viruses, we decided to include in the analysis exhaled breath samples collected from 44 patients recruited from a similar study conducted earlier from January 2013 to March 2014. Therefore, in final analysis, 256 patients were included, contributing to 151 exhaled breath samples collected without wearing a face mask and 152 exhaled breath samples collected with wearing a face mask.

Most of the 256 patients were between 18 and 50 years old (77%) and were recruited within 2 days since symptom onset (79%). Infection by at least one of the respiratory viruses included in the viral panel was identified in 153 (60%) of all patients. Infection by enterovirus/rhinovirus was most frequent ($n=65$, 25%), followed by influenza viruses ($n=48$, 19%) and human coronaviruses ($n=24$, 9%).

Four (80%) of five patients aged 11 to 17 years tested positive by the viral panel, including enterovirus/rhinovirus ($n=1$), influenza A virus ($n=2$), and influenza B virus ($n=1$). The respiratory virus infection was confirmed by RT-PCR in nose swabs in the two patients infected with influenza A virus but not in patients infected by other respiratory viruses. For all four patients, none had respiratory viral RNA recovered in their throat swabs or from the coarse or

TABLE. Efficacy of surgical face masks in reducing respiratory virus dissemination

Virus type	No of PCR+ exhaled breath samples					
	Fine particles ≤5 μm			Coarse particles >5 μm		
	Mask	Control	P value	Mask	Control	P value
Enterovirus/rhinovirus	0/25	0/33	-	0/25	0/33	-
Influenza A virus	3/15	6/17	0.44	0/15	4/17	0.10
Influenza B virus	1/9	1/4	1.00	0/9	1/4	0.31
Human coronavirus (NL63, OC43, HKU1)	0/16	1/12	0.43	0/16	0/12	-
Parainfluenza virus (P1, P2, P3, P4)	0/6	0/5	-	0/6	0/5	-
Respiratory syncytial virus	0/2	1/4	1.00	0/2	0/4	-
Human metapneumovirus	1/1	1/2	1.00	0/1	0/2	-

fine particles of the exhaled breath. In addition, 79 (66%) of 119 patients aged ≥18 years tested positive by the viral panel, including enterovirus/rhinovirus (n=33, 27%), influenza A virus (n=16, 13%), influenza B virus (n=3, 3%), and human coronaviruses (n=12, 10%). The respiratory virus infection was confirmed by RT-PCR in nose swabs in patients infected with enterovirus/rhinovirus (30%), influenza A (94%) and B (33%) viruses, and human coronaviruses (17%). Together with other respiratory viruses, we detected among the 153 patients respiratory viral RNA by RT-PCR in 20 (10%) of 152 exhaled breath samples, in the fine fraction in control group (10/77, 13%) and in mask group (5/75, 7%), and in the coarse fraction in control group (5/77, 6%) and in mask group (0/75, 0%).

We selected influenza A, enterovirus/rhinovirus, and human coronavirus for further analysis given the relatively larger sample sizes. We could not detect a significant difference on the effect of surgical masks on viral shedding of influenza A virus in the coarse (P=0.10) and fine (P=0.44) fraction of exhaled breath; nor that of human coronavirus in the fine fraction (P=0.43) [Table].

Discussion

Surgical face masks are inexpensive and easily accessible and are therefore widely used during epidemics of respiratory infections, both as a source control measure in ill persons and as a preventative measure against infection.

Viral RNA was observed in nose/throat swab and in exhaled breath regardless of the number of cough/sneezes produced during collection. We detected influenza A virus and human coronaviruses viral shedding in the fine fraction of exhaled breath from patients with as little as two coughs, and in the coarse fraction in one influenza-infected patient who never coughed, demonstrating that patient could shed virus through exhaled breath to the environment with limited or even without coughing

in both fine and coarse fractions of exhaled breath.

Human rhinovirus, RSV, and adenovirus have been detected in three exhaled breath samples from adults with mild to severe asthma.¹ Among asthmatic children with human rhinovirus infection, 11.5% of virus was detected in exhaled breath samples (compared to 25.5% in nasal wash samples) and one sample had co-infection with non-human rhinovirus.² Viral RNA has been detected from human rhinovirus (45%) and parainfluenza virus (26%) from exhaled breath. Among the patients (29/53) with positive human rhinovirus viral RNA in exhaled breath, they also observed 1 to 3 cases of concurrent detection of parainfluenza virus type 1 and 3, and influenza A virus.

The major limitation of our study was the large proportion of influenza-confirmed patients with undetectable viral shedding in exhaled breath samples. We could have increased the sampling duration beyond 30 minutes to increase the viral load being captured, at the cost of acceptability in some participants. An alternative approach is to invite patients to perform forced coughs during exhaled breath collection.³ However, many of our patients did not cough much or at all, and in our present study we focused on recovering respiratory virus in exhaled breath in a real-life situation. We found that surgical masks were effective in preventing virus dissemination in coarse fraction of exhaled breath even when a participant coughed many times.

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Disclosure

The results of this research have been previously published in:

(1) Leung NHL, Chu DKW, Shiu EYC, et al. Respiratory virus shedding in exhaled breath and efficacy of face masks. *Nat Med* 2020;26:676-80.

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