


Concise Communication

Transfer efficiency of an enveloped virus, human coronavirus 229E, from various hard surface fomites to finger pads of the hands

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Abstract

Respiratory viruses can be transmitted by fomite contact, but no data currently exist on the transfer of enveloped viruses. The transfer efficiency of human coronavirus from various hard surfaces ranged from 0.46% to 49.0%. This information can be used to model the fomite transmission of enveloped viruses.

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The transfer efficiency of viruses from various types of fomites (ie, inanimate objects and surfaces) to the finger pads of hands is key in the development of pathogen exposure and quantitative microbial risk assessment models.^{1,2} Previous studies have documented the transfer efficiencies of nonenveloped viruses from various types of fomites to finger pads.^{3,4} In this study, we evaluated the transfer efficiency for enveloped viruses such as coronavirus (HCoV surrogate 229E).

Material and methods

Preparation of the test virus

Human coronavirus 229E (HCoV 229E) was procured from the American Type Culture Collection (ATCC VR-740; Manassas, VA) and propagated using the MRC-5 cell line (ATCC CCL-171). Infected cells were freeze-thawed and clarified lysates underwent a polyethylene glycol (PEG) extraction [12% (w/v) PEG (8000 mw) and 0.5 M sodium chloride] overnight at 4°C. The suspension was centrifuged (10,000 ×g for 60 minutes), and the virus pellet was resuspended in 0.01 M phosphate-buffered saline (PBS; pH 7.4) to 10% of the original suspension volume. Aliquots were stored at –80°C until use on the respective study dates. Virus stock titers were determined prior to the study by thawing and diluting (1:10) a stock vial using 0% fetal bovine sera (FBS) in minimal essential media (MEM). Dilutions were plated in replicates of 6 onto MRC-5 monolayers prepared in multiwell trays and incubated for 7 days (35°C) in a 5% CO₂ atmosphere. Wells were scored for cytopathogenic effects and recorded as tissue culture infectious dose at the 50% end point (TCID₅₀) per mL.⁵

Pre- and post-experiment hand decontamination

The study participant was healthy 75-year-old male whose hands lacked visible cracks, abrasions, cuts, or other compromising skin conditions. Permission was obtained and the protocol was approved by University of Arizona Institutional Research Board prior to conduct of the study; a human subjects review was not required. Prior to all experiments, the study participant's hands were washed using an antibacterial liquid hand soap for 45 seconds, rinsed with deionized water, and dried using paper towels. Each hand was then sprayed twice with 70% ethanol and rubbed over the hands and wrists for 15 seconds followed by air drying for a minimum of 5 minutes. After conducting fomite-to-finger viral transfer experiments, the study participant's finger pads were sprayed twice using 70% ethanol, and the entirety of each hand was wrapped within 70% ethanol-saturated paper towels for 30 seconds. The hands were then washed, rinsed, and dried as previously described.

Test carrier preparation and inoculation with HCoV 229E

On the test dates, a vial of stock virus (~10⁷ TCID₅₀/mL) was thawed and amended with FBS to achieve an organic load of 5% (v/v). Cleaned, sanitized (70% ethyl alcohol) test carriers (Table 1) were inoculated with 10 µL of virus, which was spread over an area of 1-cm² using a bent pipette tip. The carriers were dried (no liquid) in a controlled humidity chamber (22 ± 1°C and 40% ± 5% relative humidity) for ~30 minutes with the petri dish lids on. A triplicate set of carriers was immediately harvested to determine levels of infectious virus per carrier just prior to the transfer experiments. Carriers were rinsed with 1 mL of 0% FBS MEM with antibiotics 3–5 times and treated with a sterile cell scraper to further facilitate virus detachment. Dilutions and plating onto MRC-5 cell monolayers followed as previously described.

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Table 1. Fomites/Surface Types Tested

Fomite/Surface Type	Description	Manufacturer or Source
Stainless steel	Gauge 304	AK Steel Corporation
Glass	Slides	VWR, Mississauga, Ontario,
Glazed porcelain	Porcelain	Home Depot, Atlanta, GA
Laminate	Vinyl floor tile	Home Depot, Atlanta, GA
Formica	Countertop tiles	Home Depot, Atlanta, GA

Fomite-to-finger transfer experiments

The remaining carriers underwent transfers from fomite to finger pad within 15 minutes after drying. The study was conducted under controlled conditions ($22 \pm 1^\circ\text{C}$ temperature; $55\% \pm 5\%$ relative humidity). One transfer trial consisted of 6 transfer events using the index, middle, and ring fingers of both hands for each surface type. The transfer protocol was conducted according to Lopez *et al*⁴ by placing the finger pad directly onto the contaminated carrier, achieving full contact with the 1-cm² inoculum zone for 10 seconds at 1.0 kg/cm² of average pressure (range, 900 g/cm² to 1,200 g/cm²).

Finger sampling

HCoV 229E was initially recovered from contaminated fingers using the nylon swab method described by Rusin *et al*⁶; however, low viral recoveries were achieved. An alternative method was then employed during which the contaminated fingers were washed in 1 mL 0% FBS MEM with antibiotics within a sterile petri dish. The individual fingers were placed into the liquid at the bottom of the petri dish and rotated touching the bottom of the petri dish for 10 seconds to facilitate viral removal, and the suspensions were diluted (1:10) using 0% FBS MEM with antibiotics. Dilutions and plating onto confluent MRC-5 host cell monolayers followed as previously described.

Results

The swab method for sampling HCoV 229E transferred to the finger pads yielded lower numbers than the fingers directly rotating into 1 mL 0% MEM with antibiotics (Table 2). Overall, the highest numbers of HCoV 229E were transferred from glazed porcelain ($49.07\% \pm 16.70\%$) to the fingers compared to stainless steel (<1%), which demonstrated the lowest mean transfer efficiency of HCoV 229E (Table 2).

Discussion

Fomite-to-finger pad transfer data has been published for nonenveloped viruses; however, it cannot be extrapolated to enveloped viruses due to differences in viral structure and methodologies. The transfer efficiencies presented herein for HCoV 229E are similar to those of nonenveloped viruses,^{3,4} although they may be greater for glazed porcelain. The reason for greater fomite-to-finger transfer from glass and glazed porcelain is unknown but may be attributable to the microscopically smoother surface of these fomites compared to others (eg, stainless steel) or the hydrophobicity of the surfaces. Properties including fomite/surface and viral hydrophobicity, and electrostatic interactions also contribute to the stability of virus-surface interfacial boundaries and should be considered factors. Lopez *et al*⁴ demonstrated a 7.1% transfer of

Table 2. Fomite-to-Finger Pad Transfer Efficiencies of Human Coronavirus 229E

Fomite/Surface Type	Mean Pre-Transfer Surface Viral Titer Log ₁₀ ± SD (n=6)	Mean Transfer to Finger Titer Log ₁₀ ± SD (n=6)	Mean Transfer Efficiency % ± SD
Stainless steel ^a	4.78 ± 0.19	0.92 ± 0.20	0.013 ± 0.003
Stainless steel ^b	4.94 ± 0.25	2.65 ± 2.70	0.46 ± 0.57
Glass ^b	4.17 ± 0.17	3.76 ± 4.06	37.24 ± 82.34
Glazed porcelain ^b	4.72 ± 0.10	4.42 ± 3.91	49.07 ± 16.70
Laminate ^b	4.28 ± 0.10	3.24 ± 2.98	6.55 ± 5.48
Formica ^b	4.44 ± 0.10	3.85 ± 3.86	25.38 ± 28.4

Note. SD, standard deviation.

^aSwab method.

^bWash method.

^cAverage represent the results of 6 replicates.

MS-2 from ceramic tiles, and both MS-2 and PRD-1 phages were transferred from glass at efficiencies of 19.3% and 33.47%, respectively. Ansari *et al*³ reported a mean transfer efficiency of rotavirus from stainless steel of 16.8%.

Respiratory viruses such as influenza, rhinoviruses, and respiratory syncytial virus, and coronavirus can be transmitted, in part, by inoculation of the nose, mouth, or eyes via contaminated hands. Thus, fomites may play a role in the transmission of these viruses, including coronavirus. For this reason, infection prevention measures recommended to preclude the transmission from an environmental surface or object by hands to the nose, mouth, or eyes include disinfection of surfaces and hand hygiene.^{3,4} In the case of rhinoviruses and respiratory syncytial virus, inoculation of the nose, mouth, or eyes may be the major route of transmission.⁷ Intranasal inoculation has been demonstrated as a route of transmission of human coronavirus 229E and SARS-CoV-2.^{8,9} With regard to fomites, modeling has shown that spread of the influenza virus depends on touching frequency and other factors in indoor environments.¹⁰ In contrast, the role of fomites in the transmission of coronavirus SARS-CoV-2 is not currently known. However, this study has demonstrated the potential for coronaviruses (eg, SARS-CoV-2) to be transferred from various fomites to the fingers.

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Conflicts of interest. Dr Rutala is a consultant to Professional Disposables International (PDI). All other authors report no conflicts of interest relevant to this article.

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