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Aerosol and surface stability of HCoV-19 (SARS-CoV-2) compared to SARS-CoV-1 Neeltje van Doremalen^{1*}, Trenton Bushmaker^{1*}, Dylan H. Morris^{2*}, Myndi G. Holbrook¹, Amandine Gamble³, Brandi N. Williamson¹, Azaibi Tamin⁴, Jennifer L. Harcourt⁴, Natalie J. Thornburg⁴, Susan I. Gerber⁴, James O. Lloyd-Smith^{3,5}, Emmie de Wit¹, Vincent J. Munster¹ 1. Laboratory of Virology, Division of Intramural Research, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Hamilton, MT, USA 2. Department of Ecology and Evolutionary Biology, Princeton University, Princeton, NJ, USA 3. Department of Ecology and Evolutionary Biology, University of California, Los Angeles, Los Angeles, CA, USA 4. Division of Viral Diseases, National Center for Immunization and Respiratory Diseases, Centers for Disease Control and Prevention, Atlanta, GA, USA. 5. Fogarty International Center, National Institutes of Health, Bethesda, MD, USA * These authors contributed equally to this article

Abstract

multiple hours and on surfaces up to days.

HCoV-19 (SARS-2) has caused >88,000 reported illnesses with a current case-fatality ratio of ~2%. Here, we investigate the stability of viable HCoV-19 on surfaces and in aerosols in comparison with SARS-CoV-1. Overall, stability is very similar between HCoV-19 and SARS-CoV-1. We found that viable virus could be detected in aerosols up to 3 hours post aerosolization, up to 4 hours on copper, up to 24 hours on cardboard and up to 2-3 days on plastic and stainless steel. HCoV-19 and SARS-CoV-1 exhibited similar half-lives in aerosols, with median estimates around 2.7 hours. Both viruses show relatively long viability on stainless steel and polypropylene compared to copper or cardboard: the median half-life estimate for HCoV-19 is around 13 hours on steel and around 16 hours on polypropylene. Our results indicate that aerosol and fomite transmission of HCoV-19 is plausible, as the virus can remain viable in aerosols for

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A novel human coronavirus, now named severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2, referred to as HCoV-19 throughout this manuscript) emerged in Wuhan, China in late 2019. As of March 3, 2020, >88,000 cases have been diagnosed in 64 countries, including 2915 deaths. The rapid expansion of this outbreak is indicative of efficient human-to-human transmission.^{2,3} HCoV-19 has been detected in upper and lower respiratory tract samples from patients, with high viral loads in upper respiratory tract samples. 4,5 Therefore, virus transmission via respiratory secretions in the form of droplets (>5 microns) or aerosols (<5 microns) appears to be likely. Virus stability in air and on surfaces may directly affect virus transmission, as virus particles need to remain viable long enough after being expelled from the host to be taken up by a novel host. Airborne transmission or fomite transmission were thought to play important roles in the epidemiology of the two zoonotic coronaviruses that emerged this century, SARS-CoV-1 and MERS-CoV. Airborne transmission may have been responsible for the largest superspreading event during the SARS epidemic of 2002-2003, and numerous nosocomial superspreading events of SARS-CoV-1 were linked to aerosol-generating medical procedures. 8-10 Fomite transmission was also suspected during the SARS epidemic, and one analysis of a nosocomial SARS-CoV-1 superspreading event concluded that fomites had played a significant role.¹¹ Given the potential impact of different routes of transmission on the epidemiology of emerging viruses, it is crucial to quantify the virological traits that may shape these aspects of HCoV-19 transmission. Here, we analyze the aerosol and surface stability of HCoV-19 and compare it with SARS-CoV-1, the most closely related coronavirus known to infect humans. ¹² We evaluated the aerosol stability of HCoV-19 and SARS-CoV-1 for up to three hours in aerosols and up to 7 days on different surfaces. We estimated decay rates of HCoV-19 and SARS-CoV-1 in each condition using a Bayesian regression model. Methods HCoV-19 nCoV-WA1-2020 (MN985325.1)¹³ and SARS-CoV-1 Tor2 (AY274119.3)¹⁴ were the strains used in our comparison. Virus stability in aerosols was determined as described previously at 65%

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relative humidity (RH) and 21-23°C. 15 In short, aerosols (<5 µm) containing HCoV-19 (10^{5.25} TCID₅₀/mL) or SARS-CoV-1 (10^{6.75-7} TCID₅₀/mL) were generated using a 3-jet Collison nebulizer and fed into a Goldberg drum to create an aerosolized environment. Aerosols were maintained in the Goldberg drum and samples were collected at 0, 30, 60, 120 and 180 minutes post-aerosolization on a 47mm gelatin filter (Sartorius). Filters were dissolved in 10 mL of DMEM containing 10% FBS. Three replicate experiments were performed. Surface stability was evaluated on plastic (polypropylene, ePlastics), AISI 304 alloy stainless steel (Metal Remnants), copper (99.9%) (Metal Remnants) and cardboard (local supplier) representing a variety of household and hospital situations and was performed as described previously at 40% RH and 21-23°C using an inoculum of 10⁵ TCID₅₀/mL. ¹⁶ This inoculum resulted in cycle-threshold values (Ct) between 20 and 22 similar to those observed in samples from human upper and lower respiratory tract. In short, 50 µl of virus was deposited on the surface and recovered at predefined time-points by adding 1 mL of DMEM. Stability on cardboard was evaluated by depositing 50 µl of virus on the surface and recovering the inoculum by swabbing of the surface, the swab was deposited 1 mL of DMEM. Three replicate experiments were performed for each surface. Viable virus in all surface and aerosol samples was quantified by end-point titration on Vero E6 cells as described previously. 16 The Limit of Detection (LOD) for the assays was 10^{0.5} TCID₅₀/mL for plastic, steel and cardboard and 10^{1.5} TCID₅₀/mL for copper (due to toxicity caused by the copper in the undiluted samples). The durations of detectability depend on initial inoculum and sampling method, as expected. To evaluate the inherent stability of the viruses, we estimated the decay rates of viable virus titers using a Bayesian regression model. This modeling approach allowed us to account for differences in initial inoculum levels across replicates, as well as interval-censoring of titer data and other sources of experimental noise. The model yields estimates of posterior distributions of viral decay rates and halflives in the various experimental conditions – that is, estimates of the range of plausible values for these parameters given our data, with an estimate of the overall uncertainty.¹⁷ We describe our modeling approach in more detail in the Supplemental Materials.

95 **Results** 96 97 HCoV-19 remained viable in aerosols throughout the duration of our experiment (180 minutes) with a reduction in infectious titer 3 hours post-aerosolization from $10^{3.5}$ to $10^{2.7}$ CID₅₀/L (mean across 98 99 three replicates). This reduction in viable virus titer is relatively similar to the reduction observed in aerosols containing SARS-CoV-1, from 10^{4.3} to 10^{3.5} TCID₅₀/mL (mean across three replicates) (Figure 100 101 1A). 102 HCoV-19 was most stable on plastic and stainless steel and viable virus could be detected up to 103 72 hours post application (Figure 1B), though by then the virus titer was greatly reduced (polypropylene from 10^{3.7} to 10^{0.6} TCID₅₀/mL after 72 hours, stainless steel from 10^{3.7} to 10^{0.6} TCID₅₀/mL after 48 hours, 104 105 mean across three replicates). SARS-CoV-1 had similar stability kinetics and live virus could be detected 106 on these surfaces up to 72 hours on polypropylene and 48 hours on stainless steel (polypropylene from 10^{3.4} to 10^{0.7} TCID₅₀/mL after 72 hours, stainless steel from 10^{3.6} to 10^{0.6} TCID₅₀/mL after 48 hours, mean 107 108 across three replicates). No viable virus could be measured after 4 hours on copper for HCoV-19 and 8 109 hours for SARS-CoV-1, or after 24 hours on cardboard for HCoV-19 and 8 hours for SARS-CoV-1 110 (Figure 1B). 111 Both viruses exhibited exponential decay in viable virus titer across all experimental conditions, 112 as indicated by linear decrease in the log₁₀TCID₅₀/mL over time (Figure 2A). From the posterior 113 distributions on decay slope parameters we computed posterior distributions for the half-life of each virus 114 in each condition (Figure 2B, Table 1). HCoV-19 and SARS-CoV exhibited similar half-lives in aerosols, 115 with median estimates around 2.7 hours, and 95% credible intervals (2.5%-97.5% quantile range) of 116 (1.65, 7.24 hours) for HCoV-19 and (1.81, 5.45 hours) for SARS-CoV-1 (Table 1). Half-lives on copper 117 were also similar between the two viruses. On cardboard, HCoV-19 showed a considerably longer half-118 life than SARS-CoV-1. Both viruses showed markedly longer viability on stainless steel and 119 polypropylene: the median half-life estimate for HCoV-19 was roughly 13 hours on steel and 16 hours on 120 polypropylene. In general, there was no statistically discernable difference in half-life between the two

viruses on any given surface except for cardboard: all other 95% credible intervals for the difference in half-lives overlapped 0 (Fig 2B, Table 1).

Discussion

HCoV-19 has caused many more cases of illness and resulted in more deaths than SARS-CoV-1 and is proving more difficult to contain. Our results indicate that the greater transmissibility observed for HCoV-19 is unlikely to be due to greater environmental viability of this virus compared to SARS-CoV-1. Instead, there are a number of potential factors which could account for the epidemiological differences between the two viruses. There have been early indications that individuals infected with HCoV-19 may shed and transmit the virus while pre-symptomatic or asymptomatic^{4,18-20}. This reduces the efficacy of quarantine and contact tracing as control measures relative to SARS-CoV-1.²¹ Other factors likely to play a role include the infectious dose required to establish an infection, the stability of virus in mucus, and environmental factors such as temperature and relative humidity. ^{16,22} In ongoing experiments, we are studying virus viability in different matrices, such as nasal secretion, sputum and fecal matter, and while varying environmental conditions, such as temperature and relative humidity.

The epidemiology of SARS-CoV-1 was dominated by nosocomial transmission and SARS-CoV was detected on variety of surfaces and objects in healthcare settings. HCoV-19 transmission is also occurring in hospital settings, with over 3000 reported cases of hospital-acquired infections. These cases highlight the vulnerability of healthcare settings for introduction and spread of HCoV-19. However, in contrast to SARS-CoV-1, most secondary transmission has been reported outside healthcare settings and widespread transmission in the community is being seen in several settings, such as households, workplace and group gatherings.

A notable feature of SARS-CoV-1 was super-spreading events, in which a single infected individual was responsible for a large number of secondary cases, well above the average number denoted by the reproduction number $R_{\rm eff}$. ^{7-11,20}A tendency toward such super-spreading events has two important consequences for the epidemiology of emerging infections: it makes any given introduction of infection

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more likely to die out by chance, but when outbreaks do occur they are explosive and can overwhelm hospital and public health capacity.²⁴ A number of hypothesized super-spreading events have been reported for HCoV-19. Given that SARS-CoV-1 superspreading events were linked to aerosol and fomite transmission, ⁶⁻¹¹ our finding that HCoV-19 has viability in the environment comparable to that of SARS-CoV-1 lends credence to the hypothesis that it too may be associated with superspreading. We found that the half-life of HCoV-19 on cardboard is longer than the half-life of SARS-CoV-1. It should be noted that individual replicate data were noticeably noisier for this surface than the other surfaces tested (Figures S1–S5), so we advise caution in interpreting this result. Here, we show that the stability of HCoV-19 and SARS-CoV-1 under the experimental circumstances tested is similar. Taken together, our results indicate that aerosol and fomite transmission of HCoV-19 are plausible, as the virus can remain viable in aerosols for multiple hours and on surfaces up to days. Acknowledgements We would like to thank Kwe Claude Yinde and Michael Letko for experimental assistance. This research was supported by the Intramural Research Program of the National Institute of Allergy and Infectious Diseases (NIAID), National Institutes of Health (NIH). JOL-S and AG were supported by the Defense Advanced Research Projects Agency DARPA PREEMPT # D18AC00031, and JOL-S was supported by the U.S. National Science Foundation (DEB-1557022) and the Strategic Environmental Research and Development Program (SERDP, RC□2635) of the U.S. Department of Defense. The findings and conclusions in this report are those of the author(s) and do not necessarily represent the official position of the Centers for Disease Control and Prevention. Names of specific vendors, manufacturers, or products are included for public health and informational purposes; inclusion does not imply endorsement of the vendors, manufacturers, or products by the Centers for Disease Control and Prevention or the US

Code and data availability

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- 174 Code and data to reproduce the Bayesian estimation results and produce corresponding figures are
- archived online at OSF: <insert link> and available on Github: <insert link>
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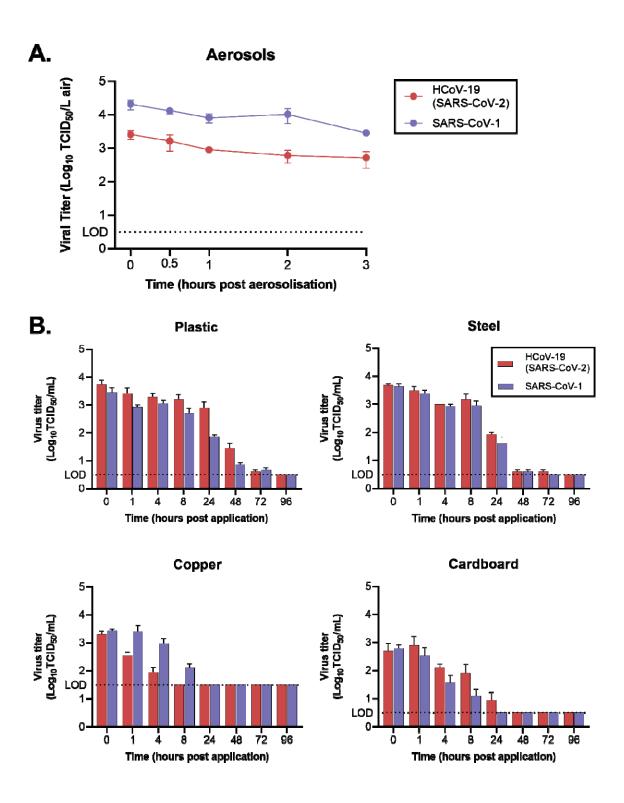


Figure 1. Viability of SARS-CoV and HCoV-19 in aerosols and on different surfaces. A) SARS-CoV and HCoV-19 were aerosolized in a rotating drum maintained at 21-23°C and 65% RH. Aerosols were

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maintained over 180 minutes and samples were collected at 0-, 30-, 60-, 120- and 180-minutes post aerosolization. Viable virus titer per liter of air is shown in TCID₅₀/L air. B) 50 μl of 10⁵ TCID₅₀/mL of SARS-CoV and HCoV-19 was applied on plastic, steel, copper and cardboard surfaces. At 1, 4, 8, 24, 48, 72, and 96 hours samples were obtained for viability assessment. All samples were quantified by endpoint titration on Vero E6 cells. Plots show the mean and standard error across three replicates. Dotted line shows Limit of Detection (LOD), 10^{0.5} TCID₅₀/mL for plastic, steel and cardboard and 10^{1.5} TCID₅₀/mL for copper.

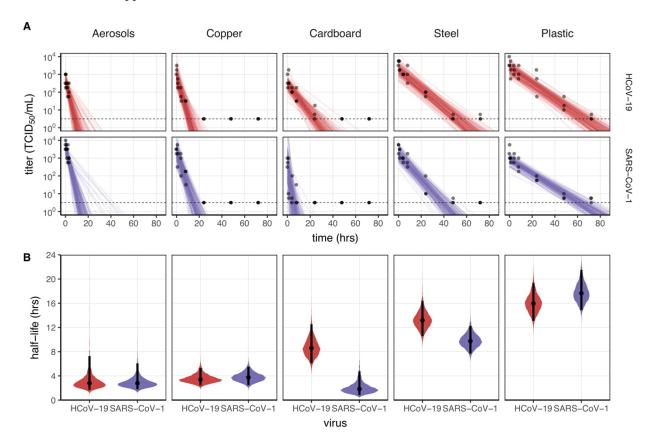


Figure 2. Estimated exponential decay rates and corresponding half-lives for HCoV-19 and SARS-CoV-1. Experimental conditions are ordered by posterior median half-life for HCoV-19. A: Regression plots showing predicted decay of virus titer over time; titer plotted on a logarithmic scale. Points show measured titers and are slightly jittered along the time axis to avoid overplotting. Lines are random draws from the joint posterior distribution of the exponential decay rate (negative of the slope) and intercept (initial virus titer), thus visualizing the range of possible decay patterns for each experimental condition.

150 lines per panel: 50 lines from each plotted replicate. Dotted line shows Limit of Detection (LOD), $10^{0.5}$ TCID₅₀/mL. B: Violin plots showing posterior distribution for half-life of viable virus. Dot shows the posterior median estimate and black line shows a 95% credible interval.

Table 1. Posterior median estimates and 95% credible intervals (2.5%–97.5% quantile range) for half-lives of HCoV-19 and SARS-CoV in aerosols and on various surfaces, as well as a median estimate and 95% credible interval for the difference between the two half-lives (HCoV-19 – SARS-CoV).

		HCoV-19	•	SARS-CoV-1			HCoV-19 - SARS-CoV-1		
	half-life (hrs)			half-life (hrs)			difference (hrs)		
Material	median	2.5%	97.5%	median	2.5%	97.5%	median	2.5%	97.5%
Aerosols	2.74	1.65	7.24	2.74	1.81	5.45	-0.00418	-2.72	4.45
Copper	3.4	2.4	5.11	3.76	2.43	5.43	-0.321	-2.31	1.78
Cardboard	8.45	5.95	12.4	1.74	0.827	4.42	6.6	3.07	10.7
Steel	13.1	10.5	16.1	9.77	7.69	12.3	3.36	-0.173	7.12
Plastic	15.9	13	19.2	17.7	14.8	21.5	-1.79	-6.31	2.51
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