Size and culturability of human-generated SARS-CoV-2 aerosol

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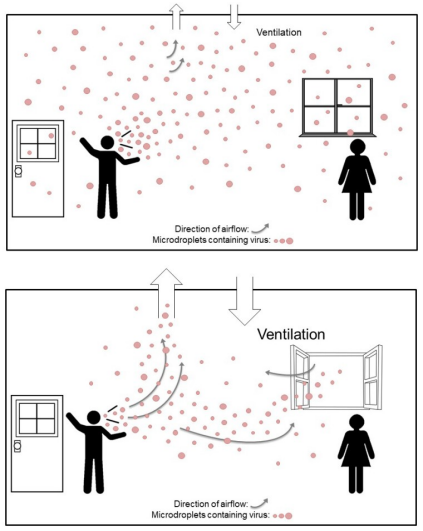
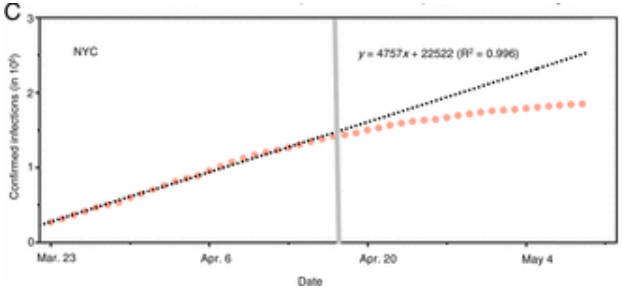
Is SARS-CoV-2 Airborne?

- The question of airborne transmission of SARS-CoV-2 is complex
- This talk will examine the portion of that question related to human production of infectious, viral aerosol
- COVID-19 is a respiratory disease, so we know that virus is produced in the respiratory tract (e.g. Zou, L., et al. 2020)
- We also know that aerosol produced in the lung and larynx are small (less than 1 µm to a few µm) and are produced by breathing and talking, as well as coughing (Morawska, et al. 2009; Johnson, et al. 2011; Somsen, et al. 2020)
- We also know that asymptomatic and presymptomatic people can transmit the virus (Gao, et al., 2020; He, et al., 2020; Oran, et al., 2020; Wei , et al., 2020; Furukawa, et al., 2020; Zou, et al., 2020)
- Several transmission events have suggested the potential for the involvement of aerosols in transmission
    - Washington Choir Practice (Hamner, et al., 2020)
    - Guangzhou Restaurant (Lu, et al., 2020)
- The impact of mask wear (Zhang et al., 2020) and the synthesis of mounting data (e.g. Borak, 2020; Morawska and Milton, 2020) have led to a growing consensus of the importance of airborne transmission in the COVID-19 pandemic



Morawska and Milton, 2020



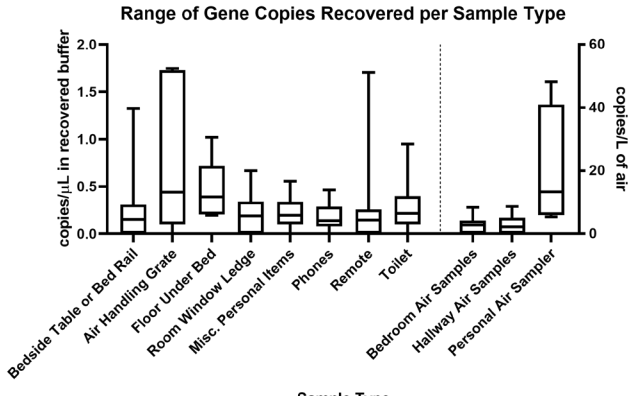
Zhang et al., 2020

Early Studies of SARS-CoV-2 in Hospitals

Table 2. Environmental and PPE Sites Sampled and Corresponding RT-PCR Results

Sites[a]	Positive samples (patient C; before routine cleaning)[b]	Cycle threshold value[c]
Environmental sites[d]		
Patient's room		
1. Cardiac table, including handle	1/1	35.44
2. Entire length of bed rail	1/1	37.95
3. Control panel on bed	0/1	
4. Call bell attached to bed	0/1	
5. Locker with hand slot	1/1	36.21
6. Chair	1/1	37.07
7. Light switches behind bed	1/1	37.54
8. Stethoscope	1/1	38.24
9. Sink, external rim	1/1	35.54
10. Sink, internal bowl	1/1	36.79
11. Floor	1/1	30.64
12. Glass window in room	1/1	35.79
13. Glass door interior	1/1	35.71
14. PPE storage area over sink	1/1	34.89
15. Air outlet fan	2/3	32.96, 37.94
Toilet area		
16. Door handle	1/1	35.83
17. Toilet bowl, surface	1/1	37.75
18. Hand rail	0/1	
19. Sink, external rim	0/1	
20. Sink, internal bowl	1/1	37.11
Anteroom		
21. Sink, external rim	0/1	
22. Sink, internal bowl	0/1	
23. Floor	0/1	
24. Glass door, room side	0/1	
25. Glass door, corridor side	0/1	
Corridor outside room		
26. Floor	0/1	
Total, No. (%)	17/28 (61)	
Staff PPE sites		
Upper front part of gown	0/2	
Lower front part of gown	0/2	
Front surface of face visor mask	0/2	
Front surface of N95 mask	0/2	
Surface of front of shoes	1/2	38.96

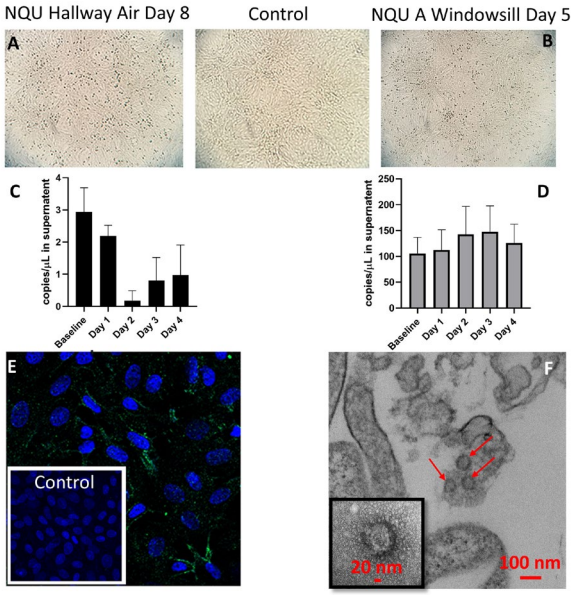
Ong, S. W. X., et al. 2020



Santarpia et al., 2020

Patient	Day of illness	Symptoms reported on day of air sampling	Clinical Ct value[a]	Airborne SARS-CoV-2 concentrations (RNA copies m^{-3} air)	Aerosol particle size	Samplers used
1	9	Cough, nausea, dyspnea	33.22	ND	>4 μm	NIOSH
				ND	1–4 μm	
				ND	<1 μm	
				ND	–	SKC filters
2	5	Cough, dyspnea	18.45	2,000	>4 μm	NIOSH
				1,384	1–4 μm	
				ND	<1 μm	
3	5	Asymptomatic[b]	20.11	927	>4 μm	NIOSH
				916	1–4 μm	
				ND	<1 μm	

Chia, P. Y., et al. 2020



Santarpia et al., 2020

- Several studies of air and surface contamination in rooms housing COVID-19 patients indicated widespread contamination
- In addition air samples or samples around ventilation indicated the potential role of fine aerosols in the observations
- Convincing data around the culturability of these aerosols was elusive

Ong, S. W. X., et al. 2020; Chia, P. Y., et al. 2020; Guo, Z. D., et al. 2020; Nissen, Karolina, et al. 2020; Santarpia, J.L, *et al.* 2020

The Size of Human-Generated SARS-CoV-2 aerosol

Patient	Day of illness	Symptoms reported on day of air sampling	Clinical Ct value[a]	Airborne SARS-CoV-2 concentrations (RNA copies m^{-3} air)	Aerosol particle size	Samplers used
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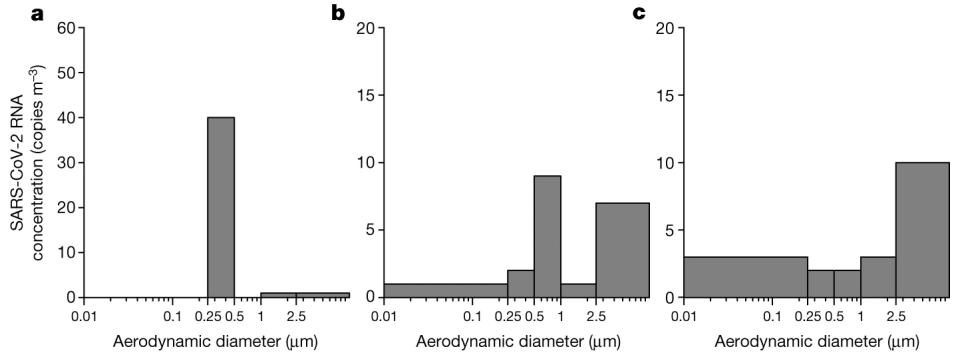
Chia, et al. 2020



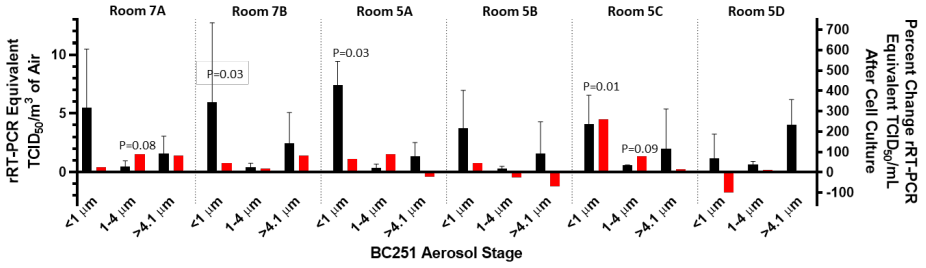
Liu, et al., 2020

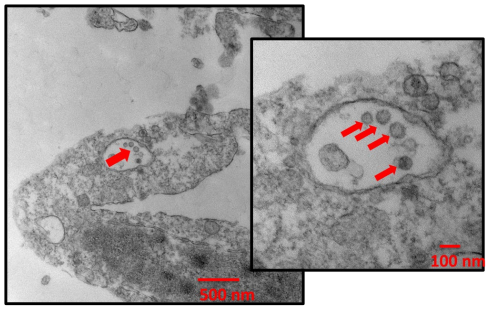
SARS-CoV-2 Detected in Aerosol by PCR and Cell Culture		
	rRT-PCR	Cell Culture
>4.1 μm	6:6 (100%)	0:6 (0%)
1-4 μm	6:6 (100%)	2:6 (33%)*
< 1 μm	6:6 (100%)	3:6 (50%)

Santarpia, et al., 2020

- Three studies have attempted to directly address the size of human-generated SARS-CoV-2 aerosol
- All three studies found evidence of SARS-CoV-2 in particles less that 5 μm
- 2 of the 3 studies found evidence for particles less than 1 μm

Aerosol Samples Replicating in Cell Culture

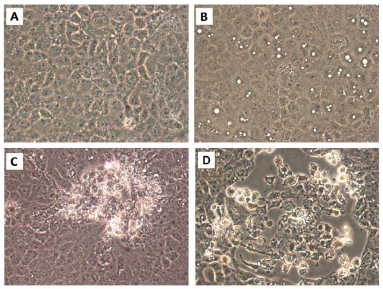




Santarpia et al., 2020

Sample ID	Virus genome equivalents/L of air[a]	$TCID_{50}$/100 µl	Viable virus count/L air
1-1 BioSpot	94	2.68E+04	74
1-2 BioSpot + HEPA	-	0	0
1-3 BioSpot	30	6.31E+03	18
2-1 VIVAS	44	1.00E+04	27
2-2 VIVA S+ HEPA	-	0	0
2-3 VIVAS	16	2.15E+03	6

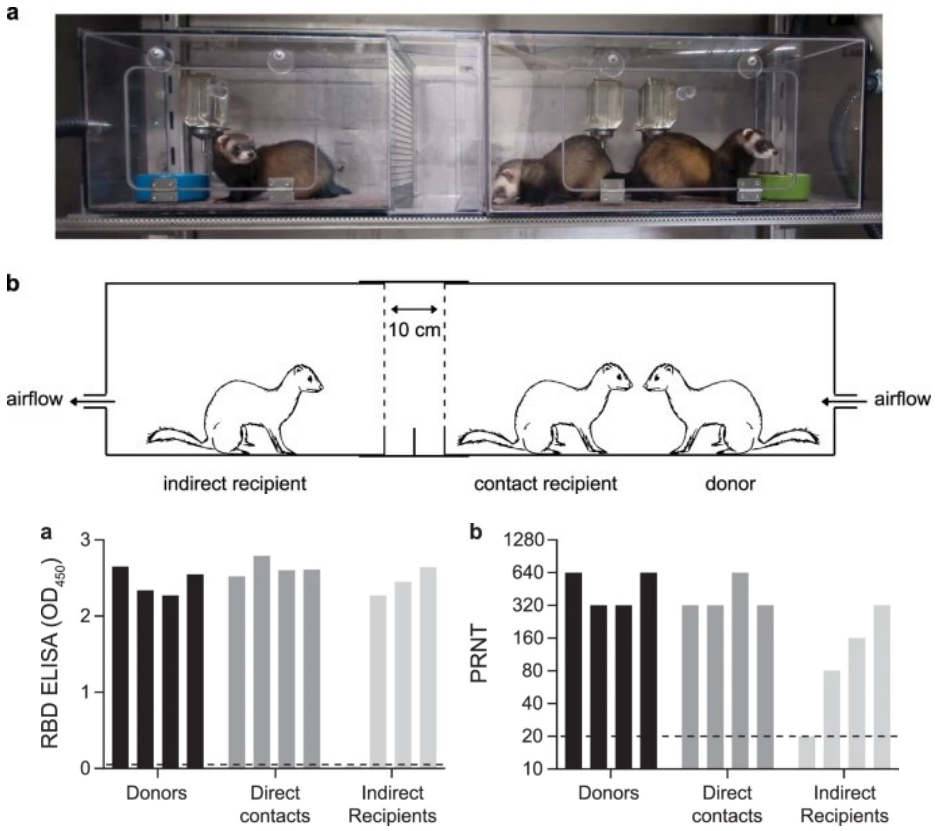
[a]From Table 2.



Lednicky et al., 2020

- Two pre-print studies indicate that aerosol particles generated by patients can replicate in cell culture
- Santarpia, et al found evidence of replication (through serial PCR and EM) of replication of collected sub-micron particles
- Lednicky, et al quantified culturable viral aerosol collected at 2 and 4.8 m from patients

Animal Studies



Richard et al., 2020

- Two studies in ferrets have indicated that indirect transmission of SARS-CoV-2 by the air is possible
- Separation distance was relatively small, so its difficult to complete rule out the role of larger particles

Richard et al., 2020;

So, what do we know?

- The rooms of people infected with COVID-19 show widespread contamination of both surfaces and air, with indications that aerosols may be involved (sample distance, ventilation, etc.)
- Asymptomatic and presymptomatic people are known to spread the virus, indicating the smaller aerosols produced only during breathing and speaking may carry the virus
- Aerosols less than 4 microns (even less than 1 micron) have been shown to contain viral RNA
- Aerosols from patient rooms have been cultured in Vero cells, and submicron samples from patient rooms have demonstrated replication in cell culture
- Ferrets have been shown to transmit the SARS-CoV-2 virus through the air

Humans infected with SARS-CoV-2 can produce infectious fine mode particles that may be able to transmit the disease after exposure to enough particles.

Additional Questions

- At what rate do people produce infectious aerosol?
- How does the production of infectious aerosol vary from person to person?
- How does the production of infectious aerosol change over the course of illness?
- What is the infectious dose of SARS-CoV-2 through the aerosol route?

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