AIR-SURFACE-AIR NEXUS IN VIRUSES SPREAD INDOOR: BREAKING THE CHAIN OF INFECTION

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LEEDS / HEALTHY BUILDINGS – JANUARY 23, 2025

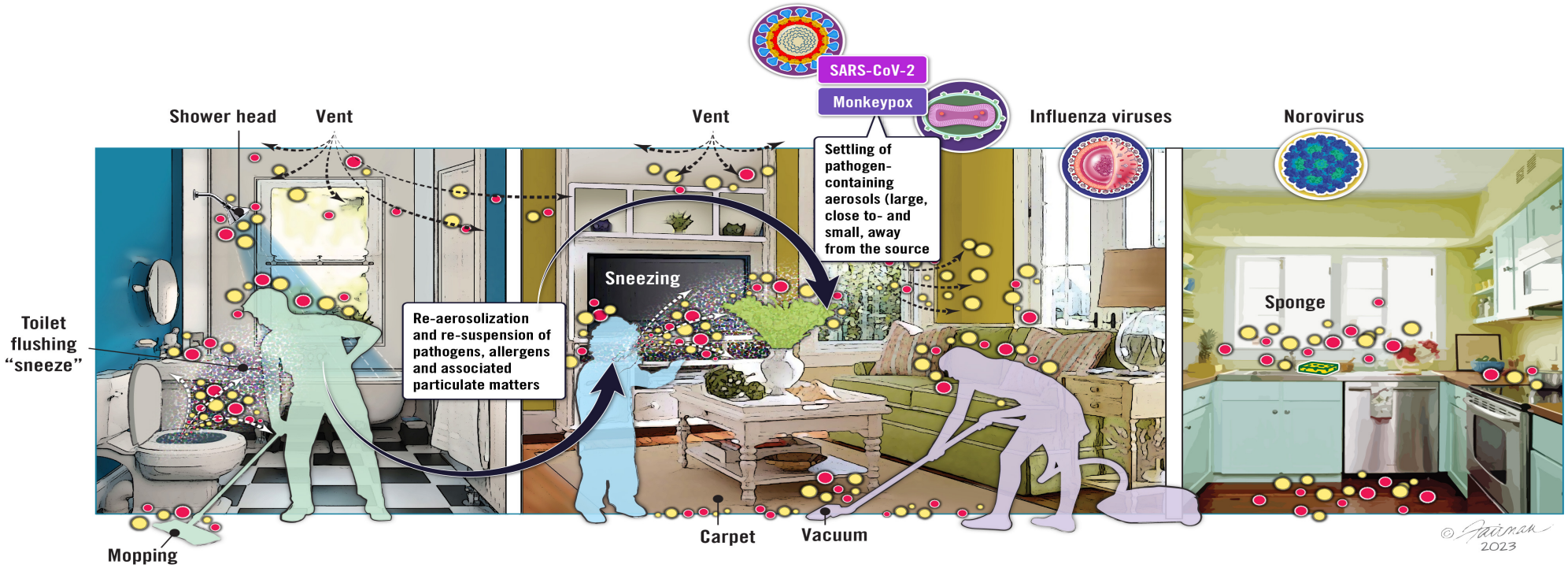
OUTLINE

1. Viruses Spread indoor in the backdrop of SARS-CoV-2/COVID-19 Pandemic & Noroviruses in restrooms
2. Role of air and fomites in virus transmission
3. Lysol Air Sanitizer: protocol development and use in assessment against airborne pathogens
4. Toilet flush generated aerosolized pathogens and cross-contamination reduction by Lysol Air Sanitizer
5. Summary

Pathogens spread indoors: Air-surfaces nexus – dynamic interdependencies & opportunities for re-aerosolization of such pathogens

Toilets sneeze as well!

- **Droplets** directly or indirectly [contaminated surfaces touch]
- **Aerosols** carrying pathogens
- **Re-aerosolized** pathogens

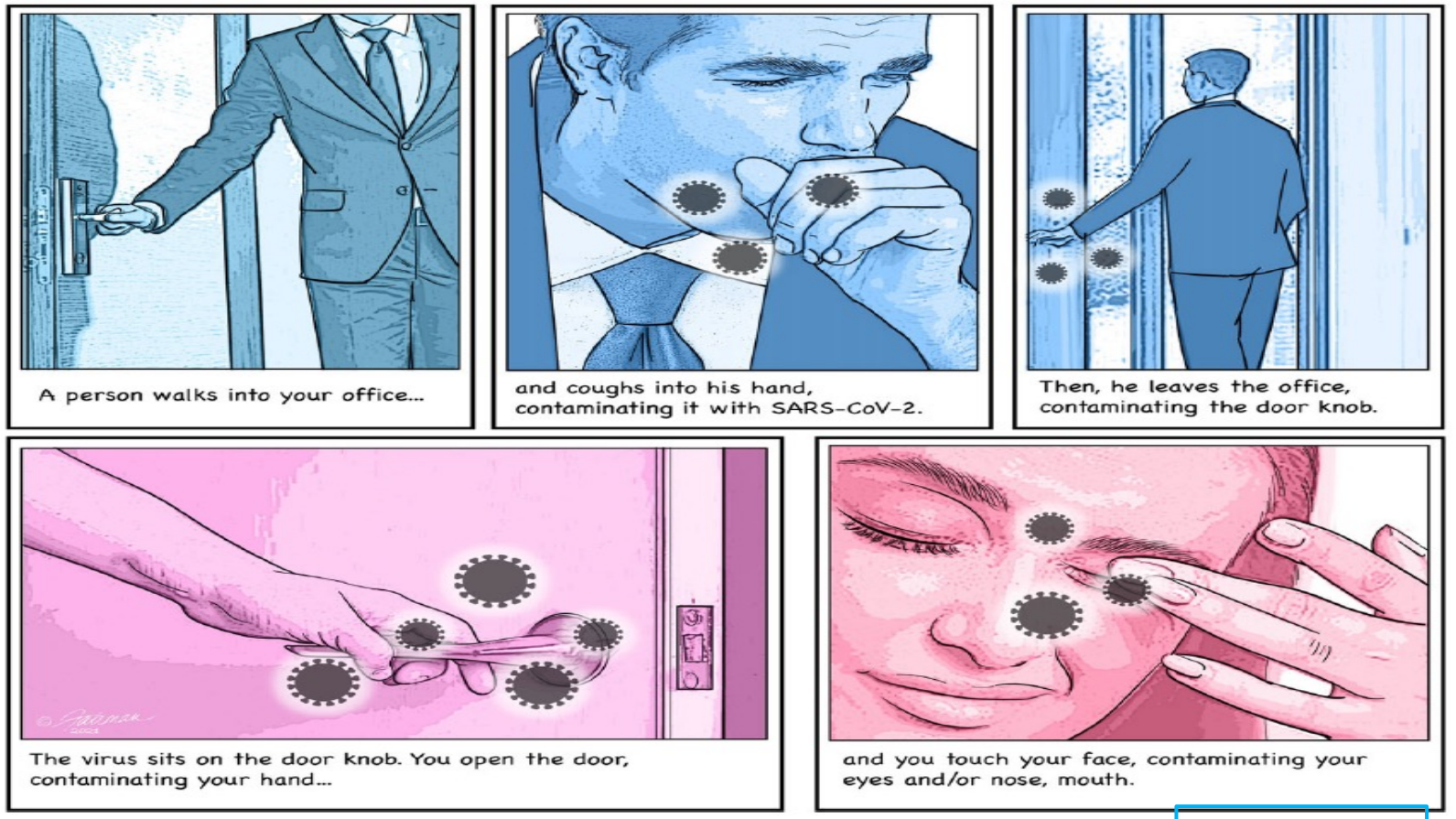


***Predisposing to Air-surface-air nexus!**

These sources may include humans, pets, plants, plumbing systems including operational toilets, shower head, heating, ventilation, vacuuming, mopping, air-conditioning systems; resuspension of settled dust indoor. The red and yellow dots represent human pathogens or harmless microorganisms.

Ijaz et .al., 2023. Environmental dissemination of respiratory viruses: dynamic interdependencies of respiratory droplets, aerosols, aerial fomites, high-touch environmental surfaces, and contribution of viral re-aerosolization / https://peerj.com/articles/16420/

SARS-CoV-2 Transmission from one person to another through virus-contaminated HITES



Ijaz et al 2021

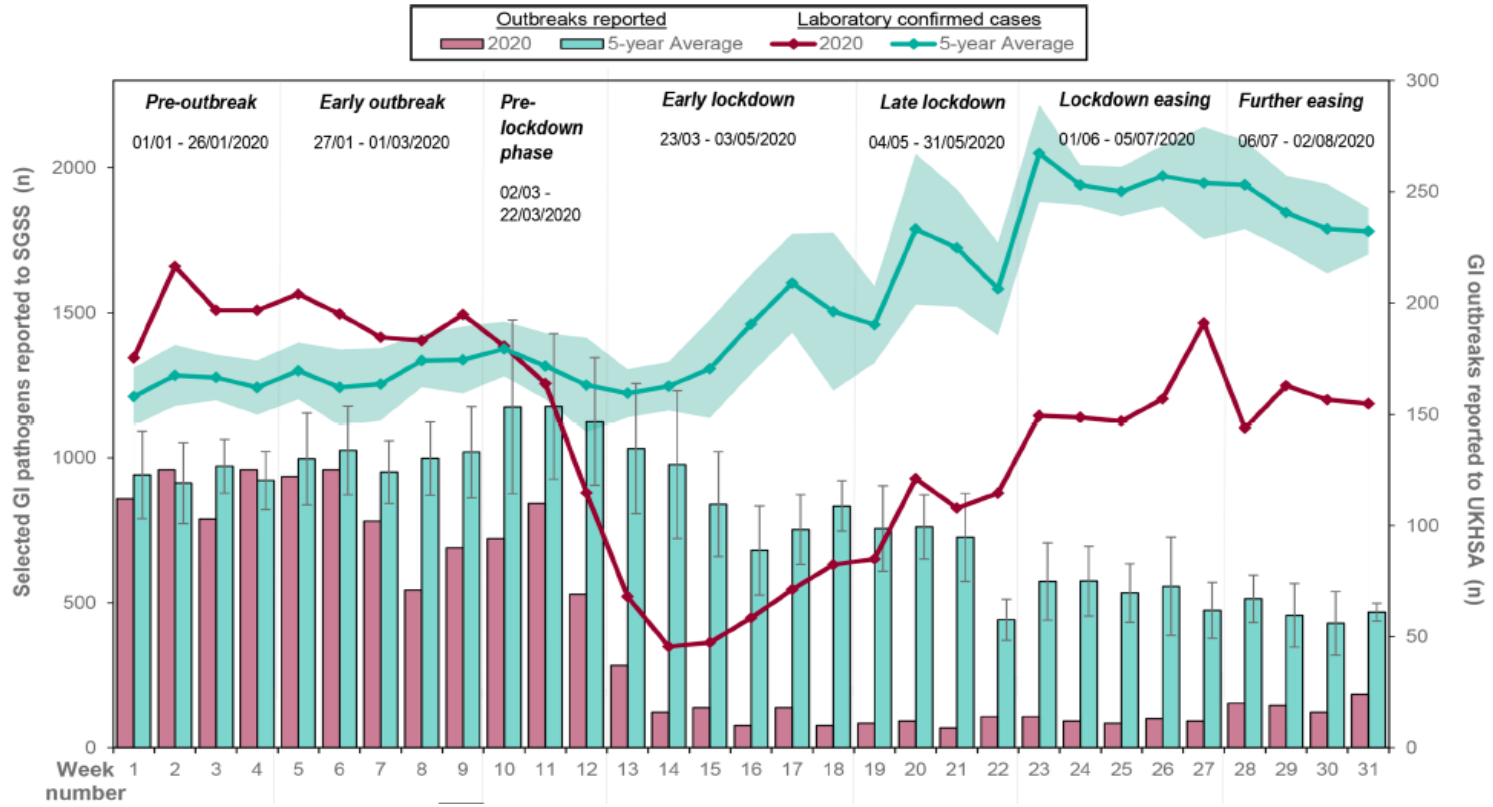
Infection Control & Hospital Epidemiology (2021), 1–2
doi:10.1017/ice.2021.57

Letter to the Editor

Indirect transmission of severe acute respiratory syndrome coronavirus virus 2 (SARS-CoV-2): What do we know and what do we not know?

M. Khalid Ijaz DVM, MSc(Hons), PhD, FRSPH[1,2], Raymond W. Nims PhD[3] and Julie McKinney PhD[1]
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Public Health Measures during COVID-19 & GI outbreaks/pathogens in 2020 compared to 5 years wkly



Open access Original research

BMJ Open

Impact of the COVID-19 pandemic on gastrointestinal infection trends in England, February–July 2020

Nicola K Love [1,2] Alex J Elliot [1,2,3] Rachel M Chalmers,[4] Amy Douglas,[1] Saheer Gharbia,[1] Jacquelyn McCormick,[1] Helen Hughes,[1,2] Roger Morbey,[1,3] Isabel Oliver,[1,5] Roberto Vivancos,[1,2] Gillian Smith [1,2,3]

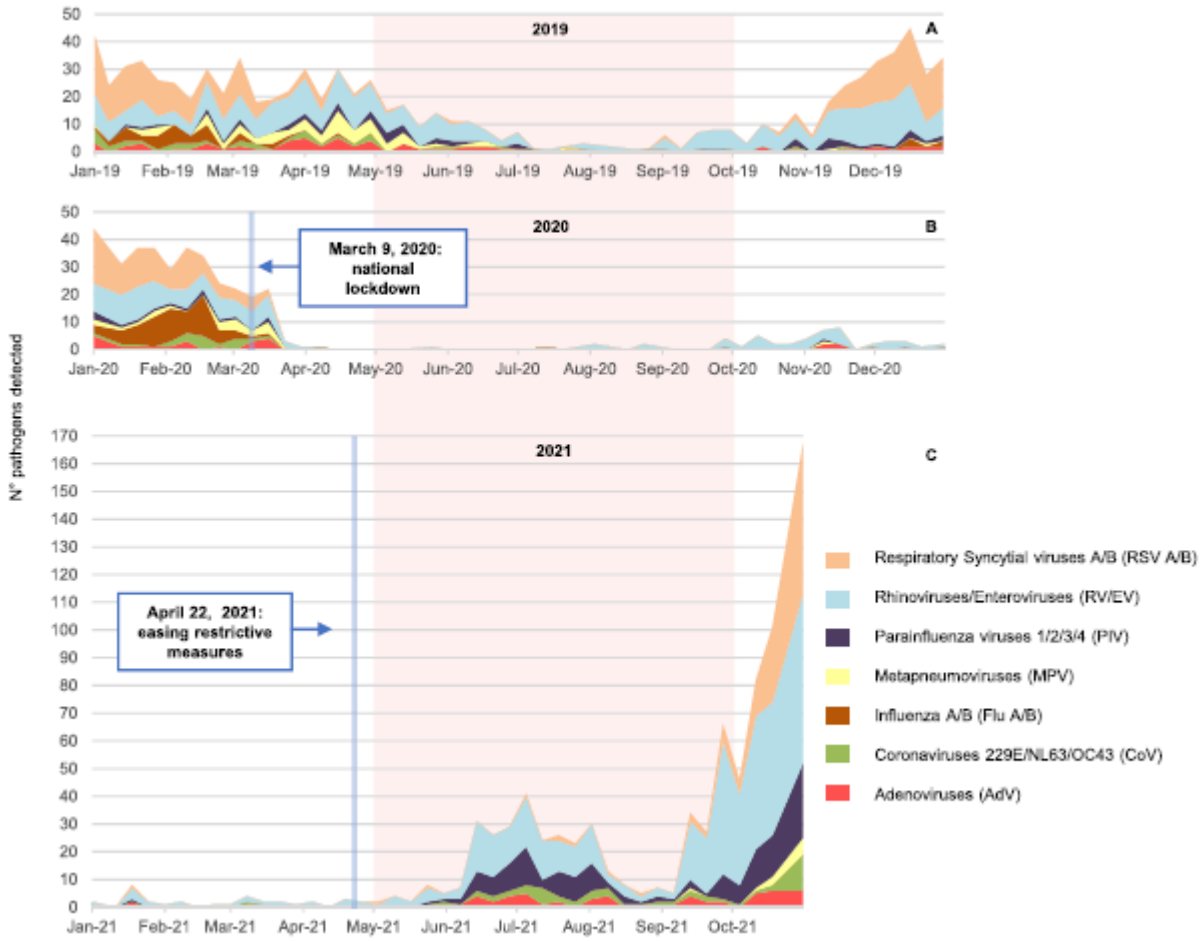
Love et al 2022

Drivers in changes of behavior/applications of NPIs including

1. **Mask**
2. **Improved hand hygiene**
3. **Social distancing**

*__Enteric pathogens__: *Campylobacter spp*, STEC O157, STEC non-O157, *Listeria spp*, non-typhoidal *Salmonella spp*, typhoidal *Salmonella*, *Shigella spp*, norovirus, *Cryptosporidium spp* and *Giardia sp*.

Number of different viral pathogens detected in (a) January–December 2019, (b) January–December 2020 and (c) January–October 2021 - Italy



Journal of Global Antimicrobial Resistance 28 (2022) 223–225

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Journal of Global Antimicrobial Resistance

journal homepage: www.elsevier.com/locate/jgar

SARS CoV-2 Dispatches

Back to the future (of common respiratory viruses)

Giordana Mattana[a], Sami Albitar-Nehme[a], Valeria Cento[b,c], Luna Colagrossi[a], Livia Piccioni[a], Massimiliano Raponi[d], Umberto Raucci[e], Anna Chiara Vittucci[f], Antonino Reale[e], Alberto Villani[f], Paola Bernaschi[a], Carlo Federico Perno[a,*]

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[d] Health Direction, Bambino Gesù Children's Hospital, IRCCS, Rome, Italy
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Mattana et al 2023

- The pink area highlights the three periods of five months (May–September) with the highest temperature, and the **white area highlights the months with the lowest temperature** (January–April and October–December).

Infectious viruses recovered from the field air samples

Table 1 Studies reporting recovery of infectious respiratory virus from air samples in field studies.

Virus	Setting	Host Cell	Sampler used	RNA load*	Positive recoveries	Comment	Reference
Smallpox virus	Healthcare (India)	Chicken egg	Porton Settling plates	NP	5 of 47 12 of 30	Air sampled from within 12″of patient's mouth	*Downie et al. (1965)*
SARS-CoV-2	Healthcare (USA)	LLC-MK2, Vero E6	BioSpot VIVAS BSS300PVIVAS prototype	Ct values ranged from 36.00 to 38.69	4 of 4	Air samples from within 2 m of the patients, with 72 h and ≥96 h duration of symptoms.	*Lednicky et al. (2020)*
SARS-CoV-2	Automobile (USA)	Vero E6	Sioutas PCIS	Ct value of 33.50	1 of 5	Driver of the car had 48 h symptomology when sampling occurred	*Lednicky et al. (2021)*
SARS-CoV-2	Laboratory (USA)	Vero E6-TMPRSS2, A549-ACE2	Gesundheit-II	$>10^4$ GC, <75 GC	2 of 100	Samples of exhaled air from COVID-19 cases with early mild infection	*Adenaiye et al. (2022)*
SARS-CoV-2	Residential bedroom (USA)	LLC-MK2, Vero E6	BioSpot VIVAS 300, NIOSH Model BC-251	2.3×10^6 GE, 1.0×10^6 GE	2 of 9	Bedroom air samples for a COVID-19 patient	*Vass et al. (2022)*
SARS-CoV-2	Laboratory USA)	Vero E6-TMPRSS2, A549-ACE2	Gesundheit-II	3.0×10^4 GC, 3.6×10^2 GC, 1.8×10^7 GC, 2.9×10^3 GC	4 of 62	Samples of exhaled air from COVID-19 cases infected with Delata and Omicron variants	*Lai et al. (2023)*
SARS-CoV-2	Hospital Rooms (Canada)	Vero E6	Liquid Spot Sampler Series 110A, SKC Eighty Four cassettes	Ct value of 32	1 of 4	Air samples collected from hospital rooms of COVID-19 patients	*Fortin et al. (2023)*

Notes.
*RNA load for culture-positive samples.
Ct, real-time PCR cycle threshold; GC, genomic copies; GE, genomic equivalents; NP, not performed.
Additional abbreviations used: h, hours; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2.

PeerJ

Environmental dissemination of respiratory viruses: dynamic interdependencies of respiratory droplets, aerosols, aerial particulates, environmental surfaces, and contribution of viral re-aerosolization

M. Khalid Ijaz[1], Syed A. Sattar[2], Raymond W. Nims[3], Stephanie A. Boone[4], Julie McKinney[1] and Charles P. Gerba[4]

[1] Global Research & Development for Lysol and Dettol, Reckitt Benckiser LLC, Montvale, NJ, United States of America
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[4] Water & Energy Sustainable Technology Center, University of Arizona, Tucson, AZ, United States of America

Infectious viruses recovered from the field fomites samples

Table 2 Studies reporting recovery of infectious respiratory virus from naturally contaminated fomite samples.

Virus	Sampling setting	Host Cell	Fomite sampled	Ct*	Positive Recoveries	Comments	Reference
HCoV-229E	Classroom (USA)	A549, MRC-5, Vero E6	Desk top	NP	4 of 8	No patient data. Surfaces disinfected daily	*Bonny, Yezli & Lednicky (2018)*
			Door knob	NP	2 of 4		
			Bed sheet		1 of 15		
MERS-CoV	Healthcare (South Korea)	Vero E6	Bed rail	No correspondence between Ct and infectious virus isolation was reported	1 of 15		*Bin et al. (2016)*
			IV fluid hanger		2 of 14		
			Computed radiography cassette		1 of 1		
			Anteroom table		1 of 7		
SARS-CoV-2	Healthcare (Canada)	Vero	Used facial tissues	5 to ~32 (Ct ≤ 25 is 84% predictive of infectious virus isolation)	4 of 5	Positive by RNA and by culture optimal within 1 week of onset of symptoms	*Lin et al. (2022)*
			Phone/call bell		2 of 10		
			Nasal prongs		3 of 4		
SARS-CoV-2	Residential (USA)	LLC-MK2, Vero E6	Mobile phone	2.29×10^4 GE	1 of 3	Mobile phone of COVID-19 patient	*Vass et al. (2023)*
Influenza A and B virus	Healthcare (Hong Kong)	MDCK	Cell phone	No correspondence between Ct and infectious virus isolation was reported	1 of 15 (Influenza A) 2 of 8 (Influenza B)	Time from symptom onset to sampling <72 h	*Xiao et al. (2021)*
Monkeypox virus	Residence (USA)	BSC-40	Paper towels on bed	Infectious virus isolates corresponded to Ct values <24.	1 of 1	Sampling occurred 15 days after the infected person left the residence. Infectious virus was isolated more often from porous than non-porous surfaces.	*Morgan et al. (2021)*
			Underwear on bed		2 of 2		
			Blanket on couch		1 of 1		
			Towel on couch		1 of 1		
			Mattress cover in hamper		1 of 1		
			Coffee tabletop		1 of 1		
Smallpox virus	Healthcare (India)	Chicken eggs	Bed pillow	NPNP	41 of 67	Patients in acute illness	*Downie et al. (1965)*
			Bed sheet		15 of 16		

Notes.

*Abbreviations used Ct, real-time PCR cycle threshold; GE, genome equivalents; HCoV-229E, human coronavirus 229E; MERS-CoV, Middle East respiratory syndrome coronaviru
SARS-CoV-2, severe acute respiratory syndrome coronavirus 2.

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Environmental dissemination of respiratory viruses: dynamic interdependencies of respiratory droplets, aerosols, aerial particulates, environmental surfaces, and contribution of viral re-aerosolization

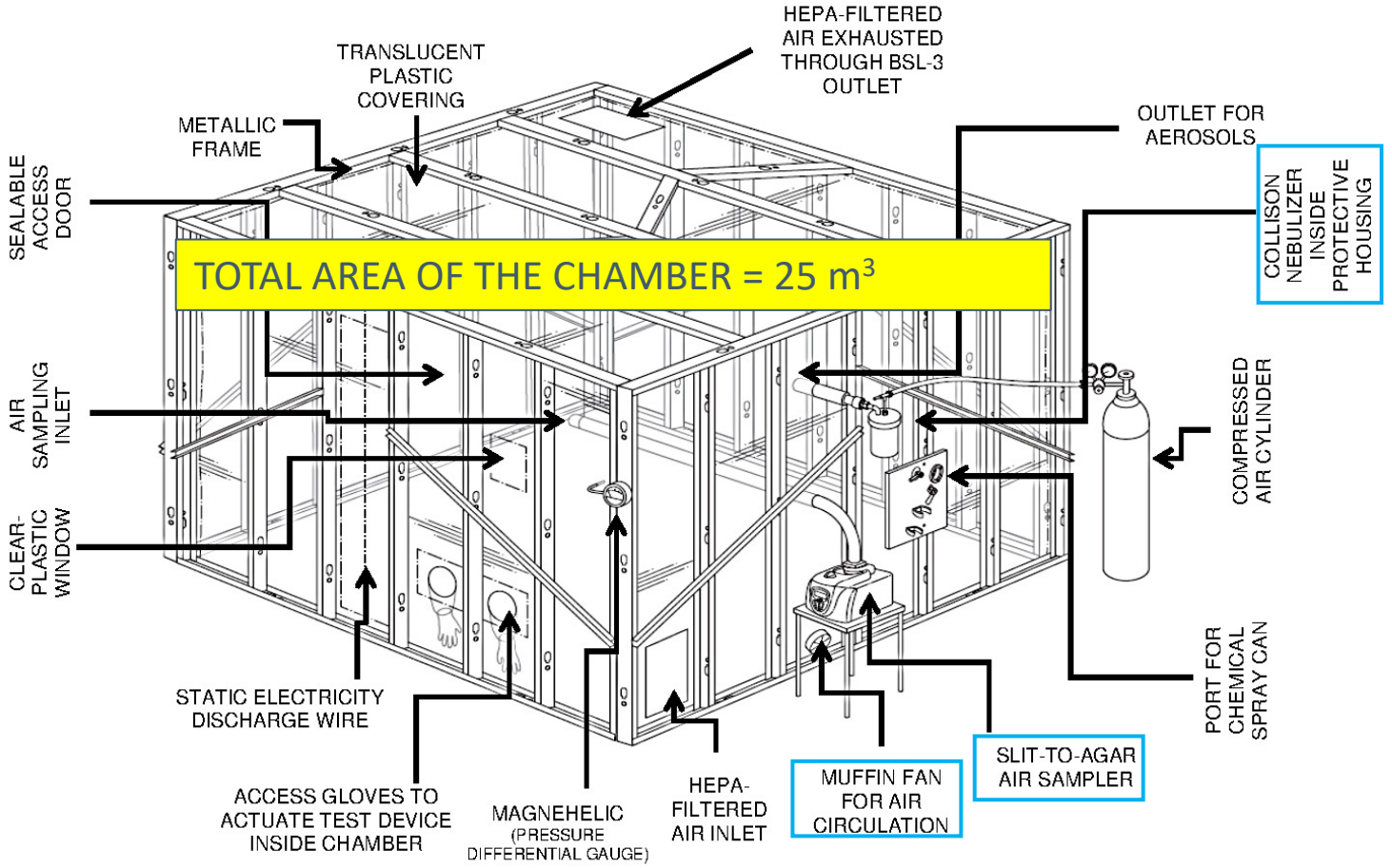
M. Khalid Ijaz[1], Syed A. Sattar[2], Raymond W. Nims[3], Stephanie A. Boone[4], Julie McKinney[1] and Charles P. Gerba[4]

[1] Global Research & Development for Lysol and Dettol, Reckitt Benckiser LLC, Montvale, NJ, United States of America
[2] Department of Biochemistry, Microbiology & Immunology, Faculty of Medicine, University of Ottawa, Ottawa, Ontario, Canada
[3] Syner-G BioPharma, Boulder, CO, United States of America
[4] Water & Energy Sustainable Technology Center, University of Arizona, Tucson, AZ, United States of America

LYSOL AIR SANITIZER TESTING FORMULATIONS AGAINST BACTERIA AND VIRUSES

Test Chamber with Essential Components

Produce them in respirable size → Keep them airborne → Collect them from the air to test viability



American Journal of Infection Control 44 (2016) e177-e182

Contents lists available at ScienceDirect

American Journal of Infection Control

journal homepage: www.ajicjournal.org

Major Article

Decontamination of indoor air to reduce the risk of airborne infections: Studies on survival and inactivation of airborne pathogens using an aerobiology chamber

Syed A. Sattar PhD [a,*], Richard J. Kibbee MLT [a], Bahram Zargar PhD [a], Kathryn E. Wright PhD [a], Joseph R. Rubino MA [b], M. Khalid Ijaz PhD [b]

[a] Department of Biochemistry, Microbiology & Immunology, Faculty of Medicine, University of Ottawa, Ottawa, ON, Canada
[b] Reckitt Benckiser, Research & Development, Montvale, NJ

American Journal of Infection Control 44 (2016) S109-S120

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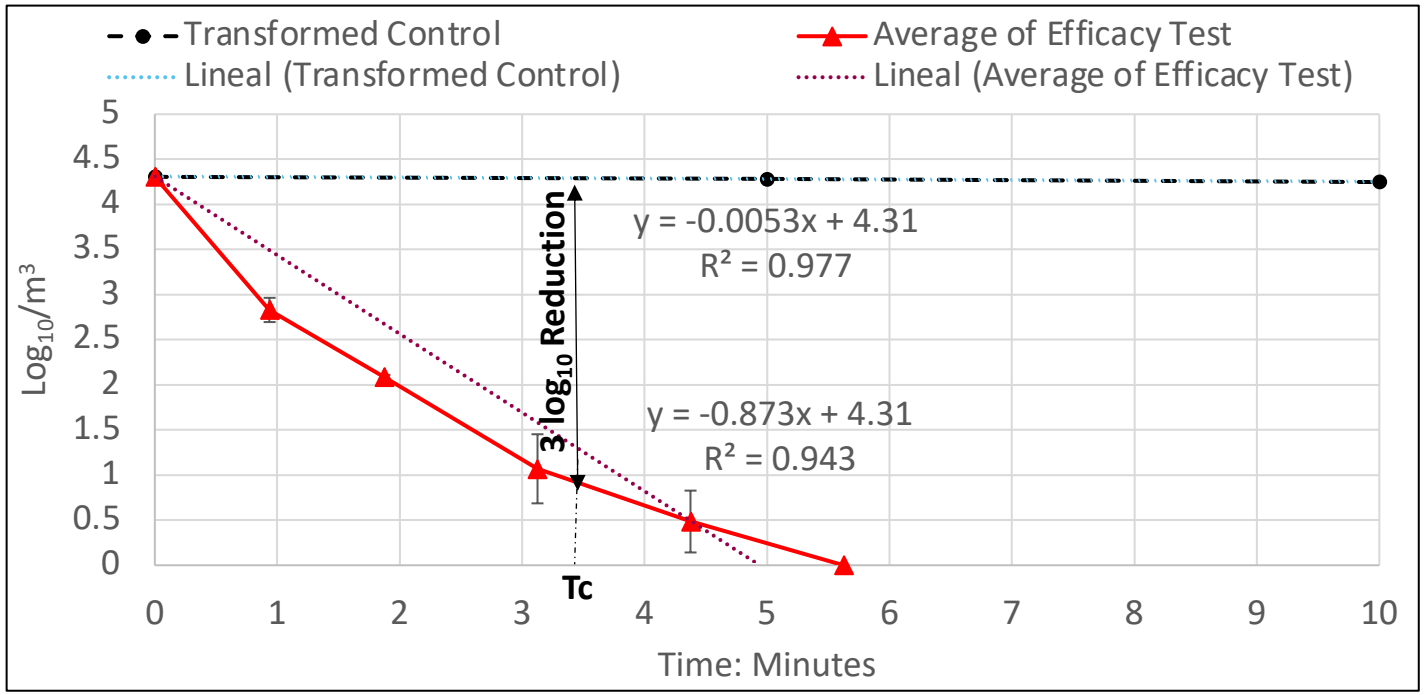
State of the Science Review

Generic aspects of the airborne spread of human pathogens indoors and emerging air decontamination technologies

M. Khalid Ijaz DVM, MSc(Honors), PhD [a,b,*], Bahram Zargar BSc Engg, MSc Engg, PhD [c], Kathryn E. Wright MA, MSc, PhD [c], Joseph R. Rubino BA, MA [a], Syed A. Sattar MSc, Dip Bact, MS, PhD [d]

[a] RB, Montvale, NJ
[b] Department of Biology, Medgar Evers College of the City University of New York (CUNY), Brooklyn, NY
[c] Department of Biochemistry, Microbiology, and Immunology, University of Ottawa, Ottawa, ON, Canada
[d] Professor Emeritus of Microbiology, Faculty of Medicine, University of Ottawa, Ottawa, ON, Canada

Lysol Air Sanitizer: Efficacy against aerosolized *Staphylococcus aureus*



- **Three lots** (e0199-069, e0032-170, e0199-070) **tested (each lot tested in triplicate)** and **demonstrated a 99.9% (3.0-log$_{10}$) reduction in the viability of airborne *S. aureus*** (suspended in ASTM tripartite soil load to simulate body secretions/excretions) **in 3.21 minutes** (3.46, 3.30, 2.86 minutes respectively) and met the acceptance criterion as required by the U.S. EPA

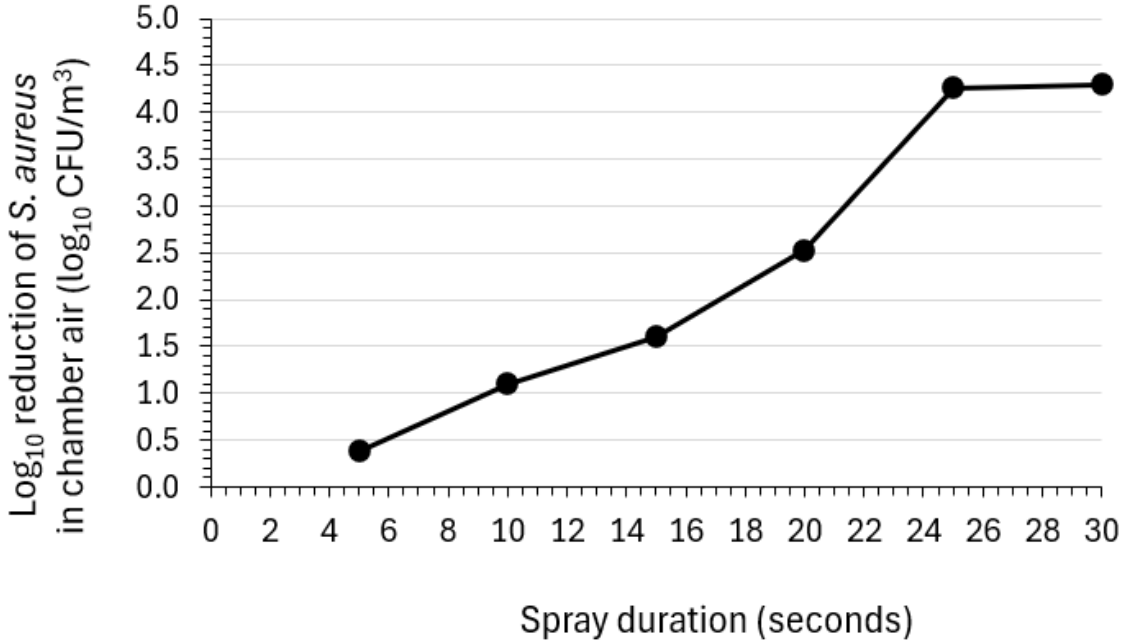
microorganisms MDPI

Article

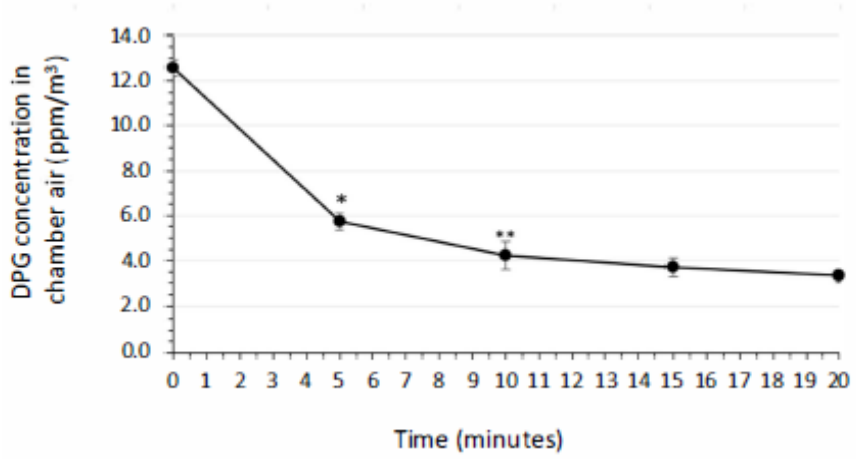
The Determination of the Rapid and Effective Activity of an Air Sanitizer against Aerosolized Bacteria Using a Room-Sized Aerobiology Chamber

Bahram Zargar [1], M. Khalid Ijaz [2,*], Anthony Kevek [2], Mark Miller [2], Julie McKinney [2] and Syed A. Sattar [1,3,*]

Dose response for the bactericidal efficacy of varying durations of spraying the air sanitizer against S. aureus – duration of spray vs. bactericidal activity.



- In six separate experiments, aerosolized *S. aureus* were challenged after which the air sanitizer was sprayed into the aerobiology chamber in increments of five seconds (i.e., 5 s, 10 s, 15 s, 20 s, 25 s, and 30 s).
- As shown in Figure 2, there was a direct correspondence between the duration of the spray and the determined bactericidal activity.



Decay curve of dipropylene glycol (DPG) in the chamber air following a 30-s spray of the air sanitizer.

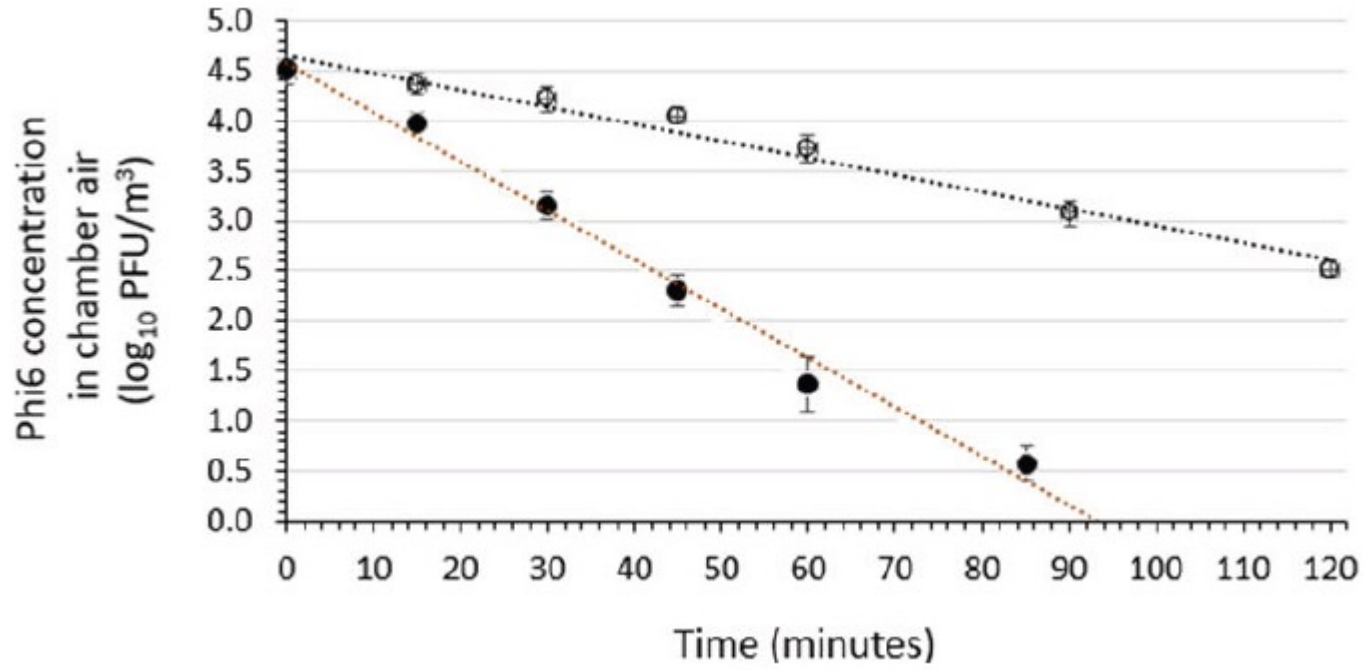


Article

The Determination of the Rapid and Effective Activity of an Air Sanitizer against Aerosolized Bacteria Using a Room-Sized Aerobiology Chamber

Bahram Zargar [1], M. Khalid Ijaz [2,*], Anthony Kevek [2], Mark Miller [2], Julie McKinney [2] and Syed A. Sattar [1,3,*]

In-air Stability of phage Phi6 aerosolized from suspending medium without and with soil load.



- Three independent runs were performed for Phi6 in the presence (open symbols) or absence (closed symbols) of a soil load.

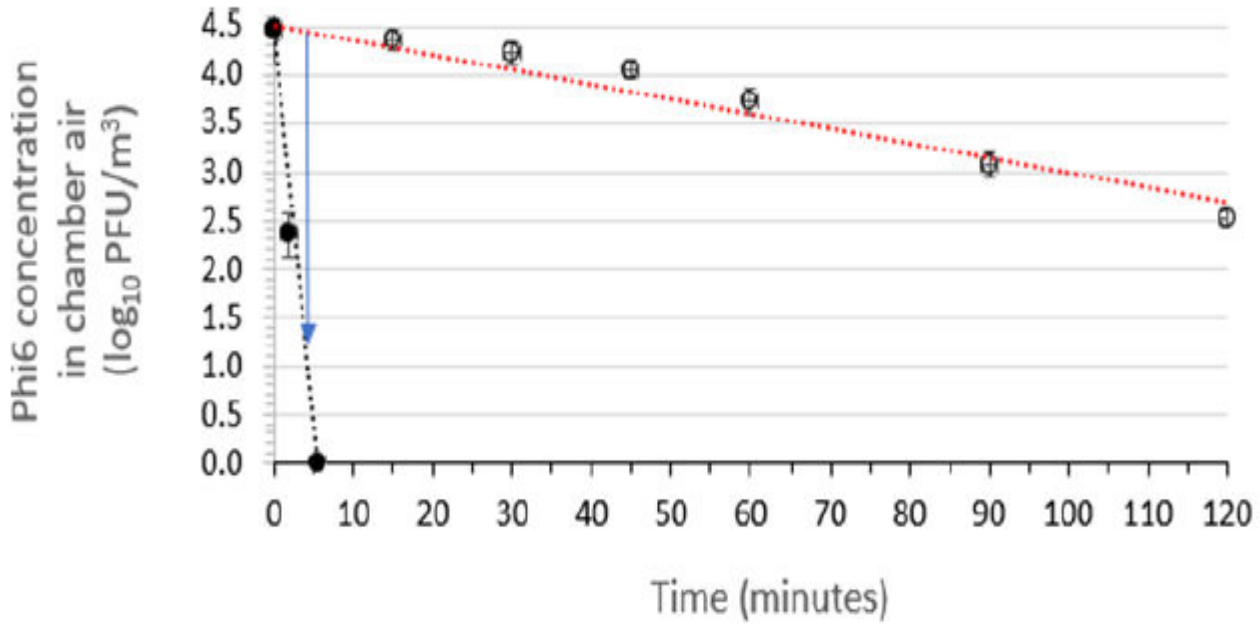
AMERICAN SOCIETY FOR MICROBIOLOGY | Applied and Environmental Microbiology

| Applied and Industrial Microbiology | Full-Length Text

Rapid virucidal activity of an air sanitizer against aerosolized MS2 and Phi6 phage surrogates for non-enveloped and enveloped vertebrate viruses, including SARS-CoV-2

M. Khalid Ijaz,[1] Bahram Zargar,[2] Raymond W. Nims,[3] Julie McKinney,[1] Syed A. Sattar[2,4]

Assessment of the air sanitizer's efficacy for inactivating aerosolized enveloped phage Phi6



- The red dotted line displays the linear regression line fit to the Phi6 concentrations in the absence of air sanitizer (open symbols), while the black dotted line displays the Phi6 post-spraying air sanitizer.
- **A 3.0 log_{10} reduction in viable Phi6 concentration was achieved within 3.6 min of air sanitizer release indicating efficacy against enveloped viruses** (e.g., SARS-CoV-2; influenza; RSV).

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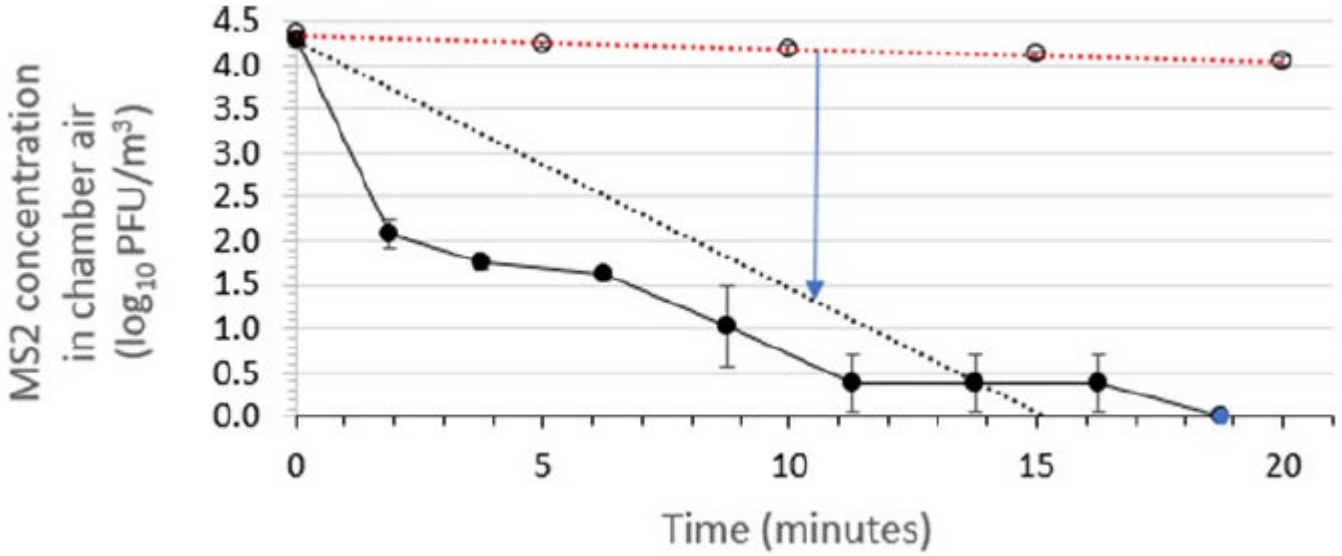
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M. Khalid Ijaz,[1] Bahram Zargar,[2] Raymond W. Nims,[3] Julie McKinney,[1] Syed A. Sattar[2,4]

Assessment of the air sanitizer's efficacy for inactivating aerosolized non-enveloped phage MS2



- Time kinetics for efficacy of **air sanitizer batch 3** for reducing the viability of aerosolized phage MS2 suspended in the soil load in a room-sized aerobiology chamber. The red dotted line displays the linear regression line fit to the MS2 concentrations in the absence of air sanitizer (open symbols), while the black dotted line displays the linear regression line fit to the MS2 concentrations in the presence of air sanitizer (closed symbols). All data points indicate mean ± standard deviation (n = 3 independent runs). **A 3.0 log_{10} reduction in viable MS2 concentration was achieved within 11.3 min** of air sanitizer release indicating efficacy against non-enveloped viruses (e.g., noroviruses and rhinoviruses).

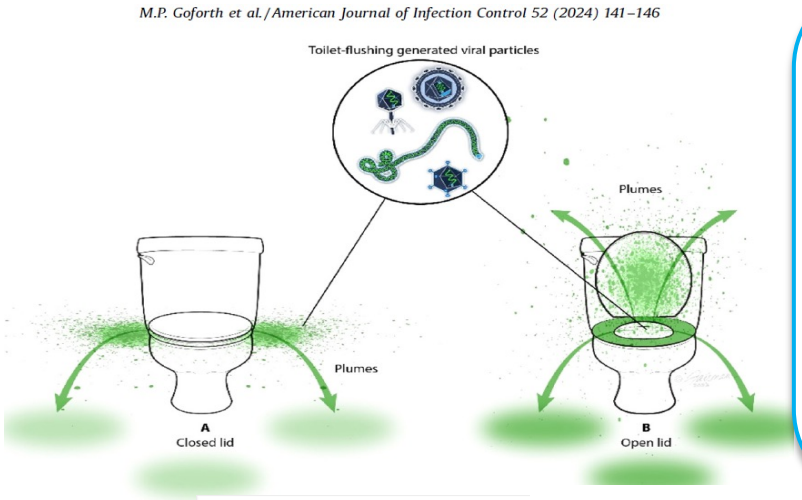
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M. Khalid Ijaz,[1] Bahram Zargar,[2] Raymond W. Nims,[3] Julie McKinney,[1] Syed A. Sattar[2,4]

Impacts of lid closure during toilet flushing and of toilet bowl cleaning on viral contamination of surfaces in United States restrooms



***Press Release**

Lysol Pro Solutions reveals how germs survive and thrive on bathroom surfaces

DISINFECTION IS KEY

- Disinfecting surfaces just twice a week can reduce germ transmission in bathrooms by up to 90%
- Disinfection of the toilet seat, sink countertop and floor are just as important as disinfection of the toilet bowl in preventing the spread of germs

American Journal of Infection Control 52 (2024) 141–146

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American Journal of Infection Control

journal homepage: www.ajicjournal.org

Major Article

Impacts of lid closure during toilet flushing and of toilet bowl cleaning on viral contamination of surfaces in United States restrooms

Madison P. Goforth BS [a], Stephanie A. Boone PhD [a,*], Justin Clark MS [a], Priscilla B. Valenzuela MS [a], Julie McKinney PhD [b], M. Khalid Ijaz DVM, MSc (Honors), PhD, FRSPH [b], Charles P. Gerba PhD [a]

[a] Department of Environmental Science, University of Arizona, Tucson, AZ
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EurekAlert! AAAS | American Society for Microbiology | The University of Arizona | Forbes | USA Today | U.S. News & World Report | Daily Mail .com | abc NEWS | Global News

***Dr. Gerba's team observed**

- With the household toilet, researchers found that there was no statistical difference in the amount of virus collected from surfaces on the toilet or the nearby floor whether the toilet was flushed with the lid up or down.

APIC Association for Professionals in Infection Control and Epidemiology | 50 YEARS 1972-2022

New Analysis Shows that Disinfection Is the Most Effective Way to Prevent Viral Contamination of Restroom Surfaces [Tania Bubb, PhD, RN, CIC, FAPIC, 2024 APIC president, press release, January 25, 2025]

Additional findings from the study include:

•Cleaning with a disinfectant and brush reduced viral contamination on the toilet by more than 99.99% and on the brush by 97.64%.

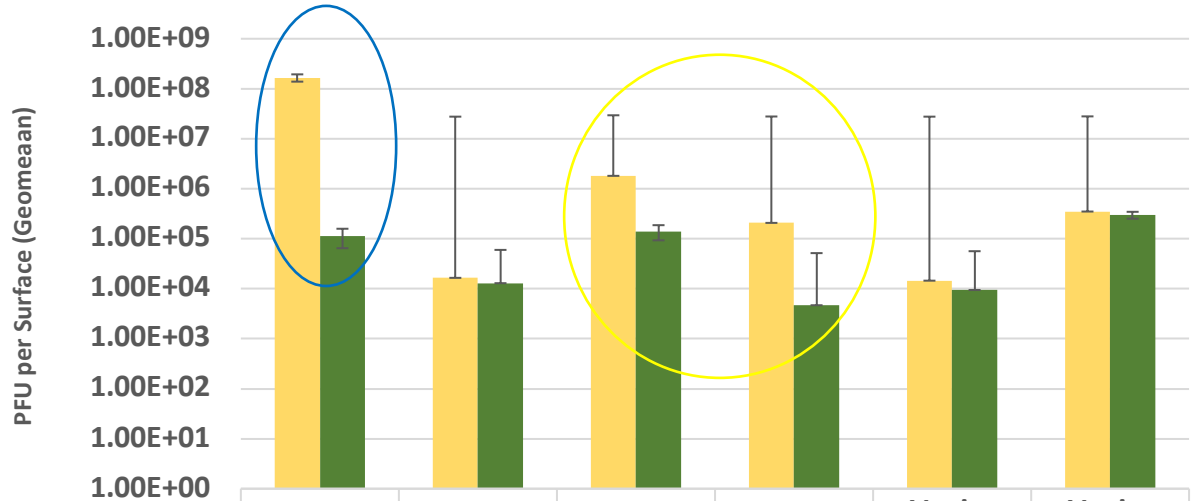
•Closing the toilet lid may alter the direction of the aerosol plume. In this study, floor surfaces in front of and to the left of the toilet were more contaminated after flushing with a closed lid than with an open lid, and floor surfaces to the right of the toilet were less contaminated.

•These findings extend to households. When one member of a household is sick, particularly with an infection that causes gastroenteritis, the regular incorporation of a disinfectant during cleaning or prior to flushing may help reduce the spread of infection to others in the household.

"This study helps establish a clearer understanding of how pathogens spread and the measures we can take to break those chains of transmission," said Tania Bubb, PhD, RN, CIC, FAPIC, 2024 APIC president. "It also underscores the importance of regular disinfection of surfaces in healthcare settings for reducing the spread of viral infections."

Reduction in restroom fomites' cross contamination with and without spraying *Lysol Air Sanitizer* (8s spray) post toilet flushing*

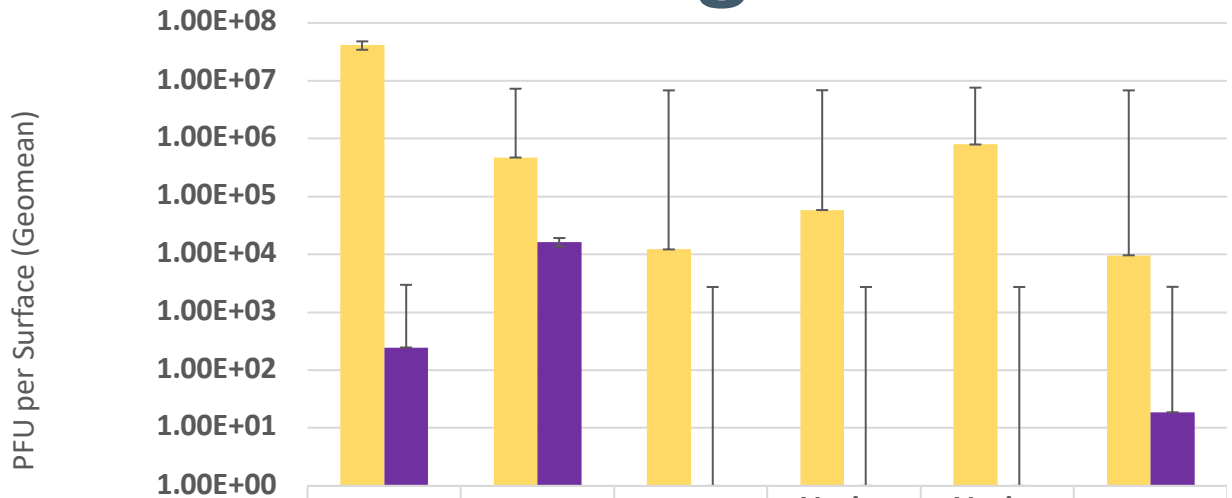




	Toilet Seat	Toilet Handle	Toilet Floor	Corner shelf	Vanity top left side -	Vanity top right Side
No Product	1.66E+08	1.65E+04	1.81E+06	2.08E+05	1.45E+04	3.50E+05
Air Sanitizer 8 sec.	1.12E+05	1.29E+04	1.40E+05	4.67E+03	9.42E+03	2.99E+05

- * The *Air Sanitizing Spray* reduced surface cross-contamination on all the surfaces by 90 to >99.9% except the toilet flush handle and vanity countertop.
- At 8 sec there was a significant difference found between product use and no use on the toilet seat (p = 0.0028).

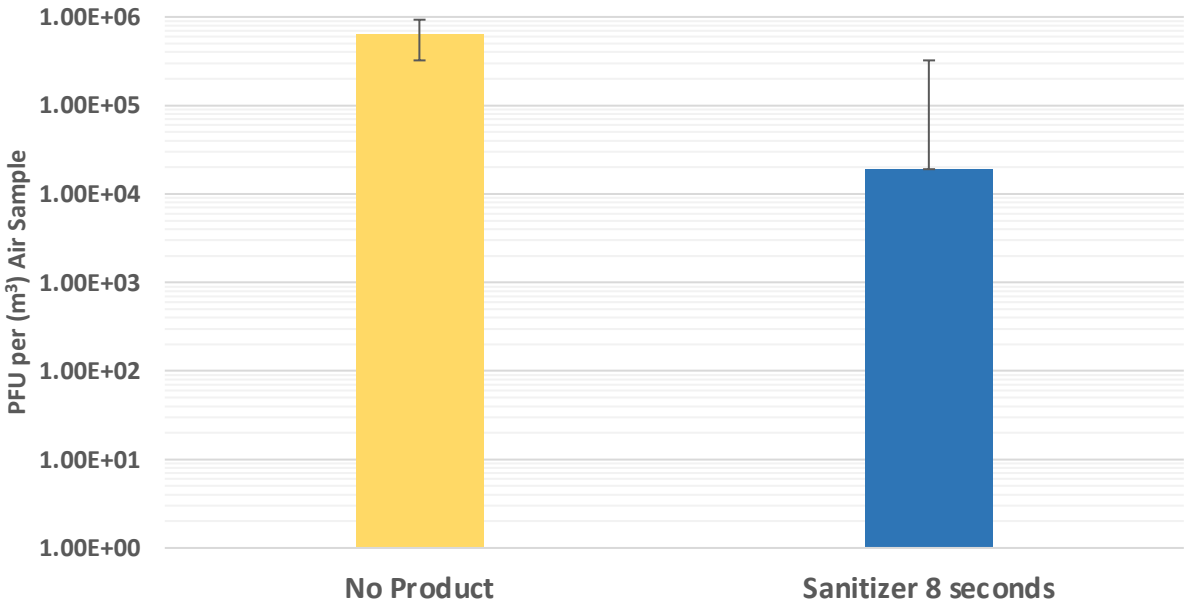
Reduction in restroom fomites' cross-contamination with and without spraying *Lysol Air Sanitizer* (30s spray) post toilet flushing*



	Toilet Seat	Toilet Floor	Corner Shelf	Vanity Top Left	Vanity Top Right	Toilet Handle
No Product	4.12E+07	4.72E+05	1.22E+04	5.84E+04	7.93E+05	9.64E+03
Air Sanitizer 30 sec.	2.46E+02	1.65E+04	1.00E+00	1.00E+00	1.00E+00	1.87E+01

- *Increasing the spray time to 30 seconds resulted in a 97% to >99.9999% reduction of virus on all the surfaces.
- A significant difference was found between no use of product and spraying the air for 30 seconds with the *Air Sanitizing Spray* ($p = 0.054$): corner shelf ($p = 0.0464$) and vanity top left ($p = 0.0483$) and vanity top right ($p = 0.0279$), on the toilet seat ($p = 0.000129$) but not on the toilet floor, and handle ($p = 0.167$).

Reduction of MS2 virus in air by spraying in restroom *Lysol Air Sanitizer* post-toilet flushing (8 seconds spray)*



- **Air Sanitizing Spray* , sampling restroom air post-toilet flushing using the Anderson Air Sampler demonstrated that infectious virus was reduced by almost 99% in the air.

Summary/Conclusions

- Indoor air is a significant vehicle for pathogen dissemination, plays role in establishing air-surfaces-air nexus highlighting the role of holistic approach for risk mitigation.
- Standardized aerobiology protocols (bacteria & viruses) have been developed to study survival & inactivation of airborne pathogens.
- Lysol Air Sanitizer (LAS) reduced airborne bacteria and viruses > 99.9% when tested against experimentally aerosolized bacteria and viruses.
- Application post-toilet flushing in American restroom, indicated that use of LAS causes a reduction in cross-contamination of toilet seat and vanity top including air space when LAS was applied post-flushing toilet in the restroom.



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