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Aerosol Generation by Modern Flush Toilets

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Abstract

A microbe-contaminated toilet will produce bioaerosols when flushed. We assessed toilet plume aerosol from high efficiency (HET), pressure-assisted high efficiency (PAT), and flushometer (FOM) toilets with similar bowl water and flush volumes. Total and droplet nuclei “bioaerosols” were assessed. Monodisperse 0.25–1.9- μm fluorescent microspheres served as microbe surrogates in separate trials in a mockup 5 m³ water closet (WC). Bowl water seeding was approximately 10¹² particles/mL. Droplet nuclei were sampled onto 0.2- μm pore size mixed cellulose ester filters beginning 15 min after the flush using open-face cassettes mounted on the WC walls. Pre- and postflush bowl water concentrations were measured. Filter particle counts were analyzed via fluorescent microscopy. Bowl headspace droplet count size distributions were bimodal and similar for all toilet types and flush conditions, with 95% of droplets <2 μm diameter and >99% <5 μm . Up to 145,000 droplets were produced per flush, with the high-energy flushometer producing over three times as many as the lower energy PAT and over 12 times as many as the lowest energy HET despite similar flush volumes. The mean numbers of fluorescent droplet nuclei particles aerosolized and remaining airborne also increased with flush energy. Fluorescent droplet nuclei per flush decreased with increasing particle size. These findings suggest two concurrent aerosolization mechanisms—splashing for large droplets and bubble bursting for the fine droplets that form droplet nuclei.

INTRODUCTION AND BACKGROUND

The potential for airborne transmission of sewage-related infectious disease was experimentally demonstrated over 100 years ago by Horrocks (1907), who found that sewage flowing smoothly in pipes as well as bursting bubbles in sewage would produce airborne microbes that could be transported substantial distances in the sewer system air while remaining viable. More recently, airborne transport of the SARS coronavirus by such sewage-related bioaerosols was proposed as the likely disease transmission mode in the 2003 SARS outbreak at the Amoy Gardens apartment complex in Hong Kong (Yu et al.

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2004; Hong Kong Special Administrative Unit Department of Health 2011). Production of both large droplet and droplet nuclei bioaerosols during toilet flushing has been shown for a variety of toilet types and microorganisms (Jessen 1955; Darlow and Bale 1959; Bound and Atkinson 1966; Gerba et al. 1975; Scott and Bloomfield 1985; Yahya et al. 1992; Barker and Bloomfield 2000; Barker and Jones 2005). As initially defined by Wells (1934), droplet nuclei are the residuals of larger droplets whose water content has largely or completely evaporated; they are of sufficiently small size that they will not settle on surfaces due to the force of gravity but rather will remain airborne and be carried on air currents. Large droplet contamination of toilet seats and lids, the surrounding floors, and nearby surfaces (Jessen 1955; Darlow and Bale 1959; Newsom 1972; Gerba et al. 1975; Yahya et al. 1992; Barker and Jones 2005) is a well-recognized contact transmission risk in health care and other facilities (Sehulster and Chinn 2003). In contrast, the potential for airborne transmission of infectious disease via “toilet plume” droplet nuclei bioaerosols has not been generally acknowledged. This is perhaps due to the difficulty in distinguishing epidemiologically between contact transmission that may have occurred via contact with large droplets or contaminated surfaces as opposed to airborne transmission that may have occurred via inhalation of droplet nuclei.

Experimental work has clearly shown that droplet nuclei toilet plume aerosols are capable of entraining microorganisms as large as bacteria (Barker and Jones 2005), can migrate well away from the toilet (Jessen 1955; Darlow and Bale 1959; Barker and Jones 2005), and can remain viable for extended periods while airborne (Jessen 1955; Gerba et al. 1975; Barker and Jones 2005). Furthermore, sequential flushes following an initial toilet contamination continue to produce bioaerosols (Barker and Jones 2005; Darlow and Bale 1959; Yahya et al. 1992). Viruses may be particularly difficult to clear from the toilet; Yahya et al. (1992) found that viral bioaerosols were still being produced even after seven flushes after contamination. These studies suggest a possible role of biological films as a reservoir and the potential for multiple bioaerosol generation events due to a single contamination.

Both the vomit and feces of some infected persons may contain extremely high pathogen loads—concentrations of 10^5 – 10^9 *Shigella* (Thompson 1955), 10^4 – 10^8 *Salmonella* (Thompson 1955), and 10^8 – 10^9 norovirus (Atmar et al. 2008) per gram of stool and at least 10^6 norovirus per milliliter of vomit (Caul 1994) have been reported. Some fraction of the aerosol droplets produced when a contaminated toilet is flushed may be expected to contain these microbes (Raabe 1968). Whether toilet bioaerosol droplets will deposit on nearby surfaces or evaporate to form droplet nuclei that move with air currents depends primarily on the initial droplet size and the initial vertical distance from a deposition surface (Wells 1934; Xie et al. 2007); for a given situation droplets smaller than a “critical size” will evaporate to droplet nuclei before reaching the surface, whereas larger droplets will not. The critical droplet size is determined by the droplet evaporation rate and the droplet's settling velocity, which are complex interrelated functions dependent on environmental conditions (Hinds 1999; Xie et al. 2007). To date no studies have reported the initial droplet size distribution of toilet plume aerosols or how the size distribution and aerosol concentration vary across toilet designs and operating modes. Therefore, it has not been possible to estimate the fraction of total aerosol that may produce potentially infectious droplet nuclei

bioaerosols, or the concentrations of droplet nuclei that might result, during toilet flushing. The purpose of the present work was to characterize the initial droplet size distribution immediately after a toilet flush and the droplet nuclei aerosol generation rate for a range of toilet types.

MATERIALS AND METHODS

Subject Toilets

Toilets are sold in a wide variety of designs, and the most common types may be conveniently classed as gravity-flow, pressure-assisted gravity flow, or pressure-valve (commercially termed “flushometer”) systems. Gravity flow is the type typically found in residences, consisting of a base containing the toilet bowl and a close-coupled or integral water tank (also termed a “cistern”). When the flush lever is activated, a valve in the tank bottom opens to allow water to flow under gravity into the toilet bowl, initiating the flush. In contrast, the flushometer toilets that are often seen in public restrooms consist of a base but no tank, with the building water supply attached directly to a flush valve via a 1-inch diameter pipe that delivers a vigorous water flow to the toilet when activated by a manual flush lever or automatic flush sensor. A recent innovation that provides the vigorous flush of a flushometer system without the requirement for a high flow capacity water connection is the pressure-assisted toilet. This toilet has the appearance of a typical gravity-flow toilet in that it has a tank attached to the base; however, a compressed air bladder pressure vessel inside the tank provides stored energy to drive the water flow and thereby simulate the flushometer system.

Prior to the mid-1990s, U.S. toilets typically had flush volumes of approximately 11 to 13 liters per flush (Lpf) (3 to 3.5 gallons per flush [gpf]), but since the enactment of the Federal Energy Policy Act of 1992 (FEPA) the maximum flush volume allowed for toilets sold in the United States is 6 Lpf (1.6 gpf) (United States Congress 1992). Furthermore, water conservation concerns have prompted greater marketing of high efficiency toilets, or HETs. The U.S. Environmental Protection Agency’s “WaterSense” specification limits HETs to no more than 80% of the 6 Lpf value, or 4.8 Lpf (1.28 gpf) (United States Environmental Protection Agency 2011). Another water-saving innovation was the introduction of dual-flush toilets providing for user selection of a lower or higher flush volume, depending on whether liquid only or solids were being flushed. The first dual-flush toilets were invented in Australia 30 years ago and have long been in widespread use there and in Europe but have only recently been widely marketed in the United States. The lower flush volume on these systems is approximately 3.8 Lpf (1 gpf).

Four toilet types were selected for this study: (1) a pre-FEPA gravity flow toilet providing 13.3 Lpf (3.5 gpf), (2) a dual-flush HET providing either 3.8 or 4.9 Lpf (1.0 or 1.3 gpf), (3) a dual-flush pressure-assisted gravity flow toilet (PAT) providing approximately 4.2 or 4.9 Lpf (1.1 or 1.3 gpf) (Figure 1), and (4) a flushometer (FOM) toilet providing approximately 5.3 Lpf (1.4 gpf). Previous investigators have noted greater aerosol production with higher flush energies (Bound and Atkinson 1966; Jessen 1955), as intuition might suggest, and the three post-FEPA toilet types were selected because their different operating modes provide different degrees of flush energy and associated bowl water agitation during flushing.

All four toilets were of the siphonic type, in which flush water enters the bowl bottom as a submerged jet directed toward the S-shaped outlet channel or “trapway” entry to induce a water flow that fills the trapway and results in a siphon effect, which then empties the bowl contents (Figure 2) (Blair 2000). Air entering the trapway when the bowl water level falls below the top of the inlet breaks the siphon and some volume of the flushed water drains back into the bowl. A secondary flush water flow passes through perforations spaced around the underside of the bowl rim and washes down the bowl walls during the flush. The jets may be angled to induce a swirling in the bowl water. The three post-FEPA toilet models had been evaluated under the Maximum Performance (MaP) Program, a joint US–Canada effort to test the clearance performance of toilets using a standard protocol, and achieved the highest MaP clearance performance rating (Gauley and Koeller 2009). The pre-FEPA toilet was selected solely on its availability in good condition from a recycling center and was thoroughly cleaned and fitted with new flush and fill valves before use.

Water Closet Apparatus

The toilets were mounted in a simulated “powder room” size water closet (WC) that was $5 \times 5 \times 7$ feet (ft) ($152 \times 152 \times 213$ centimeters [cm]) in interior dimension, for a volume of 175 cubic feet (ft³) (5 m^3) (Figure 3). The WC was constructed on stilts so that the toilet flush outfall could be captured for volume measurement. The walls and ceiling were of painted gypsum board, and the floor was covered with sheet linoleum. Seals and clamps around a 3×6 ft (91×183 cm) clear plastic access door provided an airtight enclosure. A high-frequency particulate air (HEPA)-filtered air supply, HEPA-filtered exhaust, and blast gate flow damper allowed the WC to be purged under slight positive differential pressure at an effective ventilation rate of approximately 18 air changes per hour (ACH) with particle-free air prior to toilet flushing. Complete clearance of background particles at least as small as 0.3 micrometers (μm) was verified using a Grimm Model 1.108 aerosol spectrometer (Grimm Aerosol Technik GmbH, Ainring, Germany). The spectrometer was placed inside the WC with the digital concentration readout visible through the clear WC door and operated continuously during the purging period. The logged concentration data were subsequently downloaded and analyzed to determine the effective ventilation rate. Air sampling ports were installed at heights of 22.5 and 64 inches (57 and 163 cm) on the two side walls (Figure 3). These ports were connected to air sampling pumps located outside the WC for monitoring and adjustment during air sampling. Pump flows were calibrated at 1.5 liters per minute (Lpm) using a bubble tube primary standard. A glove port in the rear wall allowed the toilet to be flushed from outside the WC.

Potential interferences by naturally occurring fluorescent particles that might be present in the mains water supply were avoided by filtering the water used for toilet flushing. For the HET and PAT toilets, it was sufficient to simply insert the particle filter into the 1/2-inch mains supply line. The FOM cannot operate from a 1/2-inch supply line and required a custom built two-tank water supply system as shown schematically in Figure 4. The first tank (Well-X-Trol WX-202, Amtrol, West Warwick, RI, USA) received mains water from a booster pump (Model MQ, Grundfos, Olathe, KS, USA) that maintained the tank water pressure between 55 and 70 psi. The water passed from the first tank through a high-efficiency particle filter (Model GE GXULQ, General Electric Co., Fairfield, CT, USA) to

the second tank, which provided particle-free water to the FOM toilet via a 1-inch supply line. FOM type toilets require a 1-inch supply line to satisfy their high flush water flow rate. Samples of the filtered water supplies verified that they were particle free.

Droplet Size Distribution and Generation Rate Measurements

Accurate measurement of the initial droplet size distribution of toilet flush aerosol is challenging due to rapid droplet evaporation under typical humidity conditions. However, the rate of evaporation may be slowed by providing a high relative humidity environment, and this was achieved by covering the toilet bowl with a plastic plate as shown in Figure 5. The sample inlet probe of a Grimm aerosol spectrometer was inserted through the plate and positioned several inches above the bowl water surface. The probe of a digital hygrometer (Traceable[®], Control Company, Friendswood, TX, USA) was also inserted through the plastic plate. The air in the enclosed bowl quickly reached saturation. After starting the aerosol spectrometer, the WC door was replaced and the WC was purged for at least 45 minutes at 18 ACH effective ventilation rate to ensure removal of all background particles. The purge air was then turned off and the toilet flushed. After a 15-min postflush sampling period, the HEPA was restarted to prepare for another measurement. The spectrometer operated continuously, providing particle concentration measurements in 15 particle size ranges spanning 0.3 to 20 μm . After each trial, the spectrometer was removed from the WC for data download and analysis.

Droplet Nuclei Bioaerosol Generation

Microbial bioaerosol generation by the three post-FEPA toilets was simulated using monodisperse suspensions of 0.25- or 0.30- μm -diameter green (0.25 μm) or red (0.30 μm) fluorescent polymer microspheres (Cat. No. G250 or R300, Thermo Scientific Inc., Fremont, CA, USA) after the method of Johnson and Lynch (2008).

A 1-mL aliquot of the 1% by volume source suspension was mixed with the toilet bowl water and a water sample collected. Delrin[®] acetal resin open-face 25-mm air sampling cassettes (Cat. No. 225-1107, SKC, Eighty Four, PA, USA) containing 0.2- μm pore size mixed cellulose ester (MCE) filters (Advantec Cat. No. A020A025A, Toyo Roshi Kaisha Ltd., Japan) were attached at each of the four sampling ports. The cassettes and o-rings were rinsed in acetone and air dried before assembly to dissolve away any polymer microsphere contamination. The Viton o-rings provided with the cassettes were replaced with perfluoroacetate (PFA or Teflon[®]) coated silicon core o-rings (PSP Inc., Denver, CO, USA) due to Viton's incompatibility with acetone and because the Viton o-rings tended to deform when the cassette cap was tightened, wrinkling the filter. Purging for 45 minutes was followed by a 5-min pause to allow damping of WC air currents before flushing the toilet. Because only the simulated droplet nuclei bioaerosols were of interest, another 5-min pause after the flush allowed time for large droplets to settle out on the floor, on toilet surfaces, or back into the bowl water. The WC air was then mixed with a small fan for 5 min to disperse the remaining droplet nuclei aerosols more evenly throughout the WC volume before starting the air sampling pumps. Pump flow rates were closely monitored and adjusted as necessary to maintain constant 1.5 Lpm flow during a 30-min sampling period, providing a 45-L air sample volume. The 0.25- μm minimum particle size was chosen because this was

the smallest fluorescent particle that could be accurately counted; larger particles are easier to count, but there was concern that they would be less likely to be aerosolized as described by Raabe (1968) and would also be less representative of microbes of virus size. The volume of captured flush water was measured, and a postflush bowl water sample was collected after the air sampling was completed.

After sampling cassettes were collected and the filters removed and mounted on oversized 75×38 mm microscope slides for top-illumination viewing and particle counting using a Nikon Model Eclipse 80i fluorescence microscope fitted with a Prior Optiscan 3-axis motorized stage, 20x planapochromat objective, Hamamatsu ORCA-Flash 2.89 camera, and NIS Elements software (AR 2.10, Build 215) (all from Nikon Instruments, Melville, NY, USA). Air samples were imaged automatically through the use of the Elements software. A 1.16×1.16 cm section of the filter (centered roughly on the filter centroid) was processed into 1200 individual images. Each of these images was scanned automatically for the presence of spheres employing user-selected criteria (area, circularity, mean intensity). Particles selected by the software as spheres were individually confirmed by reviewing selected images. The sum of the four filters' particle counts divided by the air volume sampled through that area of the filter (approximately 25% of the total air flow) provided an estimate of the mean WC air concentration (in particles per cubic meter). This was multiplied by the WC volume to provide an estimate of the number of simulated droplet nuclei "bioaerosols" generated during the flush. Because this was a counting method spanning the entire surface of the area selected for counting, the limit of detection of the method was 1 particle in 47.9 L of air (26.6% of the total sampled air) filtered through an area of 4.41 cm^2 (26.6% of the total filter area, i.e., 1.1025 cm^2 on each of 4 filters of 4.15 cm^2 filtration area), or 20.9 particles per cubic meter.

Control trials for the aerosolization experiments were conducted in the same manner as the data trials except that the toilet was not flushed. That is, all aspects of the experimental procedure were followed except the toilet was not actually flushed. These trials assessed the potential for air filter contamination during any step in the procedure from mounting the filters in the cassettes to analyzing the filters under the microscope.

An aliquot of each water sample, diluted as necessary, was filtered through the same type of $0.2\text{-}\mu\text{m}$ MCE filter used for the air sampling, and the filter was then mounted on a slide for counting. Aliquots of filtered toilet supply water and dilution water control samples were also filtered for counting. Water samples were generally processed by manual counts using a random field technique rather than automated counts due to their high areal density (Johnson and Lynch, 2008). For most samples (those less densely populated), 50 fields were counted at $200\times$ total magnification using an optical grid while for the densest samples (i.e., the preflush samples) ten fields were counted at $400\times$ magnification. Very low density water samples were counted automatically as described above using a 0.503×0.503 cm field divided into 275 images. The particle count divided by the aliquot volume and multiplied by the dilution factor provided an estimate of the suspension concentration in each water sample. The ratio of pre- to postflush concentrations was a measure of toilet clearance. The product of the difference in pre- and postflush concentrations and the bowl water volume or the preflush concentration and the bowl water volume provided essentially the same

estimate of the number of fluorescent particles flushed, because clearances were usually on the order of 3 logs or 99.9%. The ratio of the number of fluorescent droplet nuclei to the number of fluorescent particles present preflush was used as the measure of droplet nuclei aerosol generation rate, G . This was scaled up via a 10^8 multiplier to give particles generated per 100 million particles present preflush.

Toilet bowl clearance and droplet nuclei generation rates were assessed for each of five flush conditions: low- and high-volume HET, low- and high-volume PAT, and FOM. Each of the assessments was conducted at least three times.

The influence of suspension particle size on droplet nuclei aerosol production was assessed in the FOM toilet using 0.25, 0.50, 1.0, and 1.9- μm -diameter fluorescent microspheres (Thermo Scientific Inc., Fremont, CA, USA). For each of six trials with each size, the toilet was seeded with a 1-mL aliquot of 1% by volume source suspension, so that the preflush bowl water concentrations were the same in terms of volume percent but different in terms of particles per milliliter due to the differences in microsphere volumes.

Statistical Data Analysis

The postflush fluorescent droplet nuclei concentration for each trial was calculated as the mean of the four air sample results for the trial. The mean and standard deviation of the mean (SEM) for each condition (toilet type and flush volume) were calculated from the individual trial means for the condition. Condition means were compared via analysis of variance (ANOVA) followed by a simultaneous pairwise comparisons test to identify statistically significant aerosol droplet count or fluorescent droplet nuclei concentration differences between toilet types and flush conditions. Parametric one-way ANOVA with Tukey's test was used for the droplet count comparisons, but unequal variances as identified by the F-test required a nonparametric Kruskal–Wallis ANOVA with Nemenyi's test for the fluorescent droplet nuclei concentration comparisons. A Type I error rate of $\alpha = 0.05$ was used in all tests.

RESULTS

Droplet Size Distributions and Generation Rates

No droplet spatter was noted on the underside of the plastic cover plate. The aerosol spectrometer recorded particle count concentrations in each of 15 particle size bins ranging from 0.3 to $>20 \mu\text{m}$ every 6 s. The measurement data revealed zero concentrations across size bins after HEPA purging and immediately before the toilet flush. After a flush the instrument usually measured particle counts in nearly all size bins during the first 15 seconds postflush except for the $>20 \mu\text{m}$ counts, which were typically at or very near zero. The counts for particles larger than approximately 5- μm -diameter reached a maximum in the 15–30 s period and declined after the first minute, suggesting removal by gravitational settling or shrinkage to smaller sizes by evaporation. Such shrinkage will occur even under saturation humidity conditions, though more slowly than under drier conditions. In contrast, counts across all of the smaller size bins increased to a peak between 1 and 2 min postflush, then also declined. The increases in the smaller sizes were in far greater numbers than the

reductions in counts for the larger particle sizes and occurred across all size bins, suggesting droplet growth by condensation in the high-humidity bowl environment rather than simply contributions to the smaller size bins by evaporation of larger droplets. As discussed by Hinds (1999) (pp. 288–292), heterogeneous nucleation around soluble condensation nuclei can occur even under unsaturated atmospheric conditions, and the droplets will grow to a maximum size that is determined by the mass of solute in the condensation nucleus and the relative humidity. The filtered flush water used in these experiments was from the building mains supply and had a total dissolved solids concentration of approximately 120 ppm. These naturally occurring dissolved minerals would be expected to act as condensation nuclei, along with those naturally present in the air in spite of HEPA filtration.

Given this dynamic situation, it is possible only to estimate the droplet count size distribution in the bowl airspace immediately after the flush. As a common reference, the particle counts measured approximately 15 s after the flush were used. This allowed time for the spectrometer to detect the larger droplets without allowing extended time for gravitational settling or increases in the smaller droplet counts. A plot of mean droplet size distributions for the four toilet types at 15 s postflush is shown in Figure 6. The droplet size distribution plots were somewhat bimodal and similar across toilet types and across flush conditions within toilet type for the dual-flush toilets. The aerosols appeared to be composed of two particle populations but with over 95% of the particles being less than 2- μm diameter.

The total droplet counts in the air space above the bowl water and below the top of the rim, shown in Table 1, were calculated from the sum of particle concentrations across size bins as measured 15 s postflush and the volume of the air space between the bowl water level and the cover plate placed over the bowl during measurement (10.0, 12.0, 13.0, and 10.8 L for the pre-FEPA, HET, PAT, and FOM, respectively). The calculations assumed a uniform concentration throughout the bowl air space, and though this could not be verified the estimate provides a basis for comparison across toilet types. After verifying homogeneity of variance using an F-test, a one-way ANOVA of the droplet generation rates indicated a statistically significant difference between toilets/flush conditions ($p < 0.0001$), and a Tukey's test (overall $\alpha = 0.05$) further indicated significant differences between the toilets but not between the low and high volume flush conditions for either the HET or PAT. As Table 1 shows, the mean droplet generation rate per liter flushed was highest for the FOM toilet (~25,600) and lowest for the HET (~2100–2200). The mean total number of droplets produced per flush was also greatest for the FOM toilet (~145,000 droplets) and lowest for the HET (~8000–10,000). Notably, the FOM toilet produced 2.7 times as many droplets as the pre-FEPA toilet even though it had less than half (40%) of the pre-FEPA's flushed water volume. It also produced 3.5 times as many droplets as the next highest post-FEPA toilet. This appears to support an association between flush energy and aerosol production.

Droplet Nuclei Bioaerosol Generation Across Toilet Types

All control blank samples were negative for the fluorescent microspheres, demonstrating the absence of background counts in both the air samples and the water samples. Pre- and postflush bowl water samples indicated 3+ log concentration reductions from approximately 10^8 to 10^5 particles/mL, consistent with the findings reported by others for microbes at

similar seeded concentrations (Darlow and Bale 1959; Gerba et al. 1975; Barker and Jones 2005). Droplet nuclei aerosol generation rates for the highest flush volumes of each toilet, as number of “bioaerosol” fluorescent particles produced per 100 million particles present preflush, are presented in Table 2. The means suggested an increasing droplet nuclei aerosolization with increasing flush energy from HET to PAT to FOM, consistent with the differences in flush droplet production shown in Table 1; however, there was considerable flush-to-flush variation in the number of fluorescent droplet nuclei particles aerosolized. Furthermore, an F-test indicated nonhomogeneous variance across conditions. The nonparametric Kruskal–Wallis ANOVA was, therefore, used to compare conditions and failed to yield a significance result. The mean generation rates as particles aerosolized per 100 million particles present in the bowl water preflush were, therefore, also not significantly different due to the similar seed concentrations and bowl water capacities.

Droplet Nuclei Aerosol Production for Larger Particle Sizes

Airborne particle production and generation rates for the 0.25, 0.50, 1.0, and 1.9 μm microspheres in the FOM toilet are summarized in Table 3. In testing for differences in the means, unequal variances as shown by the F-test required that nonparametric Kruskal–Wallis ANOVA be employed rather than parametric ANOVA. The nonparametric ANOVA yielded a significant result ($p < 0.015$), and a Nemenyi's multiple simultaneous pairwise comparisons test (overall $\alpha = 0.05$) indicated the 0.25- μm airborne concentration to be significantly different from the 1.0- and 1.9- μm concentration but not statistically significantly different from the 0.50- μm concentration. The 0.50-, 1.0-, and 1.9- μm concentrations were also not statistically significantly different from one another. It should be noted that the airborne particle concentrations measured in individual trials of the larger particles, particularly the 1.0 and 1.9 μm particles, were often at the limit of detection of the counting method, i.e., no particles or only 1 particle detected.

DISCUSSION

The three modern (post-FEPA) toilets studied were seen to vary greatly in initial flush droplet production for nearly identical flush volumes. Droplet generation rates per liter of water flushed appeared consistent across lower and higher volume flush options in the two dual-flush toilets (HET and PAT), and mean droplet generation rates increased with perceived higher flush energy (an admittedly qualitative judgment by the investigators based on sound level and apparent agitation) from HET to PAT to FOM. These results support the conclusion by Jessen and subsequent investigators that flush droplet production increases with increasing flush energy. Flush energy cannot be directly measured, and at any rate is a surrogate descriptor for the degree of agitation the water undergoes during the flush. The effects of agitation on aerosol production from biological suspensions have been studied with regard to aerated activated sludge sewage treatment systems and aerated fermenters. Brandi et al. (2000) and Sanchez-Monedero et al. (2008) noted that mechanical agitation increased bioaerosol production from aerated sewage treatment plants, and Pilacinski et al. (1990) observed an increase in both the number and size of aerosol droplets produced from aerated fermentor broths as the degree of mechanical agitation increased. It seems plausible

that the effects of flush agitation on bioaerosol production from contaminated toilets would be similar, but this is an area requiring further research.

Results for comparisons of fluorescent droplet nuclei particle generation across particle sizes should be viewed with some caution. The very low concentrations observed for the larger particles, particularly the 1.0 and 1.9 μm particles, would be expected to induce volatility in the estimated means and also inflate the variance estimates, reducing the power of statistical tests to detect real differences. Although longer air sampling times or higher air sampling rates would have increased the number of filter counts, this would have been at the risk of oversampling the 175 ft³ (5 m³) WC air volume. Future studies might be designed to constrain the air volume into which the droplet nuclei disperse to provide a more concentrated atmosphere and thereby increase the counts, though it is recognized that this would introduce an additional dimension of artificiality.

Aerosol generation rates expressed as airborne particles produced per 100 million particles present in the bowl preflush were widely different and had no clear pattern except that the generation rate value for the 1.9 μm particles was an order of magnitude larger than the other generation rates. However, in other experiments not reported here involving a much wider range of preflush particle loadings (5–6 logs), we found that aerosol generation is not proportional to the preflush bowl loading. This observation was in agreement with the findings of others in sequential flush experiments showing that airborne particle concentrations do not vary in proportion to the bowl water particle concentration (Darlow and Bale 1959; Newsom 1972; Gerba et al. 1975; Barker and Jones 2005). The measure, though we present it for completeness, appears to have little utility in characterizing flush aerosol generation.

The observation that flush aerosol generation is not proportional to the preflush bowl water particle loading seems contrary to logic if one assumes that the aerosolization mechanism is related to simple splashing. Such an assumption appears to be supported by observations that aerosol production seems to increase with increasing flush energy. Nevertheless, the data show the splash generation model does not hold up, at least for droplet nuclei aerosol production, and we therefore propose a different mechanism be considered. The somewhat bimodal droplet size distributions of the flush droplets were consistent with an aerosol generation mechanism that includes bubble bursting. Bubbles bursting at the liquid surface will produce two populations of droplets: fine “film droplets” and larger “jet droplets” (Tomaides and Whitby 1976). Film droplets are produced by the breakup of the bubble film layer as the bubble rises through the liquid surface and range from submicrometer size to approximately 20 μm (Garner et al. 1954; Blanchard 1963; Cipriano and Blanchard 1981), whereas jet droplets are produced at the tip of a jet of liquid projected upward from the center of the bubble depression in the liquid surface (MacIntyre 1972). Jet droplets are typically an order of magnitude larger than film droplets (Newitt et al. 1954) and may be projected tens of centimeters above the water surface at initial ejection velocities of up to several meters per second (Gunther et al. 2003). The absence of visible spatter on the toilet cover plate and particle counts in the >20- μm Grimm spectrometer data from size distribution measurements suggest that jet drops were not a substantial constituent of the toilet flush aerosols. The number of film droplets decreases with decreasing bubble size

(Blanchard 1963; Day 1964), from as many as 1000 film droplets for a 6-mm-diameter bubble (Blanchard and Syzdek 1982) to very few film droplets for bubbles less than approximately 1 mm (Gunther et al. 2003). In contrast, the number of jet droplets increases with decreasing bubble size, with bubbles less than 0.3-mm-diameter producing five or more jet droplets (Blanchard and Syzdek 1982). Bubbles in the flush water may be produced when air is entrained in agitated water, as by a submerged jet or during a turbulent flush, in a manner similar to air entrainment in seawater by wave action and whitecaps (see, e.g., the discussion by Kerman 1986). The resulting bubble burst aerosol would thus be expected to be somewhat bimodal, as was seen by Baron and Willeke (1986) when they measured droplet aerosols above heated whirlpools agitated with jets. Bubble-burst production of aerosols, including bioaerosols, has been studied above bodies of sea water and fresh water (Blanchard and Syzdek 1970, 1972; Baylor et al. 1977; Blanchard and Syzdek 1978; Blanchard et al. 2011), and to some extent in whirlpool baths (Baron and Willeke 1986), but to date has not been explored as a mechanism for toilet aerosol production. Since bioaerosol of respirable size produced by bubble bursting in a whirlpool contaminated with *Legionella pneumophila* has been associated with an outbreak of Pontiac Fever (Mangione et al. 1985), and a number of studies have demonstrated the potential for toilets to produce bioaerosols during flushing (reviewed briefly above and in detail by Johnson et al. 2013). We propose bubble bursting as a likely mechanism for droplet nuclei bioaerosol production from pathogen-contaminated toilets and believe this to be a logical area for further research.

Our results and those of previous investigators present a consistent body of data that demonstrates the potential for generation of infectious droplet nuclei bioaerosols when a contaminated toilet is flushed. Pathogens including *Shigella*, *E. coli*, *Clostridium difficile*, SARS coronavirus, and norovirus (Thomson 1955; Caul 1994; Atmar et al. 2008) can be present in vomit or stools of infected persons and can survive on surfaces for weeks or even months (Kramer et al. 2006). As has been shown, hundreds to thousands of potentially infectious bioaerosol particles, capable of remaining airborne for extended periods and migrating with air currents, may be generated in a single flush of a toilet contaminated with these organisms. Whether a subsequent bioaerosol exposure results in disease would of course depend on the organism's viability under existing environmental conditions, the number of organisms inhaled and their virulence, and the exposed person's immune status among other factors (Cox 1987), but it is difficult to imagine that such transmission is not occurring. Separating the incidence of disease transmission by contact and droplet routes from that by the airborne route is a challenge that remains to be met.

A limitation of this study was the small number of replicate trials per experimental condition (3 to 6), which likely provided inadequate statistical power in some data analyses due to the sometimes substantial within-condition variability in aerosol production. Future work should allow for larger sample sizes as determined by power calculations using these observed variances. A second limitation was the data volatility inherent in measurements of very low airborne particle concentrations, as seen with larger particle sizes, which limited the interpretability of these data. A third limitation, from the risk assessment perspective, was the use of monodisperse fluorescent polymer particles rather than actual microbes. However, the purpose of this work was to characterize the initial droplet size distribution immediately

after a toilet flush and the potential for droplet nuclei aerosol generation for a range of toilet types, with an eye toward elucidating the toilet design factors governing aerosol production, rather than to provide data directly applicable in risk assessment. Further work using specific microbes of interest is required to better estimate airborne infectious disease transmission risk.

SUMMARY AND CONCLUSIONS

The WC research platform and analytical method were effective in assessing toilet flush droplet nuclei generation. For all toilet types examined, toilet flush aerosols were highly concentrated and the initial droplet size distributions were somewhat bimodal. The number of droplets appeared to increase with increasing flush energy, with statistically significant differences in droplet production across toilets. The FOM type toilet, which is ubiquitous in business, health care, and other public environments, produced far more droplets than the other toilets, both as total droplets and droplets per liter flushed.

All three modern toilets produced hundreds to thousands of droplet nuclei “bioaerosol” particles with each flush, though there were differences across toilets in droplet nuclei aerosol production as measured by mean airborne concentration produced. Aerosol production appeared to increase with increasing flush energy, in agreement with the droplet aerosol measurements immediately postflush. However, aerosol production was not proportional to preflush bowl water particle loading as would be expected for an assumed splash aerosolization model. An alternative bubble burst aerosolization mechanism is proposed that may be responsible for these results and similar results by other investigators.

There were also statistically significant differences in droplet nuclei production for different suspension particle sizes, with mean airborne concentrations decreasing as particle size increased. The mean air concentrations (or number of nuclei produced per flush) decreased with microsphere particle size.

These results provide additional support for concerns that flush toilets could play a role in airborne transmission of infectious disease via droplet nuclei bioaerosols. Further research is needed to separate the incidence of toilet flush aerosol-related airborne infectious disease transmission, if it exists as seems likely, from transmission by other routes.

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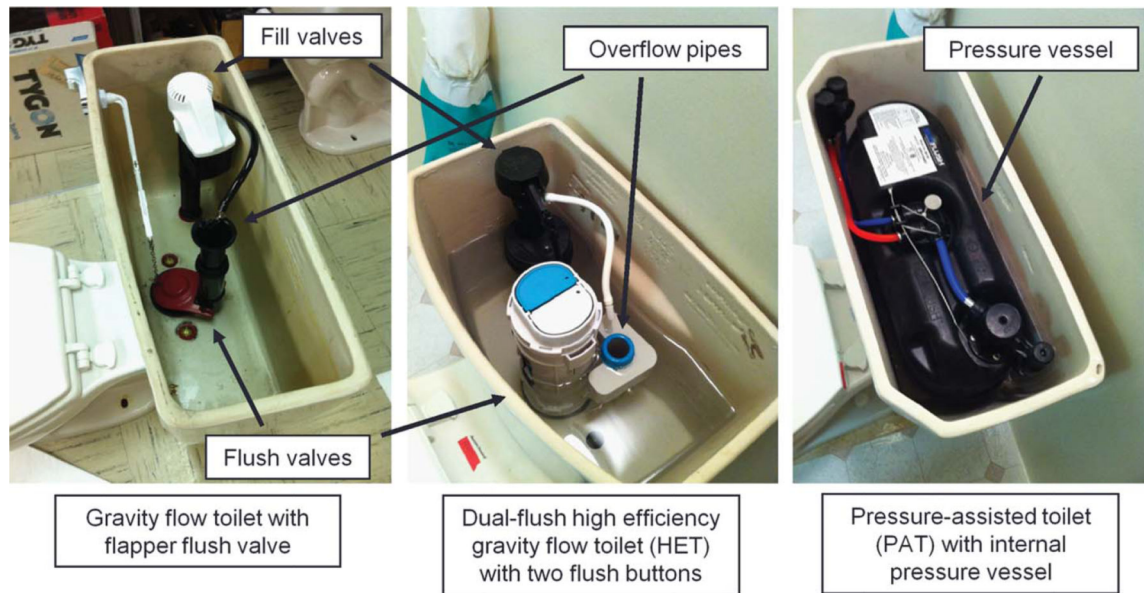


FIG. 1. In-tank flush mechanisms of the pre-FEPA, dual-flush high efficiency, and dual-flush pressure-assisted gravity flow toilets used in this work. (Color figure available online.)



FIG. 2. Typical water flow in the base of a siphonic toilet. (Color figure available online.)

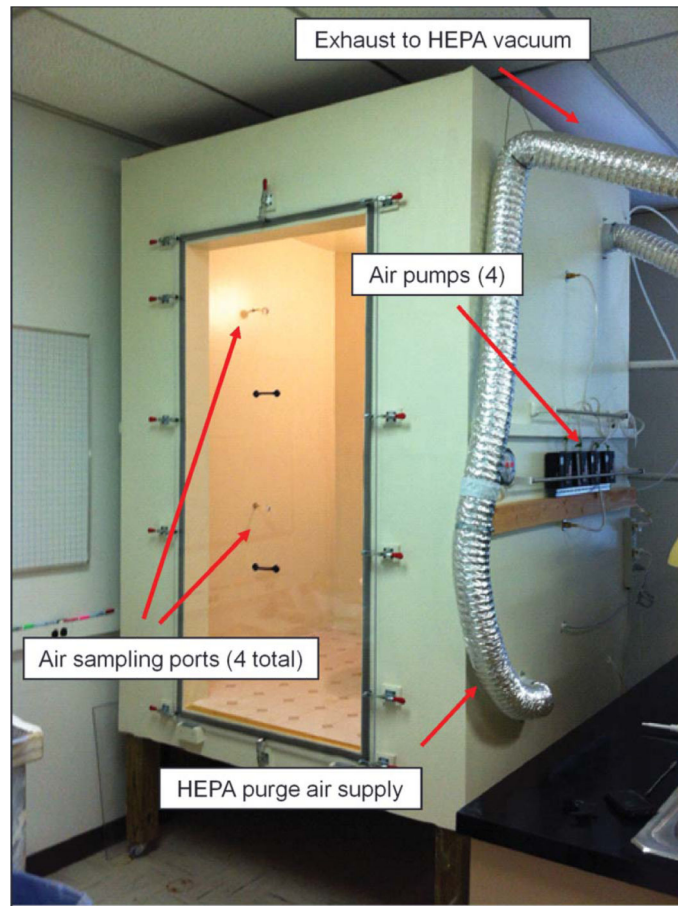


FIG. 3. Elevated, controlled-environment water closet (WC) with airtight door, balanced HEPA-filtered flush air and exhaust, air sampling ports on side walls, and externally-mounted air sampling pumps. (Color figure available online.)

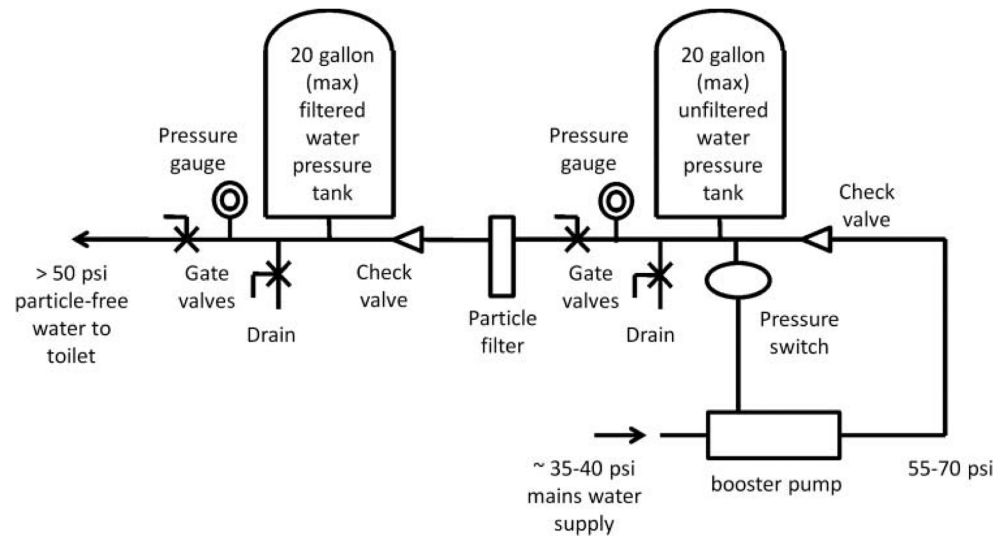


FIG. 4. Schematic of the flushometer flush water supply system incorporating a fine particle filter and pressure boost.

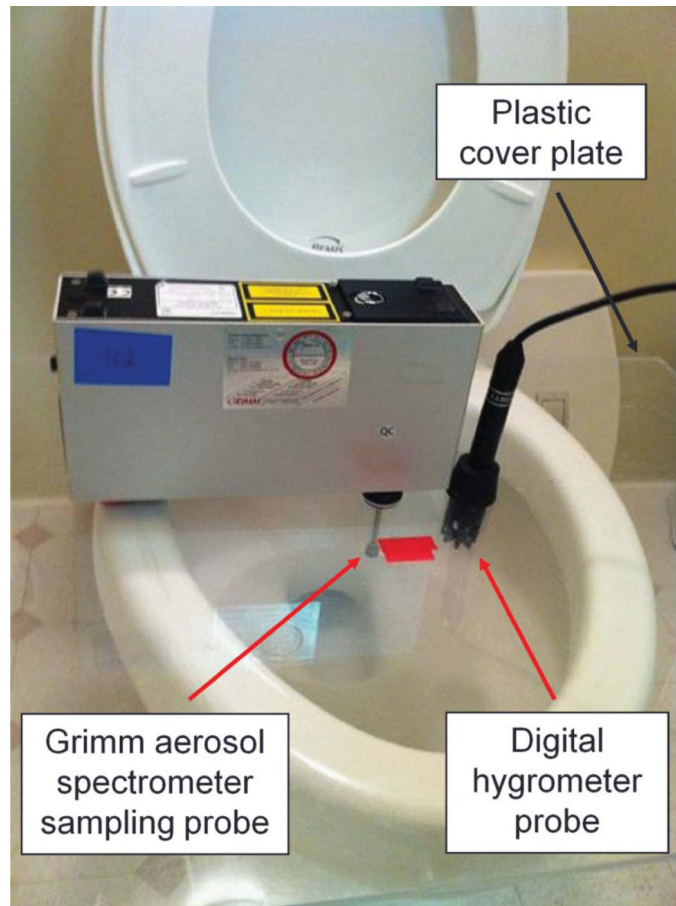


FIG. 5. Droplet size distribution measurement. A Grimm aerosol spectrometer sampled the droplet aerosol from the enclosed volume immediately above the bowl water surface at relative humidity exceeding 99%. (Color figure available online.)

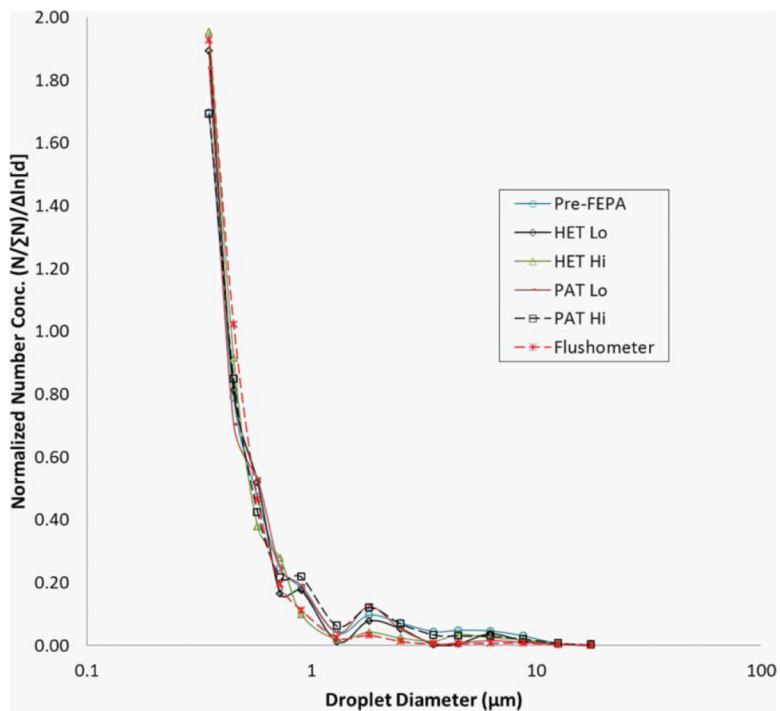


FIG. 6. Normalized frequency plot of the droplet particle size distributions measured in the toilet bowl at 15 s postflush for the toilet types and flush conditions studied: pre-FEPA gravity flow, dual-flush volume high efficiency (HET), dual-flush volume pressure-assisted (PAT), and flushometer (FOM). Each curve represents the mean of three trials. (Color figure available online.)

TABLE 1

Flush droplet generation

Toilet type and flush condition	Total droplets produced *	Droplet generation rate * (droplets/L flushed)
Pre-FEPA gravity flow (13.3 Lpf)	54,363 (6764)	4,165 (570)
HET, low-volume flush (3.8 Lpf)	8220 (616)	2,237 (158)
HET, high-volume flush (4.9 Lpf)	10,620 (1060)	2,100 (189)
PAT, low-volume flush (4.2 Lpf)	25,762 (1855)	6,546 (545)
PAT, high-volume flush (4.9 Lpf)	40,521 (1955)	8,001 (308)
FOM (5.3 Lpf)	145,214 (8325)	25,663 (1525)

Each toilet type was statistically significantly different from the others, but there was no statistically significant difference between flush conditions for a given dual-flush toilet (HET or PAT).

* Mean (standard error), $n = 3$ trials per condition.

TABLE 2

Fluorescent droplet nuclei (DN) generation by toilets with similar flush volumes and seed concentrations, simulated with 0.25- or 0.30- μm diameter polymer microspheres

Toilet type and flush	Size (μm)	DN particles	Mean (SEM [*]) DN particles	Mean (SEM [*]) air conc. (DN/m ³)	Number of particles present preflush	DN generation rate (DN produced per 100 million present preflush)	Mean (SEM [*]) DN generation rate (DN produced per 100 million present preflush)
HET (4.9 Lpf)	0.30	1512	1164 (229)	235 (46)	1.52×10^{11}	0.995	0.256 (0.151)
	0.30	1096			1.23×10^{12}	0.089	
	0.30	1282			6.68×10^{11}	0.186	
	0.25	1893			9.92×10^{11}	0.191	
	0.25	230			1.44×10^{12}	0.016	
	0.25	973			1.74×10^{12}	0.056	
PAT (4.9 Lpf)	0.25	148	1253 (734)	253 (148)	1.13×10^{12}	0.013	0.072 (0.037)
	0.25	2643			1.89×10^{12}	0.140	
	0.25	968			1.54×10^{12}	0.063	
FOM (5.3 Lpf)	0.25	3432	2539 (218)	513 (44)	1.33×10^{12}	0.259	0.233 (0.028)
	0.25	2252			1.20×10^{12}	0.188	
	0.25	2808			1.79×10^{12}	0.157	
	0.25	1900			7.75×10^{11}	0.245	
	0.25	2285			6.47×10^{11}	0.353	
	0.25	2560			1.29×10^{12}	0.198	

* SEM = standard error of the mean.

TABLE 3

Droplet nuclei concentrations for various particle sizes flushed using the flushometer toilet ($n = 6$ trials per condition)

Particle size (μm)	Mean (SEM [*]) bowl water preflush concentration (particles/mL)	Mean (SEM [*]) airborne particle concentration produced (particles/m ³)	Mean (SEM [*]) airborne particle generation rate (airborne particles produced per 100 million particles present in the bowl preflush)
0.25	5.073×10^8 (7.427×10^7)	512.6 (44.0)	0.235 (0.029)
0.50	1.679×10^8 (2.028×10^7)	65.8 (18.4)	0.092 (0.034)
1.0	2.035×10^7 (1.968×10^6)	10.3 (4.6)	0.113 (0.052)
1.9	1.660×10^6 (7.009×10^5)	20.1 (9.9)	8.27 (6.86)

The 0.25- μm concentration was significantly different from the 1.0- and 1.9- μm concentration but not different from the 0.50- μm concentration, and the 1.0- and 1.9- μm concentrations were not significantly different from one another. Nemenyi's test with overall $\alpha = 0.05$.

* SEM = standard error of the mean.