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The potential spread of infection caused by aerosol contamination of surfaces after flushing a domestic toilet

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ABSTRACT

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Aims: To determine the level of aerosol formation and fallout within a toilet cubicle after flushing a toilet contaminated with indicator organisms at levels required to mimic pathogen shedding during infectious diarrhoea.

Methods and Results: A semisolid agar carrier containing either *Serratia marcesens* or MS2 bacteriophage was used to contaminate the sidewalls and bowl water of a domestic toilet to mimic the effects of soiling after an episode of acute diarrhoea. Viable counts were used to compare the numbers of *Serratia* adhering to the porcelain surfaces and those present in the bowl water before and after flushing the toilet. Air sampling and settle plates were used to determine the presence of bacteria or virus-laden aerosols within the toilet cubicle. After seeding there was a high level of contamination on the porcelain surfaces both under the rim and on the sides of the bowl. After a single flush there was a reduction of 2·0–3·0 log cycles cm^{-2} for surface attached organisms. The number of micro-organisms in the bowl water was reduced by 2·0–3·0 log cycles ml^{-1} after the first flush and following a second flush, a further reduction of *c.* 2·0 log cycles ml^{-1} was achieved. Micro-organisms in the air were at the highest level immediately after the first flush (mean values, 1370 CFU m^{-3} for *Serratia* and 2420 PFU m^{-3} for MS2 page). Sequential flushing resulted in further distribution of micro-organisms into the air although the numbers declined after each flush. *Serratia* adhering to the sidewalls, as well as free-floating organisms in the toilet water, were responsible for the formation of bacterial aerosols.

Conclusions: Although a single flush reduced the level of micro-organisms in the toilet bowl water when contaminated at concentrations reflecting pathogen shedding, large numbers of micro-organisms persisted on the toilet bowl surface and in the bowl water which were disseminated into the air by further flushes.

Significance and Impact of the Study: Many individuals may be unaware of the risk of air-borne dissemination of microbes when flushing the toilet and the consequent surface contamination that may spread infection within the household, via direct surface-to-hand-to mouth contact. Some enteric viruses could persist in the air after toilet flushing and infection may be acquired after inhalation and swallowing.

Keywords: aerosols, environment, gastroenteritis, infection risk, MS2-bacteriophage, toilet.

INTRODUCTION

Infectious gastroenteritis is caused by a variety of micro-organisms which have the potential to contaminate surfaces in toilets and bathrooms because they are excreted in large numbers during episodes of acute diarrhoea. Flushing the toilet is known to produce aerosols that are capable of causing surface contamination within the toilet and bathroom (Darlow and Bale 1959; Bound and Atkinson 1966; Newsom 1972; Gerba *et al.* 1975). Many enteric pathogens are spread by the faecal–oral route and it has been suggested that the fallout of droplets containing faecal material, after

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flushing the toilet, is an important infection hazard within the bathroom (Hutchinson 1956; Darlow and Bale 1959; Gerba *et al.* 1975).

Viruses are a significant cause of gastroenteritis worldwide and virtually all children aged 3–5 years acquire a rotavirus infection. Individuals with acute diarrhoea may shed $>10^{10}$ infectious rotavirus particles per ml of faeces (Hart and Cunliffe 1999) and toilet flushing could spread aerosols containing the virus onto surfaces in the bathroom. The virus spreads rapidly within families and adults also become infected, although they generally suffer from asymptomatic or mild illness. In the UK, over the last decade the reported incidence of norovirus has increased considerably and it is estimated that at least 3 million cases occur annually (Evans *et al.* 1998; Wheeler *et al.* 1999). The virus produces a rapid onset of diarrhoea and vomiting in both adults and children and large numbers of infectious virus particles are found in both vomit and faeces. The infective dose of both norovirus and rotavirus is presumed to be as low as 10–100 virus particles (LeBaron *et al.* 1990) which undoubtedly contributes to their high infectivity, spreading mainly through contact with infected individuals and virus-contaminated environmental fomites. Norovirus outbreaks can be difficult to control because the virus spreads rapidly in closed environments often resulting in secondary attack rates of >50% (Caul 1994).

The risk of environmental contamination occurring in the bathroom is likely to be greatest during acute diarrhoeal illness when billions of micro-organisms are being flushed down the toilet. During such episodes faecal material is likely to contaminate not only the bowl water but also the porcelain surfaces within the toilet bowl. Flushing produces aerosols from the force of the water running down the surfaces of the bowl and from the turbulence caused by mixing with water contained in the bowl. Previous studies have shown that toilet design influences aerosol production. Bound and Atkinson (1966) found that a siphonic toilet produced much lower concentrations of contaminated particles than the older style 'wash-down' pan by a ratio of 1 : 14. Newsom (1972) reported that the splashing produced by flushing varied with cistern height and bowl design and noted that a double-trap siphonic toilet produced more splashes than a 'wash-down' type. Obviously there is considerable variation in the design of modern flush toilets which is likely to affect the amount of turbulence, splashing, and aerosol production.

This report considers the infection risk after flushing a toilet contaminated with indicator organisms at levels required to mimic pathogen shedding during infectious diarrhoea which could be $>10^{10}$ particles per ml. A domestic close-coupled siphonic toilet, a type used widely in the UK, was used to examine the dynamics of aerosol formation and contamination of environmental surfaces after flushing. The separate effects of bacteria adhering to the porcelain sidewalls as opposed to bacteria present in the toilet bowl water on the formation of bacterial aerosols was determined. In addition, we investigated the effects of sequential flushing on environmental contamination.

MATERIALS AND METHODS

Toilet

A domestic toilet, situated in a room of 2·6 m^{-3}, in the home of one of the authors (J.B.) was used throughout (see Fig. 1). The cistern had a reservoir containing 12 l of flush water and the toilet bowl contained 2 l of water. The surface area of the internal bowl sides above the water line was 1150 cm^2. Before seeding with micro-organisms the toilet bowl water and porcelain surfaces were scrubbed with a chlorine-containing disinfectant (50 000 ppm of free available chlorine) and flushed six times to eliminate traces of the cleaning compound. This procedure was also used to decontaminate the toilet after individual experiments.

Organisms

For bacterial contamination a pigment-producing strain of *Serratia marcesens* (NCTC 10211) was used throughout

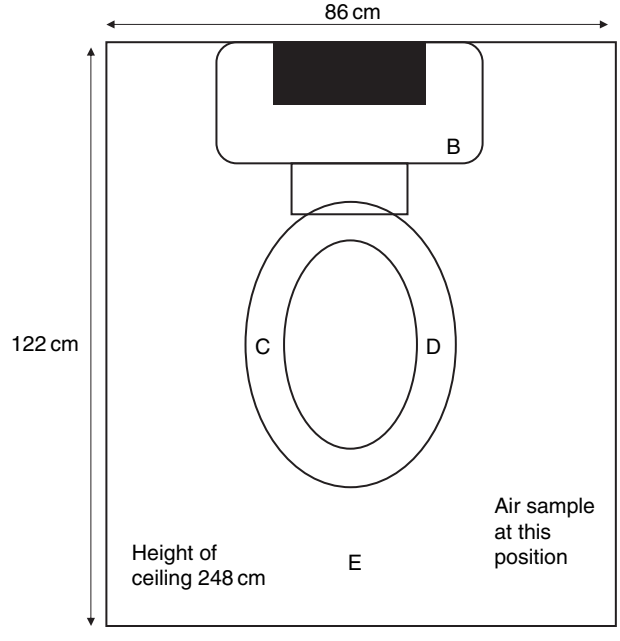

Fig. 1 The relative positions of settle plates which were exposed for 30 min after flushing the toilet. A: a shelf behind the toilet, 83 cm above the seat; B: the cistern, 41 cm above the seat; C: toilet seat, left; D: toilet seat, right; E: 30 cm in front of the toilet, level with the toilet seat

because it can be easily identified on nonselective agar and it has a low decay constant when sprayed in aqueous suspension (Darlow and Bale 1959). The organism was grown to stationary phase in 100 ml buffered peptone water (Oxoid Ltd, Basingstoke, UK) on an orbital shaker at 37°C for 24 h to give *c.* 10^9 CFU ml^{-1}. The suspension was centrifuged (2080 *g* for 30 min) before washing and suspending in 10 ml of 1/4 strength Ringer's solution (RS; Oxoid Ltd). The washed suspension was added to 80 ml of semisolid agar (0·2% w/v, Technical Agar; Oxoid Ltd) and mixed thoroughly to produce a seed inoculum containing *c.* 10^{10} bacteria.

Virus contamination was achieved using MS2 bacteriophage (ATCC 15597-B1) which is a nonpathogenic virus that can be easily propagated in the laboratory. MS2 is a nonenveloped virus, known to be relatively stable in the environment, which has been used previously as an indicator for enteric viruses (Jones *et al.* 1991; Havelaar *et al.* 1993; Dore *et al.* 2000; Allwood *et al.* 2003). Bacteriophage propagation was performed using an agar-overlay technique using *Escherichia coli* (ATCC 15597) as the host (Adams 1959). Briefly, a soft agar/host covering was prepared by overlaying agar plates (tryptone soya agar; Oxoid Ltd) with 2·5 ml of melted 0·5% agar (same medium) which contained two drops of a 6-h culture of the host in tryptone soya broth (TSB; Oxoid Ltd). The soft agar was allowed to harden and the surface covered with *c.* 0·5 ml of the concentrated bacteriophage suspension. After 24 h incubation at 37°C, the soft agar was scraped off the surface of the plates and suspended in TSB. The extract was centrifuged at 3000 *g* for 20 min to sediment the cellular debris and agar. The supernatant containing the bacteriophage was passed through a 0·2-μm filter and the filtrate stored at 4°C. Prior to use the bacteriophage suspension was allowed to equilibrate to RT. To quantitate the virus, 10-fold dilutions of the stock suspension in TSB were assayed by the overlay method. Plaques were counted after 24 h incubation at 37°C and the results expressed as plaque-forming units (PFU ml^{-1}). To seed the toilet with virus, 1·5 ml of stock bacteriophage suspension was added to 80 ml of semisolid agar (0·2% w/v) and mixed thoroughly to produce a seed inoculum containing *c.* 10^{10} PFU of virus.

Toilet seeding

Experiments were carried out to establish the dynamics of aerosol formation and surface contamination after seeding the toilet with *S. marcesens*. Key experiments were repeated using MS-2 bacteriophage to determine whether a similar pattern of contamination occurred when the toilet was seeded with a virus. Semisolid agar (0·2% w/v, 80 ml) was used as the carrier for the seed inoculum because it had the consistency of a loose stool. The inoculum was applied with a 50-ml syringe; either directly to sidewalls of the toilet bowl to give, as far as possible, an even coating on the porcelain surface above the water line, to simulate the splashing effects associated with acute diarrhoea or directly to the bowl water avoiding contamination of the sidewalls. The toilet was flushed 5 min after applying the inoculum. Preliminary tests showed that the toilet was not contaminated with pigment-producing *Serratia* species or with MS2 bacteriophage prior to seeding.

To study the aerosol formation produced by *Serratia* adhering to the sidewalls of the toilet, as opposed to the bacteria present in the bowl water, after applying the inoculum, the bowl water was disinfected with sodium hypochlorite at a final concentration of 5000 ppm of free available chlorine, before the toilet was flushed. After 30 min disinfection the residual chlorine was neutralized for 15 min by adding 8 g of sodium thiosulfate to the bowl water (final concentration 0·4%). Preliminary experiments had shown that after this level of disinfection and neutralization *Serratia* was not detected in the bowl water nor did the water exhibit residual antibacterial activity.

Microbiological sampling

The contaminated toilet bowl surface was sampled using cotton swabs (25 cm^2) moistened in RS which were rubbed over an area of 50 cm^2. The swabs were placed in 6 ml of RS and homogenized for 30 s using a stomacher. To determine bacterial counts 10-fold dilutions were prepared in RS and 0·1 ml aliquots spread onto nutrient agar plates (NA; Oxoid Ltd) which were incubated at 30°C for 18 h. Swabs for virus determination were also homogenized in RS and dilutions assayed by the agar overlay technique. The toilet bowl water was sampled by removing an aliquot with a disposable sterile plastic pipette into a 25-ml universal container.

Bacterial air samples were collected onto NA immediately after flushing the toilet, using a portable, single-sieve, impacter MicroBio MB1 (FW Parrett Ltd, London, UK). This device meets the basic criteria for a suitable reference sampler although it does not differentiate particle sizes (Griffiths and Stewart 1999). The sampler was positioned 30 cm in front of the toilet at a height of 20 cm above the toilet seat with the lid open. The door to the toilet cubicle was closed during sampling. Air sample volumes of between 100 and 600 l were collected, depending on whether the samples were collected after the first, second or third flush after seeding. A control 500-l air sample was taken prior to flushing the toilet to establish that there were no *Serratia* species or MS2 bacteriophage particles present in the air. Virus-laden aerosols were detected using 0·2% semisolid agar for the entrapment medium. Bacteriophage was detected by thoroughly mixing the entrapment medium and assaying using the overlay technique as described above.

Time	MS2 bacteriophage PFU m^{-3}	*Serratia* CFU m^{-3}	
	Untreated bowl water	Untreated bowl water	Bowl water disinfected and neutralized
Before flush	Not detected	Not detected	Not detected
After flush			
1 min	2420 (691)	1370 (527)	351 (58)
30 min	178 (91)	75 (25)	1 (0·25)
60 min	27 (25)	13 (8·5)	2·6 (0·5)

Values given within parenthesis are standard error of the mean for three replicates.

Table 1 MS2 bacteriophage and *Serratia* detected in air samples for up to 60 min after a single toilet flush. The effect of untreated bowl water and bowl water that was disinfected and neutralized prior to flushing on the dissemination of *Serratia* is shown

Settle plates containing NA, exposed for 30 min after each flush, were used to determine the fallout of bacterial aerosols onto five surfaces surrounding the toilet (Fig. 1). Settle plates for virus capture contained 0·2% semisolid agar which was assayed as for the air samples.

RESULTS

The number of *Serratia* or MS2 bacteriophage disseminated into the air after a single flush of the toilet, 5 min after the inoculum had been applied to the sidewalls, for three replicate experiments is shown in Table 1. One minute after flushing, when the toilet bowl contained untreated water, the mean air count for *Serratia* was 1370 CFU m^{-3} which declined to 75 and 13 CFU m^{-3} after 30 and 60 min respectively. The toilet water contained *c.* 10^8 CFU ml^{-1} of *Serratia* prior to flushing and 60 min thereafter the numbers declined to *c.* 10^6 CFU ml^{-1} (data not shown). When the toilet bowl water was disinfected and neutralized prior to flushing the number of bacteria released into the air was greatly reduced. One minute after flushing the air count was 351 CFU m^{-3} and this fell to <5 CFU m^{-3} after 30 and 60 min. Compared with the number of bacteria released into the air, almost twice as many virus particles were detected. One minute after the first flush 2420 PFU m^{-3} of MS2 bacteriophage were detected, declining to 178 and 27 PFU m^{-3} after 30 and 60 min respectively.

Table 2 reveals the level of contamination on surfaces surrounding the toilet 30 min after the toilet was flushed for three replicate experiments. The number of bacteria detected on the settle plates was greatest when the inoculum was applied to the sidewalls of the toilet. Counts were highest on the toilet seat (47 and 50 CFU per plate) which was more likely to have been contaminated by splashes but the shelf and the cistern which were 83 and 41 cm above the seat had mean counts of 38 and 45 CFU respectively. There was considerable variation in the counts obtained between three replicate experiments, presumably reflecting the variation in the distribution of the inoculum on the sidewalls and the flush hydrodynamics. The settle plate counts obtained after applying the inoculum directly to the water were less than half of those obtained when the inoculum was applied to the sidewalls. With one exception, the level of surface virus contamination was broadly similar to the bacterial contamination after the inoculum had been applied directly to the sidewalls.

Figure 2 shows the number of MS2 bacteriophage particles or *Serratia* attached to the porcelain surfaces of the toilet bowl 5 min after applying the inoculum to the sidewalls and 60 min after flushing. Levels of contamination

Sample sites	Location	Settle plate counts 0–30 min after flush		
		MS2 bacteriophage PFU per plate	*Serratia* (CFU per plate)	
		Inoculum applied to the sidewalls	Inoculum applied to the sidewalls	Inoculum applied to the bowl water
A	Shelf	38 (20·5)	38 (21)	14 (8)
B	Cistern	45 (16·5)	45 (28)	11·5 (4·5)
C	Seat (left)	171 (54)	47 (23·5)	20 (8)
D	Seat (right)	69 (31)	50 (18)	24·5 (4)
E	In front of toilet	42 (18·5)	34·5 (19)	11 (2·5)

Values given within parenthesis are standard error of the mean for three replicate experiments.

Table 2 MS2 bacteriophage and *Serratia* detected within 30 min of a single flush, on settle plates at various locations surrounding the toilet. For *Serratia* the inoculum was applied either to the sidewalls, or directly to the bowl water but for MS2 bacteriophage the inoculum as applied to the sidewalls only

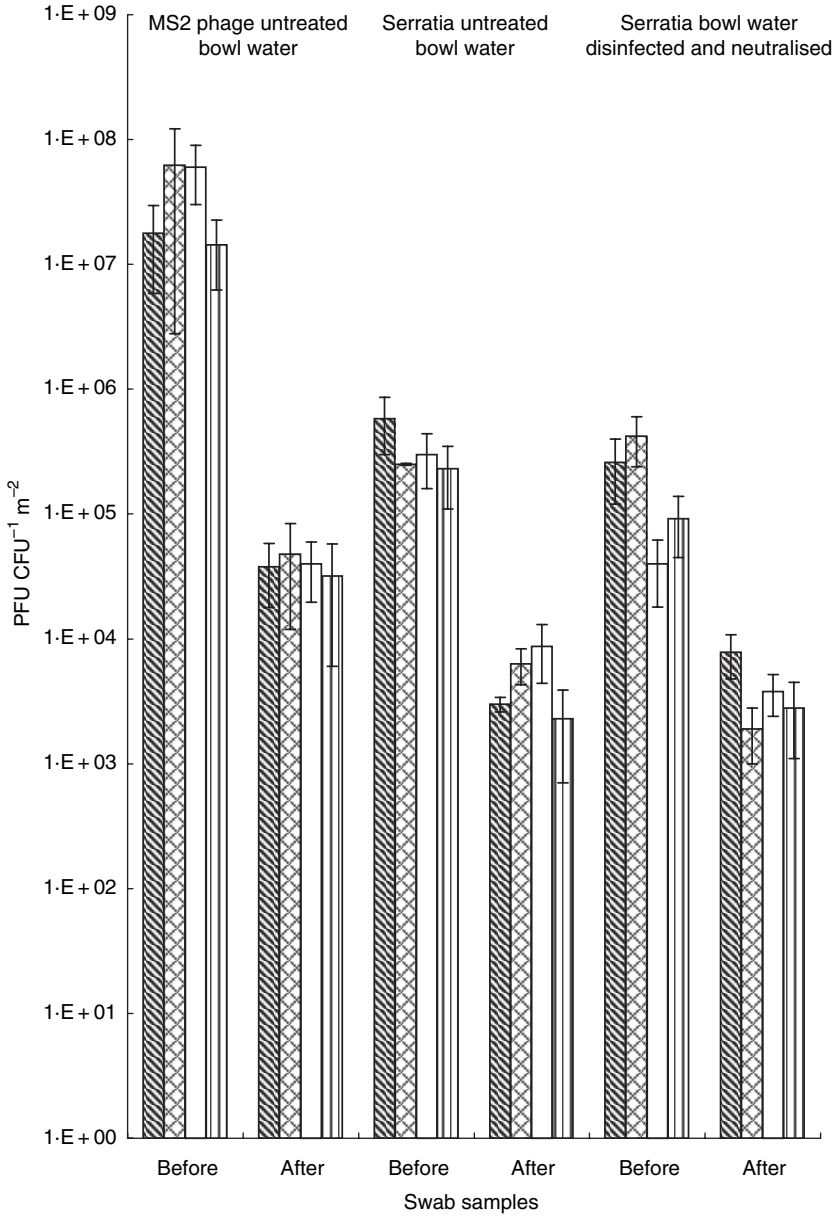

Fig. 2 Persistence of MS2 bacteriophage and *Serratia* on the porcelain surfaces of the toilet before and 60 min after flushing a seeded toilet. For *Serratia*, the effect of flushing the toilet when the bowl water contained untreated bowl water is compared with disinfection and neutralization of the bowl water prior to flushing (bars represent the standard errors of the means for three replicate experiments). ▧, Side (lt); ▩ side (rt); □, under rim (lt) and ▥, under rim (rt)

on the sidewalls and under the rim were broadly similar. Although the inoculum was not applied directly under the rim it was readily colonized with micro-organisms. This probably occurred from a 'splash-back' effect as the force of the inoculum hitting the sides of the bowl bounced back under the rim, similar to 'splash' effects that are likely to occur with acute episodes of diarrhoea. The initial level of contamination with MS2 bacteriophage on the bowl surface was *c.* 5×10^7 PFU cm^{-2} which was about 100-fold greater than for the initial bacterial contamination. Flushing the toilet reduced the level of surface attached bacteriophage by *c.* 3 log cycles cm^{-2}. The initial surface counts of *Serratia* ranged from 4×10^4 to $5{\cdot}8 \times 10^5$ CFU cm^{-2} and after flushing, there was *c.* 100-fold cm^{-2} reduction. Following a second flush the number of surface attached *Serratia* declined by a further 10-fold cm^{-2} (data not shown). When the toilet was flushed after first disinfecting and neutralizing the bowl water prior to flushing the number of surface attached bacteria were broadly similar to the levels found when flushing in the presence of untreated bowl water. This indicates that the majority of surface attached bacteria are unlikely to have been derived from the bowl water splashing onto the bowl sides through turbulence.

Figure 3a,b compares the reduction in the bacterial loading of the toilet water and the bacterial aerosol formation after three sequential flushes. Before flushing, the bowl

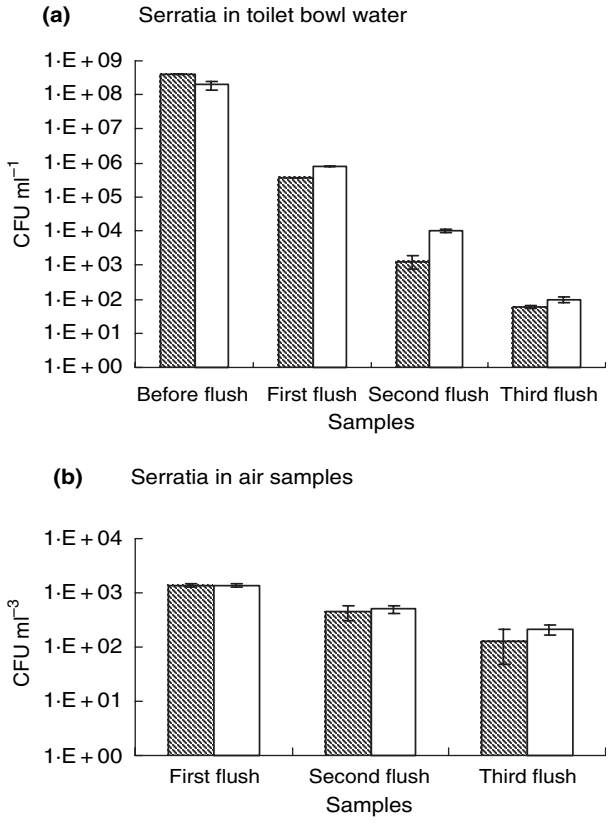

Fig. 3 Investigation of sequential flushes on bacteria persisting in the bowl water (a) and bacteria released into the air (b), comparing the effects of either; applying the inoculum to the sidewalls, or directly to the bowl water (bars represent the standard errors of the means for three replicate experiments). ▨, Bowl water and □, side walls

water contained *c.* 4×10^8 CFU ml^{-1}, when the inoculum was applied directly to the water. The numbers in the bowl water were reduced to *c.* 2×10^8 CFU ml^{-1} after applying the inoculum to the sidewalls and clearly a considerable fraction of the semisolid agar inoculum ran down the walls contaminating the bowl water. A single flush reduced the number of bacteria in the bowl water by *c.* 2·0–3·0 log cycles and after the third flush the level had decreased to *c.* 10^2 CFU m^{-1}. Application of the inoculum to either the sides walls or the bowl water made little difference to the level of bacteria released into the air which was greatest after the first flush (≅1300 CFU m^{-3}). After the second flush the number of bacteria in the air declined to *c.* 500 CFU m^{-3}. A third flush reduced the air count to 128 and 207 CFU m^3, respectively, for the inoculum applied to either the bowl water or the sidewalls. Therefore, the reduction in air sample counts was not as great as in the bowl water, clearly, indicating the residual sidewall contamination as being a major contribution to the air loads.

DISCUSSION

This investigation simulated the effects of a person using a toilet during an attack of acute diarrhoea when there is likely to be substantial contamination of both the internal toilet bowl walls and the bowl water. We were able to show that considerable numbers of both bacteria and virus-laden particles were released into the air after flushing when seeded with *c.* 10^{10} micro-organisms, to mimic levels of bacterial/viral shedding that are known to occur during infectious diarrhoea (Thomson 1954; Hutchinson 1956; LeBaron *et al.* 1990; Caul 1994). One minute after the first flush *c.* 1370 CFU m^{-3} of *Serratia* were detected but 30 and 60 min thereafter, the air count had declined by 20- and 100-fold respectively. In contrast, when the toilet was flushed after first disinfecting and neutralizing the bowl water, the concentration in the air 1 min after flushing was *c.* 350 CFU m^{-3}. These data demonstrates that both the bacteria attached to the sidewalls and those present in the bowl water contribute to the aerosol formation. MS2 bacteriophage was also released into air after toilet flushing with levels of contamination about twice that for bacteria, with 2240 PFU m^{-3} of virus particles detected in the air after the first flush. The air counts for both bacteria and viruses may have been considerably higher as a single-sieve impactor is known to be inefficient at capturing small particle sizes (Griffiths and Stewart 1999). Darlow and Bale (1959) estimated that *c.* 80% of air-borne particles released after flushing a toilet seeded with a liquid culture containing 10^{11} *Serratia* were probably <4 μm. It is possible that our air sampling technique did not detect particles of <5 μm which are likely to remain suspended in the air for several hours but could, nevertheless, eventually settle onto surfaces.

Closing the toilet lid had little effect in reducing the number of bacteria released into the air which was *c.* 1000 CFU m^{-3} after the first flush (data not shown). Although splashes would probably have been contained by closing the lid, there was a gap of 15 mm between the top of the porcelain rim and the seat, and also a gap between the seat and the lid of 12 mm which would allow aerosols to escape into the room. Conversely, Darlow and Bale (1959) found that closing the lid reduced the aerosol concentration by a ratio of 1 : 2 but their measurements were performed using a 'wash-down' toilet and an impinger air sampler. In contrast, Bound and Atkinson (1966) found that closing the lid did not significantly reduce the bacterial count in the air from a 'wash-down' toilet seeded with *E. coli* using a slit sampler positioned at seat level.

Sequential flushing of the seeded toilet resulted in prolonged air-borne transmission but with decreasing numbers of bacteria. Compared with the number of bacteria released into the air after the first flush, a second flush resulted in a threefold decrease and after the third flush the

numbers had declined by almost 10-fold. The decline in air-borne bacteria correlated with the decreasing numbers present in the bowl water. We found that reduction in numbers in the bowl water after flushing was similar to those reported by Newsom (1972). The first flush reduced the viable count in the water by 2–3 log cycles and by a similar amount after the second and third flushes. Even so, after the third flush up to 2×10^5 CFU were present in the 2 l volume of the bowl water. In contrast, the number of *Serratia* detected on the porcelain surfaces remained fairly constant after the initial flush showing that the organism had adhered to the surface. After applying the inoculum there was widespread contamination of the sidewalls; *Serratia* surface counts ranged from 10^4 to $>10^5$ CFU cm^{-2}. Although flushing reduced the initial level of colonization by about 2 log cycles, *c.* 10^3 CFU cm^{-2} persisted on the surfaces despite repeated flushing (data not shown). When the bowl water was disinfected and neutralized prior to flushing it did not alter the level of bacteria attached to the sidewalls. Thus the bacteria surviving on the sidewalls are unlikely to have been derived from the bowl water splashing back onto the walls as the toilet was flushed.

We also found that the recess under the rim of the toilet was heavily colonized with the test organisms. The recess under the rim of the toilet bowl has previously been found to be an area where *Salmonella* persisted in domestic homes where a family member had recently suffered an attack of salmonellosis with acute diarrhoea (Barker and Bloomfield 2000). The rim is an area of the toilet where limescale often accumulates, which aids bacterial retention and it can be difficult to clean effectively even with a toilet cleaner and scrubbing brush. Gerba *et al.* (1975) also found that a persistent fraction of seeded bacteria were absorbed onto the porcelain surface of the toilet and they concluded that subsequent elution of these organisms was responsible for continuing residual contamination in the toilet bowl water. In contrast, we found that after the initial seed inoculum was flushed from the sidewalls the numbers on the surface remained constant for several days of normal toilet use and thorough cleaning and disinfection using a toilet brush was required to remove the marker organisms to undetectable levels.

Thirty minutes after flushing the toilet surface contamination was detected at various locations surrounding the toilet. The level detected was probably a minimum value because micro-organisms are subject to stress by aerosolization and can be further damaged by dehydration and impaction (Dark and Callows 1973; Griffiths and DeCosemo 1994; Griffiths 1998). The highest level of surface contamination was closet to the aerosol source, at the toilet seat level, however, the marker organisms were also found on the cistern and on a shelf, 41 and 83 cm above the toilet seat respectively. The particles captured by the settle plates were likely have been >20 μm because these are known to settle within a relatively short period compared with smaller-sized particles which can remain suspended for several hours (Chatigny *et al.* 1979). Our results support earlier studies (Darlow and Bale 1959; Gerba *et al.* 1975) that there is a risk that pathogens contaminating bathroom surfaces could spread to other family members. Organisms may be picked up by the clean hands of an uninfected person and cause infection, either by direct transfer from surface-to-hand-to-mouth, or transfer by handling ready-to-eat foods (Barker *et al.* 2004). The number of bacteria/viruses found in the toilet or on surrounding surfaces must be compared with the infectious dose. Although bacteria may multiply if they contaminate food and reach levels required for infection, clearly this does not happen with viruses. Nevertheless, many faecal–oral pathogens such as norovirus, rotavirus, *Campylobacter* and *E. coli* 0157 have infective doses as low as 10–100 micro-organisms (Dupont *et al.* 1972; LeBaron *et al.* 1990; Tauxe 1992; Caul 1994; Griffin *et al.* 1994; McDonnell *et al.* 1995) and we speculate that surface-to-hand-to-mouth transfer could occur with the levels of contamination that we found on the surfaces surrounding the toilet.

The possibility that aerosols containing enteric pathogens could cause infection after being swallowed following deposition in the nose or pharynx was suggested by Darlow and Bale (1959) Recent epidemiological studies have provided convincing evidence to support this hypothesis. The likelihood of air-borne transmission of norovirus was demonstrated in an outbreak at a restaurant where no food source was implicated but analysis of the attack rate showed an inverse correlation with the distance from a person who had vomited (Marks *et al.* 2000). In infected persons up to 10^{11} g^{-1} of virus particles have been detected in stools during viral gastroenteritis and with an average stool weighing 100 g the toilet bowl could contain 10^{13} virus particles. If there is a 2-log reduction in loading after an initial flush, the bowl water could still contain 10^{11} virus particles. Multiple trips to the toilet during diarrhoea are likely to result in large numbers of pathogens persisting in the toilet, both on the porcelain surfaces and in the bowl water. Our studies have shown that such contamination is likely to result in continuing air-borne spread on subsequent flushes. It would not be unreasonable to suggest that the persistence of enteric viruses within the air could be a potential infection risk via inhalation and swallowing. Air-borne contamination could help to explain the high level of secondary spread of norovirus, within closed communities.

In normal use the toilet is unlikely to present a great risk to health as formed stool is quickly washed away and does not create large numbers of bacterial aerosols (Newsom 1972). In our opinion the health risk of using the toilet is likely to arise during acute episodes of gastroenteritis with the shedding of large numbers of pathogens. In this

investigation, we were able to show when simulating loose stool that material deposited both on the sidewalls and in the bowl water were involved in the dissemination of micro-organisms into the air and onto surrounding surfaces. Epidemiological studies from recurrent outbreaks of norovirus infection in successive cohorts of guests in hotels and on cruise ships (Ho *et al.* 1989; Gellert *et al.* 1994; Cheesbrough *et al.* 2000), suggests spread from infected persons after vomiting by settling of aerosol particles onto surfaces which are then touched by hands. In addition, these studies suggested that splashing or aerosol generation during toilet flushing may spread virus particles onto contact surfaces such as the toilet seat or flush handle. Combined with our experimental data we believe that the potential spread of enteric disease by contact with surfaces in bathrooms harbouring pathogens cannot be ignored and must be regarded as a serious infection risk.

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