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Impact of prescribed cleaning and disinfectant use on microbial contamination in the home

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Abstract

Aims: To identify and quantify the presence of *Escherichia coli*, *Staphylococcus aureus*, *Salmonella*, hepatitis A and norovirus in households and to assess the effect of chlorine and quaternary ammonium–based disinfectants following a prescribed use.

Methods and Results: Eleven sites distributed in kitchen, bathroom, pet and children's areas of two groups of 30 homes each: (i) a nonprescribed disinfectant user group and (ii) a disinfectant protocol user group. During the 6-week study, samples were collected once a week except for week one when sample collection occurred immediately before and after disinfectant application to evaluate the disinfectant protocol. The concentration and occurrence of bacteria were less in the households with prescribed use of disinfectants. The greatest reductions were for *E. coli* (99%) and *Staph. aureus* (99·9999%), respectively. Only two samples were positive for HAV, while norovirus was absent. Disinfection protocols resulted in a significant ($P < 0{\cdot}05$) microbial reduction in all areas of the homes tested compared to homes not using a prescribed protocol.

Conclusions: The study suggests that disinfectant product application under specific protocol is necessary to achieve greater microbial reductions.

Significance and Impact of the Study: Prescribed protocols constitute an important tool to reduce the occurrence of potential disease-causing microorganisms in households.

Introduction

Foodborne pathogens cause a range of diarrhoeal and flu-like illnesses. An estimated 1·8 million childhood deaths are associated with disease-causing organisms acquired via food consumption with the greatest number of cases occurring in developing countries (WHO 2008). Developed countries also suffer from a significant burden of diarrhoeal illnesses (Scallan *et al.* 2005). More than 130 million people from European countries suffer from foodborne diseases and diarrhoea is the most common symptom, while in Australia around 17 457 098 cases occurred in 2003 (WHO 2008).

In the United States, foodborne pathogens cause an estimated 76 million cases of illnesses, 325 000 hospitalizations and 5000 deaths per year (Scott 1996; Mead *et al.* 1999). This result in an estimated annual cost of $6·9 billion because of work absenteeism, cost of medication and hospitalization. This helps to contribute to the annual diarrhoeal burden of 0·72 episodes per person (Allos *et al.* 2004; Imhoff *et al.* 2004).

México has reported a significant and continuous decrease in deaths associated with foodborne pathogens; during the past 25 years, child mortality rate declined from 64 to 23 per 1000 live births yet the number of diarrhoeal episodes is similar to other developing world regions. Households in these regions have been found to harbour a wide variety of microbial pathogens (Chaidez and Gerba 2000; Terrés-Speziale and Casas Torres 2002; Sepulveda *et al.* 2007).

The presence of disease-causing micro-organisms associated with diarrhoea has been isolated from the household environment (Allos *et al.* 2004; Scott 1996). The pathogens that have been isolated from households sites are *Campylobacter jejuni*, *Clostridium perfringens*, *Escherichia coli* O157:H7, *Listeria monocytogenes*, nontyphoid *Salmonella* and *Staphylococcus aureus*, together cause an estimated 3603 526–7130 767 foodborne cases and between 2654 and 6546 deaths per year in the United States (Buzby *et al.* 1996). According to Kagan *et al.* (2002), the infectious disease transmission has been demonstrated to occur from 6 to 60% within households in which one member is ill. Even though the most common cause of foodborne acquired infections has been attributed to the global food distribution and international travels, the 80% of *Salmonella* and *Campylobacter* infections are acquired in the home in European countries as England, Wales and the Netherlands. The efficiency of cleaning and disinfecting is important to reduce microbial dispersion because of cross-contamination between people, animals (domestic and non-domestic) kitchen, toys and contact surfaces (Cogan *et al.* 1999, 2002; Fredriksson-Ahomaa *et al.* 2001; Otokunefor *et al.* 2003; Iwanicka-Grzegorek *et al.* 2005; Seepersadsingh *et al.* 2005; Van Asselt *et al.* 2008; Rutland *et al.* 2009). Proper food handling and home hygiene procedures are effective tools to prevent microbial spread (Kitamoto *et al.* 2009); however, these procedures are inconsistently applied among home dwellers, which affects the microbicidal effect of disinfectant products (Barker *et al.* 2003; de Jong *et al.* 2008); moreover, contaminated dishcloths or another cleaning utensils can serve as reservoirs and disseminators of pathogenic micro-organisms (Kagan *et al.* 2002); in fact, there are viral particles like enteric viruses, which survive during laundering process and can be transferred during washing (Gerba and Kennedy 2007). Furthermore, laboratory studies have confirmed that during food preparation, bacteria are able to survive, multiply and spread throughout the kitchen and other home areas (Cogan *et al.* 1999; Rusin *et al.* 2002; Castro-del Campo *et al.* 2004); the presence of moist is a factor that favours the survival and spread of micro-organisms (Rusin *et al.* 1998); moreover, it also favours the growth of pathogens such as *Salmonella* that is able to survive and grow by biofilm formation on surfaces, and this ability is an important risk to human health (Barker and Bloomfield 2000). The most important means for maintaining efficient microbial control include minimizing the microbial load from outside sources, efficient microbial control for vulnerable home sites and adequate cleaning and disinfecting processes (Wirtanen and Salo 2003).

This study was conducted to determine and quantify the occurrence of *E. coli*, *Salmonella*, *Staph. aureus*, hepatitis A virus and norovirus in different locations within households in a community in Mexico and to assess the disinfectant effectiveness of chlorine and quaternary ammonium–based disinfectants under specific application protocols provided to homeowners.

Materials and methods

A total of 60 homes were selected for this study. In order for the participants to be included, they had to meet the following criteria: a nonemployed housewife, children under 12 years of age, a pet and home owner willing to follow a prescribed disinfection protocol.

All houses were situated within the Culiacan city limits, the capital city of the north-western state of Sinaloa, Mexico, with an urban population of approximately one million inhabitants (INEGI, 2005).

Homes were divided into two groups of 30 homes each. Home dwellers were informed about the nature of the study and signed a consent form to obtain access to their homes. Group 1 was designated as 'nondisinfectant protocol users' (control group), and they were asked to follow their common cleaning procedures. This group used sodium hypochlorite (5%), pine oil and detergents during household cleaning before enrolment in the study. They were not asked to change their cleaning or disinfecting habits for this study. All products were local brands made in Mexico. Group 2 was designated as 'disinfectant protocol users' (test group), and they were given specific training about cleaning and disinfecting procedures as well as written protocols and specific disinfectant products (Table 1). Eleven sites within each home were selected and divided into kitchen sites (counter top, sponge, dishcloth, cutting board and sink); bathroom sites (sink, toilet bowl, and toilet seat and shower tile), pet and children toy sites. The samples were collected on a weekly basis during a period of 6 weeks, giving a total N of 3960. During the first week of the study, both home groups (prescribed and nonprescribed) were sampled immediately before and after (initial and week 1) cleaning/disinfectant application. After that, sample collection took place on weekly basis after disinfectant application (Table 1). The day of sampling varied and home owners were not told in advance the day sampling would take place.

Sample collection

Household sites (kitchen, bathroom, pet area and toys) were divided into surfaces (counter top, kitchen sink, cutting board, bathroom sink, shower tile, toilet bowl, toilet seat, pet area and children toy) and cleaning tools (sponges/dishcloths). When sampling surfaces, an area of 30×30 cm was wiped with a sterile sampling sponge prewetted with 15 ml of phosphate-buffered solution

Table 1 Prescribed disinfectant protocol

Disinfectant in provided product	Area	Frequency	Preparation	Application procedure
Quaternary ammonium compound	Counter top and sink	Twice a day	1 Top (10 ml)	Fill a top and drop on a dishcloth, wait for 5 min and rinse
2·1% Sodium hypochlorite compound/degreaser	Dishcloth, sponge and cutting board	Twice a day	60 ml per l	Dip the utensils in the solution, wait for 5 min, remove and dry
2·5% Sodium hypochlorite	Sink and toilet seat	Twice a week	30 ml per 500 ml	Using a sponge, dip it, dry and clean the surfaces, then rinse
5% sodium hypochlorite	Toilet bowl	Twice a week	Direct	Apply directly on the walls and wait for 5 min
Citric acid/ionic surfactants	Shower tile	Twice a week	Direct	Apply directly on the surface at a 15 cm of distance, then rinse
2·5% sodium hypochlorite	Homework table and toys	Twice a day	60 ml per 500 ml	Dip a dishcloth in the solution, dry clean the surface, then rinse
5% Sodium hypochlorite	Pet dish or pet area	Twice a week	240 ml per 4 l	Pet dish: apply the solution and rinse Rest area: spread the solution

(Whirl-Pak, Fort Atkinson, WI, USA). Kitchen sponges and dishcloths were placed into a sterile Ziplock bag containing 100 ml of buffered peptone water (Difco, USA). The items were hand-shaken for 2 min and then squeezed to detach micro-organisms. The items were then removed, and the liquid placed on ice and transported to the Food and Environmental Microbiology Laboratory of CIAD Culiacán within 2 h of collection. The volume holding the prewetted sampling sponges used to sample surfaces was adjusted to 25 ml to obtain a proper sample volume.

Microbiological analysis

Samples were analysed for *E. coli* and *Staph. aureus* using Chromagar ECC (Chromagar, USA) and mannitol salt agar (Bioxon, Mexico), respectively. *Escherichia coli* and *Staph. aureus* plates were incubated for 24 h at 45°C and 37°C, respectively. Colonies exhibiting blue colour on ECC agar were identified as *E. coli* (APHA 1998a). Bacterial colonies on this media were then confirmed as *Staph. aureus* using polymerase chain reaction (PCR) (Wang *et al.* 1997). *Salmonella* was identified by the most probable number (MPN) method and confirmed by PCR (Wang *et al.* 1997). After confirmation, positive strains were sent to the Instituto Nacional de Diagnótico y Referencia Epidemiológica (INDRE, México) for serotyping by Kauffman–White method (Wang *et al.* 1997; APHA 1998b).

PCR confirmation of *Salmonella* and *Staphylococcus aureus*

For each suspected colony of *Salmonella* and *Staph. aureus*, a DNA heat lyses extraction was conducted, and a portion of 2 μl of the lysed product was added to 23 μl of PCR mixture (Promega, Madison, WI, USA) containing free Mg buffer 1×, 3 mmol l^{-1} $MgCl_2$, 30 μmol l^{-1} of each DNTP (dATP, dCTP, dTTP, dGTP), 1 U of Taq DNA polymerase and 240 μmol l^{-1} of specific *Salmonella* primers SAL III (TATCGCCACGTTCGGGCAA) and SAL IV (TCGCACCGTCAAAGGAACC) (Sigma-Aldrich, St Louis, MO, USA), and for *Staph. aureus*, the primers employed were SA I (GCGATTGATGGTGATACGGTT) and SA II (CAAGCCTTGACGAACTAAAGC) (Sigma-Aldrich) as described for *Salmonella* PCR. PCR was conducted in a thermal cycler Mastercycler (Eppendorf, Hamburg, Germany), and the amplification conditions were 1 cycle of 94°C for 15 s, then 35 cycles of 94°C for 3 s, 50°C for 10 s and 74°C for 35 s and finally 1 cycle of 74°C for 2 min and 45°C for 2 s and stabilized at 4°C (Wang *et al.* 1997).

PCR products were separated by electrophoresis in 1% agarose gels containing ethidium bromide (1 μg ml^{-1}), and amplified DNA bands were observed using a UV transilluminator (Spectroline, Westburg, NY).

Hepatitis A and Norovirus identification

Viral testing was conducted on kitchen sponges and dishcloth samples. RNA extraction was conducted based on the user's manual QIAMP Viral Mini kit (Qiagen, Duesseldorf, Germany).

For hepatitis A, reverse transcription PCR was conducted using the access RT-PCR System kit (Promega); 32·75 μl of RNA was added to 17·25 μl of PCR mixture containing 1 μl of dNTPs (40 mmol l^{-1}), 10 μl buffer 5×, 2 μl $MgSO_4$ (25 mmol l^{-1}), 0·25 μl RNAases inhibitor (40 U μl^{-1}), 1 μl AVM retro-transcriptase (5 U μl^{-1}), 1 μl Taq polymerase (50 U μl^{-1}) and 1 μl of the primers L3 (50 mmol l^{-1}) (CTTCCTGAGCATACTTTGAGTC) and L4 (50 U μl^{-1}) (CCAGACCTCCATTGAACT). The

amplification condition was 1 cycle of 45°C for 45 min and then 94°C during 2 min, 94°C during 0·5 min, 51°C during 1 min, 68°C during 2 min and 68°C during 10 min.

For norovirus, reverse transcription PCR was conducted using the access RT-PCR System kit (Promega); 5 μl of RNA was added to 40 μl of PCR mixture containing 27·75 μl free RNAases water, 1 μl dNTPs (40 mmol l^{-1}), 10 μl buffer 5×, 2 μl $MgSO_4$, 0·25 μl RNAases inhibitor, 1 μl AVM retro-transcriptase (5 U μl^{-1}), 1 μl Taq polymerase and 1 μl of the primers MJV12 (50 μmol l^{-1}) (TAY CAYTATGATGCHGAYTA) and RegA (50 μmol l^{-1}) (CTCRTCATCICCATARAAIGA). The amplification condition was 1 cycle of 45°C for 45 min and then 94°C for 2 min, 94°C for 0·5 min, 50°C for 1 min, 68°C for 2 min and 68°C for 7 min.

Statistical analysis

Statistical analysis was carried out using a SPSS software ver. 17 (SPSS Inc., Somers, NY). Comparison between home groups was made using a repeated measurements design, with home groups and weeks as factors for *E. coli* and *Staph. aureus*. Comparison within weeks in each group was made using student *t*-test (Bonferroni adjustment).

The results of the study are expressed as mean and minimum–maximum range. A value of $P < 0{\cdot}05$ was considered as statistically significant.

Results

Table 2 shows the comparison between two home groups for the presence of *E. coli*. Using a prescribed protocol, cleaning and disinfecting with chlorine and quaternary ammonium–based products resulted in higher reductions in studied microbes than in the control group. The use of this protocol resulted in significant reduction in the studied organisms compared to the control group (Fig. 1).

The greatest *E. coli* concentrations were larger in the kitchen areas such as counter top, sink, sponge and dishcloth with mean values of 3·27, 3·80, 3·70 and 3·80 $\log_{10}$, and the maximum and the minimum were (0–10·0), (0–8·85), (0–9·85) and (0–10·6), respectively. The bathroom, pet area and toys were the least contaminated sites, except for the bathroom toilet (Table 2).

For the group using the prescribed disinfectant protocol, a significant *E. coli* reduction was observed when week 1 and week 5 were compared. On the other hand, the control group showed only a limited reduction in *E. coli* during this same period.

Table 3 shows a comparison between the two home groups for the presence of *Staph. aureus*. It also shows that *Staph. aureus* numbers were lower than *E. coli* in the studied households. Again disinfectant use with a prescribed protocol resulted in higher reductions when compared with the control group. *Staphylococcus aureus* was 1 $\log_{10}$ less on average in the test group compared to the control. The greatest number of *Staph. aureus* was isolated in toilet bowl and on the toilet seat. The toilet bowl showed the greatest concentration with a mean of 1·16 and range of (0–8·31) $\log_{10}$ followed by toilet seat 1·11 (0–9·99) $\log_{10}$. In addition, the control group showed an increasing number of *Staph. aureus* in the kitchen sponge during the study (Fig. 2).

For the disinfecting protocol group, a greater *Staph. aureus* reduction was observed in week 5, while the numbers of *Staph. aureus* in the control group were higher in later weeks after the first week of the study.

Incidence of *Salmonella* in kitchen sponges and dishcloths

A total of 720 samples (360 kitchen sponges and 360 dishcloths) from both the test and control groups were tested for the presence of *Salmonella* (Table 4). Overall, *Salmonella* was present in 1·38 and 2·22% of the sponges and dishcloths analysed from both disinfection and control group households, ranging from 300 to 110 000 MPN as minimum and maximum values. Serotyping results of the isolates are shown in Table 5. Several isolated serotypes have been associated with foodborne outbreaks.

Hepatitis A and Norovirus

A total of 720 samples (360 kitchen sponges and 360 dishcloths) from both the disinfection and control groups were selected to evaluate the presence of hepatitis A and norovirus. Only two samples were positive for HAV and norovirus was never detected.

Discussion

This study detected the presence of *E. coli*, *Salmonella* spp., *Staph. aureus* and hepatitis A virus in 11 different household areas. The kitchen and bathroom appeared to be the most contaminated sites. The greatest microbial reductions were observed in those households using a standard disinfecting and cleaning protocol compared to those which did not (Tables 2 and 3). This was most easily seen with *E. coli* and *Staph. aureus,* which showed a decrease tendency during the course of the study in the protocol using homes.

Previous studies have been conducted to identify and quantify faecal indicator bacteria (coliforms and faecal coliforms) and common bacterial pathogens in household

Table 2 *Escherichia coli* on surfaces in households using a prescribed protocol and control groups

Area	Home group	Initial*	Week 1*	Week 2*	Week 3*	Week 4*	Week 5*
Counter top	TG	3·271 (0–10·00)	0·994 (0–6·00)[a] $P = 0·006$	0·957 (0–5·96)	1·203 (0–9·86)	0·997 (0–6·30)	0·299 (0–4·56)
	CG	1·866 (0–10·45)	0·951 (0–6·95)	0·7925 (0–6·10)	0·696 (0–5·89)	0·8711 (0–6·48)	1·014 (0–6·48)
Sink	TG	3·805 (0–8·85)	1·498 (0–6·30)[a] $P = 0·002$	2·502 (0–7·46)	1·866 (0–7·97)	1·268 (0–5·99)	0·842 (0–5·69)
	CG	2·856 (0–7·85)	2·758 (0–7·93)	1·876 (0–8·35)	1·546 (0–8·28)	2·032 (0–8·60)	0·913 (0–6·30)
Sponge	TG	3·701 (0–9·85)	0·527 (0–5·66)[a] $P = 0·000$	1·564 (0·539–2·591)	1·891 (0–7·62)	1·692 (0–7·48)	1·212 (0–6·00)
	CG	3·270 (0–10·12)	2·824 (0–10·43)	3·156 (0–7·75)	1·367 (0–8·43)[a] $P = 0·043$	2·196 (0–8·60)	2·022 (0–10·30)
Dish cloth	TG	2·443 (0–8·30)	0·324 (0–5·47)[a] $P = 0·007$	1·28 (0–9·85)[a] $P = 0·033$	1·50 (0–9·91)	1·272 (0–7·88)	1·164 (0–7·85)
	CG	3·796 (0–10·60)	1·961 (0–9·69)[a] $P = 0·034$	2·781 (0–8·03)	0·738 (0–8·37)[a] $P = 0·053$	2·636 (0–8·48)	2·032 (0–10·30)
Cutting board	TG	2·054 (0–7·00)	0·481 (0–5·77)[a] $P = 0·009$	0·805 (0–8·45)	0·654 (0·7·78)	0·263 (0–3·95)	0·579 (0–4·43)
	CG	1·251 (0·8·58)	0·263 (0–3·95)	0·790 (0–4·95)	0·438 (0–4·65)	0·597 (0·10·30)	1·019 (0–5·99)
Sink	TG	1·618 (0–10·30)	0·283 (0–4·26)[a] $P = 0·005$	0·671 (0–10·17)	0·423 (0–4·80)	0·881 (0–6·00)	0·333 (0–5·16)
	CG	0·555 (0–8·28)	0·241 (0–7·23)	0·131 (0–3·95)	0·00 (0–0)	0·886 (0–6·00)	0·00 (0–0)
Shower tile	TG	2·531 (0–10·00)	0·665 (0–5·69)[a] $P = 0·013$	0·848 (0–7·42)	1·78 (0–8·03)	1·180 (0–8·00)	0·651 (0–7·85)
	CG	1·888 (0–7·43)	0·905 (0–6·17)	1·272 (0–7·77)	1·197 (0–7·92)	1·454 (0–5·61)	1·461 (0–8·30)
Toilet bowl	TG	4·145 (0–10·48)	0·283 (0–4·56)[a] $P = 0·000$	0·981 (0–10·13)	0·907 (0–5·73)	1·025 (0–9·85)	0·694 (0–6·90)
	CG	1·728 (0–6·82)	2·311 (0–7·07)	1·962 (0–9·80)	1·182 (0–7·89)	1·659 (0–10·00)	2·228 (0–10·48)
Toilet seat	TG	2·022 (0–8·48)	0·298 (0–5·00)[a] $P = 0·010$	0·578 (0–6·30)	0·00 (0–0)	0·563 (0–7·30)	0·00 (0–0)
	CG	1·403 (0–6·35)	0·488 (0–5·95)	0·398 (0–6·64)	0·425 (0–6·95)	0·839 (0–6·30)	0·752 (0–7·30)
Pet area	TG	3·038 (0–10·00)	0·455 (0–5·77)[a] $P = 0·000$	0·559 (0–5·82)	0·487 (0–5·18)	1·115 (0–5·90)	0·307 (0–5·28)
	CG	1·351 (0–8·34)	0·656 (0–7·88)	0·538 (0–7·00)	1·020 (0–9·23)	0·874 (0–8·30)	0·215 (0–6·48)
Children toy	TG	1·745 (0–7·30)	0·631 (0–6·48)	0·419 (0–6·45)	0·593 (0–5·23)	0·131 (0–3·95)	0·131 (0–3·95)
	CG	0·943 (0–5·90)	0·371 (0–6·58)	0·00 (0–0)	0·147 (0–4·43)	0·390 (0–6·48)	0·261 (0–7·85)

TG test group (disinfectant protocol users); CG control group (nondisinfectant protocol users); *Mean $\log_{10}$ CFU of *E. coli* per 900 cm^2; Range $\log_{10}$ CFU of *E. coli* per 900 cm^2 is indicated in parenthesis. [a]Statistical significance between consecutive weeks (current and previous week) within the same home group and no letter means no significance.

settings and to determine those disinfecting products that are the most effective in reducing pathogens (Rusin *et al.* 1998; Cogan *et al.* 1999). However, no previous studies have been focused on the evaluation of a prescribed cleaning and disinfecting protocol in a home in a developing country. As observed in previous studies, a critical step for the reduction in micro-organisms was the following application procedure of chlorine and/or quaternary ammonium–based products on household surfaces.

During the course of the intervention, the studied bacteria decreased in the disinfectant protocol users group, while, in contrast, bacterial numbers in the control group either increased or remained unchanged.

The disinfectant protocol users group showed a tendency to reduce microbial concentrations, with mean reductions of 2·5 $\log_{10}$ and reduction percentages between 99 and 99·9999% (2–6 $\log_{10}$) for *E. coli* and *Staph. aureus*, which remained low during the study. In addition to logarithmical reduction, microbial load showed a decrease with regard to presence of positive samples on the different household sites, where it was possible to appreciate the reduction in the presence of *E. coli* and *Staph. aureus*

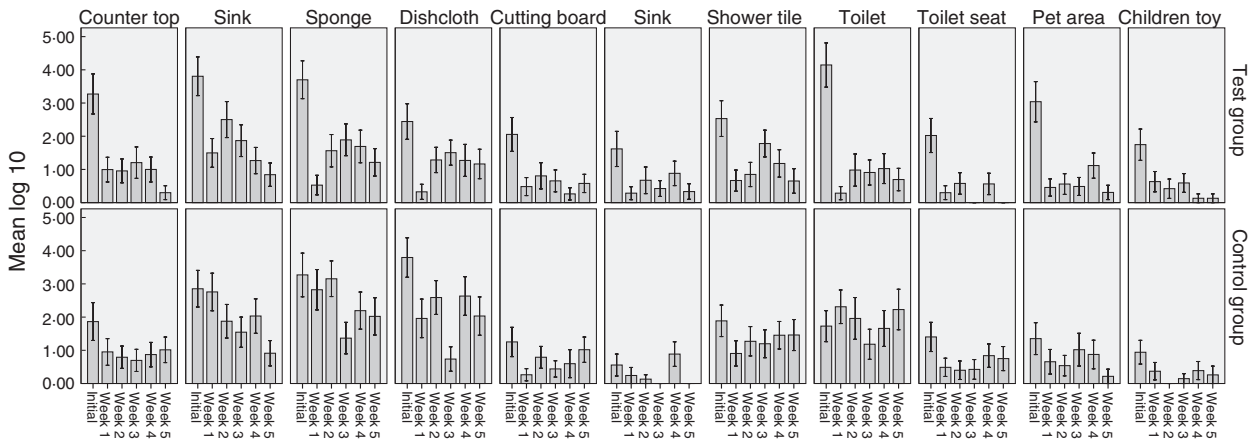

Figure 1 *Escherichia coli* comparison between home groups during the periods of sampling.

Table 3 *Staphylococcus aureus* on surfaces in households using a prescribed protocol and control groups

Area	Home group	Initial*	Week 1*	Week 2*	Week 3*	Week 4*	Week 5*
Counter top	TG	0·427 (0–7·90)	0·00 (0–0)	0·184 (0–5·55)	0·507 (0–8·58)	0·174 (0–5·23)	0·00 (0–0)
	CG	0·200 (0–6·00)	0·00 (0–0)	0·00 (0–0)	0·00 (0–0)	0·00 (0–0)	0·00 (0–0)
Sink	TG	0·771 (0–7·94)	0·00 (0–0)	0·802 (0–7·07)	0·457 (0–8·41)	0·00 (0–0)	0·00 (0–0)
	CG	0·509 (0–9·21)	0·00 (0–0)	0·206 (0–6·21)	0·206 (0–6·20)	0·00 (0–0)	0·00 (0–0)
Sponge	TG	0·594 (0–6·43)	0·214 (0–6·43)	0·00 (0–0)	0·173 (0–5·21)	0·00 (0–0)	0·00 (0–0)
	CG	0·489 (0–8·58)	0·00 (0–0)	0·588 (0–9·00)	0·00 (0–0)	0·00 (0–0)	0·147 (0–4·43)
Dishcolth	TG	0·913 (0–8·03)	0·276 (0–8·28)	0·284 (0–8·54)	0·202 (0–6·07)	0·333 (0–5·45)	0·00 (0–0)
	CG	0·486 (0–8·03)	0·00 (0–0)	0·537 (0–10·20)	0·151 (0–4·56)	0·00 (0–0)	0·00 (0–0)
Cutting board	TG	0·683 (0–8·26)	0·00 (0–0)	0·198 (0–5·94)	0·379 (0–5·88)	0·219 (0–6·58)	0·00 (0–0)
	CG	0·00 (0–0)	0·241 (0–7·23)	0·00 (0–0)	0·170 (0–5·10)	0·00 (0–0)	0·00 (0–0)
Sink	TG	0·197 (0–5·94)	0·273 (0–8·20)	0·685 (0–8·10)	0·679 (0–10·20)	0·363 (0–5·46)	0·00 (0–0)
	CG	0·00 (0–0)	0·00 (0–0)	0·718 (0–9·88)	0·00 (0–0)	0·00 (0–0)	0·00 (0–0)
Shower tile	TG	0·438 (0–7·18)	0·416 (0–7·40)	0·170 (0–5·10)	0·00 (0–0)	0·140 (0–4·22)	0·171 (0–5·16)
	CG	0·397 (0–5·98)	0·611 (0–6·49)	0·00 (0–0)	0·179 (0–5·39)	0·00 (0–0)	0·227 (0–6·83)
Toilet bowl	TG	1·157 (0–8·31)	0·00 (0–0)	0·00 (0–0)	0·450 (0–8·03)	0·147 (0–4·0·42)	0·00 (0–0)
	CG	0·507 (0–8·75)	0·676 (0–8·53)	0·284 (0–8·54)	0·480 (0–8·02)	0·540 (0–9·23)	0·262 (0–7·86)
Toilet seat	TG	0·294 (0–8·84)	0·00 (0–0)	0·488 (0–8·07)	0·720 (0–8·23)	0·00 (0–0)	0·00 (0–0)[a]
	CG	0·610 (0–6·23)	1·106 (0–9·99)	0·00 (0–0)	0·00 (0–0)	0·00 (0–0)	0·00 (0–0)
Pet area	TG	0·209 (0–6·28)	0·00 (0–0)	0·00 (0–0)	0·00 (0–0)	0·00 (0–0)	0·00 (0–0)
	CG	0·301 (0–9·03)	0·436 (0–6·64)	0·00 (0–0)	0·194(0–5·83)	0·00 (0–0)	0·00 (0–0)
Children toy	TG	0·438 (0–7·93)	0·280 (0–8·40)	0·197 (0–5·91)	0·00 (0–0)	0·00 (0–0)	0·00 (0–0)
	CG	0·163 (0–4·91)	0·213 (00–6·41)	0·00 (0–0)	0·210 (0–6·33)	0·00 (0–0)	0·00 (0–0)

TG test group (disinfectant protocol users); CG control group (nondisinfectant protocol users); *Mean $\log_{10}$ CFU of *Staph. aureus* per 900 cm^2; Range $\log_{10}$ CFU of *Staph. aureus* per 900 cm^2 is indicated in parenthesis. [a]Statistical significance between consecutive weeks (current and previous week) within the same home group, and no letter means no significance.

from 50 to 100% during the time of study, mainly for the disinfectant protocol users group.

It is evident that using disinfectants following a prescribed protocol helps to increase microbial reductions. This was the case when the protocol and control groups were compared to each other. In the disinfectant protocol users, the bacterial numbers in the sponges declined after the initial sampling period and remained low. On the

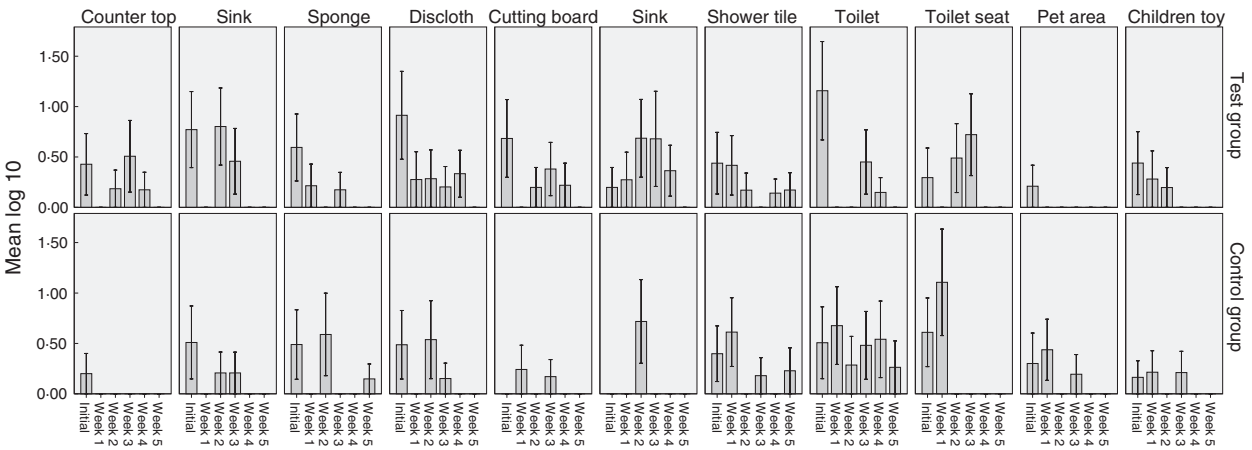

Figure 2 *Staphylococcus aureus* comparison between home groups during the periods of sampling.

Table 4 Presence of *Salmonella* in kitchen sponges and dishcloths

Home	Sample	Positive	Negative	Maximum	Minimum
TG*	Sponge	2	178	1200	610
	Dish cloth	3	177	2100	320
CG†	Sponge	2	178	1600	300
	Dish cloth	6	174	110 000	930

*TG, test group (disinfectant protocol users); †CG, control group (nondisinfectant protocol users), Maximum and minimum expressed as most probable number (MPN) values.

Table 5 *Salmonella* serotypes identified in kitchen sponges and dishcloths

Sample	Serotype
Sponge	Rough somatic antigen (1)
	Saint Paul (1)
	Anatum (1)
	Oranienburg (1)
Dish cloth	Infantis (1)
	Oranienburg (4)
	Saint Paul (1)
	Anatum (1)
	O11 antigenic factor (2)

Number of positives strains per each serotype is indicated in parenthesis.

other hand, bacterial numbers remained higher in the control group. Dishcloths showed a significant reduction ($P = 0{\cdot}001$) in bacterial numbers in the protocol group, between the initial week and week 1, while there was not any other significant reduction ($P > 0{\cdot}05$) observed, which can be associated with fails during the application of the protocols. Rusin *et al.* (1998) noted that the highest faecal and coliforms concentrations were found in the sponge, dishcloth and kitchen sink and the lowest concentrations were found at bathroom sites.

HAV was only identified twice of 720 samples collected, although virus viability was not determined. No norovirus was identified. This suggests that enteric virus levels in the studied households were low or below the detection level of the test methods.

Hypochlorite and quaternary ammonium–based disinfectants have been demonstrated to lower the concentration of total coliforms, faecal coliforms and heterotrophic bacteria in the home (Rusin *et al.* 1998). Previous studies suggested that water rinsing of kitchen surfaces is a critical step in achieving hygiene conditions; however, disinfectant products have the greatest impact on microbial reduction (Cogan *et al.* 2002). According to Barker *et al.* (2003), disinfectant products should be considered to improve the hygiene conditions at home environment reducing cross-contamination during food handling. Oundo *et al.* (2008) suggested that food handlers may potentially harbour pathogens such as enteroaggregative *E. coli* and can be spread at kitchen sites posing risk to home dwellers. Van Asselt *et al.* (2008) suggested that *Camp. jejuni* could be transferred during food preparation from hands to cutting boards and to knives. The adherence to the application of hygiene procedures helps to reduce microbial populations at homes, thus reducing the risks of infection (Cogan *et al.* 1999).

Conclusions

Potential pathogens were found in several household areas (kitchen sponges, dishcloth and counter top). The use of a prescribed cleaning and disinfecting protocol involving chlorine and quaternary ammonium–based disinfectants was found to lower the numbers of these bacteria, reducing the risks of household-acquired

infections. On the other hand, hygiene education is important in order for the impact to be significant in reducing the levels of contamination at critical sites within household environment.

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