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Detection of viruses and body fluids which may contain viruses in the domestic environment

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SUMMARY

The domestic environment was investigated for the presence of viruses and body fluids that may contain viruses. A range of surfaces in 39 homes (17 visited on 2 occasions) were sampled by swabbing and analysed using cell culture, reverse transcription polymerase chain reaction for enteroviral RNA, haemoglobin as a marker for blood, amylase as an indicator of urine, saliva and sweat, and protein as an indicator of general hygiene. Haemoglobin was found on 1·9% of surfaces sampled and of the positive samples 30% were from articles frequently handled. Amylase ($>$ 5 U/l) was found in 29·3% of samples tested. Protein was found in 97·8% of samples tested. Enteroviral RNA, indicating the presence of virus, was detected in 3 out of 448 samples tested; they were from a tap handle, telephone handpiece and a toilet bowl. No viruses were isolated in cell culture, however significant problems were encountered with bacterial and fungal contamination. This work demonstrates that only testing environmental samples for bacteria and ATP may not give a total view of the microbiological problem in the home. A range of test methods is useful to gain a broad view of the problems of hygiene in the home and to allow comparative studies of specific areas such as the kitchen and bathroom.

INTRODUCTION

The importance of viruses as human pathogens needs no explanation. Whilst many studies have been carried out to define the occurrence and routes of transmission of virus infections in hospitals [1] and day-care centres for children [2, 3], no such studies have been carried out in the domestic environment where many infections occur. During and after infection, virions are shed in large numbers in many body fluids including blood, faeces, urine, saliva and nasal secretions. They may pose a risk to health if they contaminate inanimate surfaces, survive, and are transferred to a susceptible host.

In laboratory studies viruses have been shown to survive on a wide range of surfaces for varying times, depending on the virus type (enveloped or non-enveloped), surface (porous or non-porous) and the relative humidity (RH) of the environment. Herpes simplex virus has been shown to survive for up 2 h on a plastic surface at 37–40 °C in a humid atmosphere [4]. Mbithi and colleagues [5] demonstrated that the survival of hepatitis A virus was inversely proportional to the RH and temperature. The 'half lives' of the virus ranged from $>$ 7 days at low RH and 5 °C to 2 h at 95% RH and 25 °C. Mahl and Sadler [6] reported the survival of adenovirus, poliovirus and herpes simplex virus for up to 8 weeks under conditions of low humidity at 25 °C.

Viruses can survive on inanimate objects, but their transfer and survival on hands also plays a part in their transmission. The transfer of rhinoviruses has been demonstrated from surfaces to hands, e.g. taps and door handles to hands in laboratory studies [7]. Once on hands of susceptible individuals virus can

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easily be transferred to the nasal mucosa where infection may occur.

In this study the occurrence of viruses and body fluids which may contain viruses was investigated by swabbing surfaces around the home. The surfaces included baths and toilets, door handles and telephones, i.e. surfaces where contact may occur directly from body fluids, hand contact or aerosols. The samples were tested for a range of viruses by isolation in cell culture, for enteroviral RNA by reverse transcription polymerase chain reaction (PCR) [8], haemoglobin indicating the presence of blood and amylase indicating the presence of saliva, serum, urine and sweat. Sampling was carried out both in winter and summer to take account of seasonal variation in viral infection.

MATERIALS AND METHODS

Volunteers and sampling sites

Volunteers were recruited from the Unilever Research, Port Sunlight Laboratory consumer panel. The criteria for selection were that they had at least one child < 12 years and were willing to answer a short questionnaire on family health, and cleaning habits. Volunteers were asked not to clean their homes 24 h prior to sampling. The study was conducted in two 5-week parts; June–July when 19 homes were sampled and January–February when 20 homes were sampled. In the second part of the study 17 of the original homes were revisited. Samples were taken from a range of surfaces in the kitchen, and bathroom.

Sampling procedure

For each site sampled two adjacent areas were swabbed. Swabs were moistened with sterile saline (0·85%), and an area of 10 cm^2 was swabbed with 10 vertical and 10 horizontal strokes. The virus transport swab (Copan 147C Viroswab) was processed according to the manufacturer's instructions. The tip of the second swab (Medline Scientific Diagnostics Code 300261.2) was cut off into a 5 ml plastic bijou containing 1 ml sterile saline. Swabs were stored on ice until returned to the laboratory.

The swabs (in their respective solutions) were mixed vigorously on a vortex mixer for 10 s and allowed to stand for 1 min, after which the swab was removed. The virus transport medium (VTM) was aseptically removed from the swab case and stored in a 2 ml sterile tube.

To avoid contamination of the PCR sample, 200 μl of the saline sample was aseptically removed and placed in a sterile tube before any other tests were carried out on the saline sample. The PCR samples were stored at -70 °C until tested.

Haemoglobin detection

The Haemostix test strip (Bayer 2816A), which is designed to detect haemoglobin in urine was used to detect haemoglobin in the environmental samples. The strip was completely immersed in the sample and then immediately removed. The edge of the strip was tapped on the side of the tube to remove excess liquid. After 1 min the colour of the test area was compared with the colour chart provided.

In order to eliminate false positive results caused by oxidizing contaminants (e.g. sodium hypochlorite), all positive samples were neutralized with an equal volume of 100 mg ml^{-1} sodium thiosulphate solution and retested as described above. Only samples giving a positive result on retesting were regarded as genuinely positive.

Amylase detection

Amylase activity was measured using the Sigma 577 kit. The test was performed according to the manufacturer's instructions. The α-amylase activity at 37 °C was calculated. Normal levels of amylase in serum, urine, and saliva are 53–123, 0–375, and 0–300 U l^{-1} respectively.

Protein determination

The Bio-Rad protein microassay was used according to the manufacturer's instructions, to determine total protein. Standard solutions of bovine serum albumin (Wilfred Smith AF2070), ranging from 2 to 1024 ml^{-1}, were prepared in sterile saline for each assay. Concentration in samples were calculated by logarithmic regression analysis.

Cell culture

A range of cells were used for virus isolation. Primary monkey kidney cells were obtained from Animal Cell Research Department, Centre for Applied Microbiology and Research, Porton Down. The cells were grown in medium 199 supplemented with 5% heat-inactivated fetal calf serum (FCS), 20 mM l^{-1} glutamine, 2·5 μg ml^{-1} amphotericin B and 0·05 mg ml^{-1}

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gentamicin and maintained in medium 199 supplemented as above but with the level of FCS reduced to 1%.

Hep-2 (human caucasian larynx carcinoma), and HeLa Ohio (human cervical carcinoma) cells were purchased from Gibco–BRL an ICN Flow, respectively. HEL cells (human embryo lung) were prepared in our own laboratory. Growth medium for cells consisted of Eagle's minimal essential medium (with Earle's salts) supplemented with 10% FCS, 1% (v/v) 100× non-essential amino acids, 2·5 μg ml^{-1} amphotericin B and 0·05 mg ml^{-1} gentamicin. Maintenance medium consisted of growth medium with FCS reduced to 1%. Stocks of cells were grown in 75 cm^2 cell culture bottles and passaged as required. Plates (48-well) for virus isolation were prepared by subculturing the cells at a dilution to give confluent monolayers in 24–48 h.

Virus isolation

Cells used for isolation were selected to allow detection of enteroviruses, parainfluenza viruses, adenoviruses, herpes and rhinoviruses. Confluent monolayers in 48-well plates were prepared as described. To each well 100 μl of sample was added (one sample/well/cell type) and the plates incubated for 1 h in an atmosphere of 5% CO_2 in air. Warmed maintenance medium (200 μl) was added to each well and the plates incubated at 33 °C for HeLa and 37 °C for all other cells. After overnight incubation, the medium was removed aseptically and replaced with fresh medium. Cells were observed daily for cytopathic changes. After 7 days incubation, samples were passaged as described above. Any samples showing signs of virus-induced cytopathic effect were passaged for a third time as described.

Reverse transcription polymerase chain reaction for enteroviral RNA

Extraction

RNA extracts were prepared from saline or cell culture samples using Ultraspec 3 (Biotecx). Extraction was performed according to the manufacturer's instructions except that glycogen (1 μl of 20 μg ml^{-1}) was used in place of resin. Extracted RNA was resuspended in 20 μl diethyl pyrocarbonate treated water [10] and stored at −20 °C prior to RT-PCR.

PCR primers and probe

Enterovirus PCR primers EP1 and EP4 were designed by Gow and colleagues [9]. These amplify all enteroviruses except hepatitis A virus. Probe P6803 (5′-ACA(TA)GGTGTGAAGAG-3′-356–370) was designed in-house from the aligned coxsackie, echo, polio and rhinovirus genome sequences using the DNA Star database. P6803 was designed with a single degenerate (mixed T/A) base, and had 100% complementarily with the four aligned viral genomes. PCR primers were synthesized on an ABI 381A DNA synthesizer and the probe obtained with a digoxigenin label from Oswel DNA Service.

Reverse transcription (RT)

RT was performed on 10 μl RNA extract for 1 h at 37 °C (the remaining 10 μl was refrozen and stored at −20 °C). RT was mediated by reverse transcriptase (Gibco) and cDNA synthesis was primed with primer EP4 (15 pmol).

PCR of viral cDNA

Fifteen μl RT product was added to 35 μl PCR mix. The remaining RT product was stored at −20 °C and used for repeat tests. The PCR mix contained buffer (60 mM Tris-HCl, 15 mM $(NH_4)_2SO_4$, 2 mM $MgCl_2$, pH 9·5), primers EP1 and EP4 (15 pmol), deoxynucleotide triphosphates and Taq polymerase (Gibco 2·5 U). PCR was for 40 cycles (denaturing 1 min 93 °C, primer annealing 1·5 min at 54 °C and DNA synthesis 2 min at 72 °C).

Detection of RT-PCR amplification products

Detection was by electrophoresis in an ethidium-bromide-stained agarose gel. PCR product (44 μl) was mixed with 11 μl 5× loading buffer [10] and 50 μl loaded onto the gel. Electrophoresed gels were observed under u.v. light. The enterovirus PCR generated a specific amplification product of 415 bp. The size of the amplification products was compared with a 100 bp DNA ladder (Gibco).

Southern blotting of PCR products and detection with digoxigenin labelled enteroprobe P6803

Agarose gels were denatured by incubating with 0·5 M sodium hydroxide/1·5 M NaCl for 1 h and neutralized

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for 1 h in 1 M Tris pH 8 containing 1·5 M NaCl. A capillary blot was set up to transfer the DNA overnight on to Hybond-N^+ membrane (Amersham). The blotted membrane was optimally u.v. cross linked. The membrane was blocked and probed according to the Boehringer method [11]. The probing was performed at 35 °C with 4 pmol P6083 in 10 ml hybridization solution. Washing was carried out at 37 °C to remove unbound probe. Following washing, anti-digoxigenin conjugate incubation and further washing, substrate colour was developed for 1·5 h in 20 ml NBT/BCIP conjugate (Sigma FAST BCIP/NBT tablets).

Table 1. *Sites included within each grouping*

Group	Sites included in each group*
Toilet	Cistern[2], bowl (above and below flush) lid[2,3], seat[2], flush handle[1], surfaces near the toilet[2], door handle[1], outer surfaces of bowl[2]
Washbasin	Bowl, taps[1], surfaces behind taps, surfaces (15 and 45 cm) above washbasin[2]
Bath	Bath surfaces, taps[1]
Telephone	Mouthpiece[2] and handpiece[1]
Baby	Cot rails[1], trainer seat[2], potty (inside[3] and out[2]), change mat[3]
Kitchen	Work surface, door handle[1], fridge door[1]

* Possible route of contamination: 1, handling; 2, aerosol contamination; 3, faecal contamination.

RESULTS

Forty-seven different sites within each house were sampled. The sites were grouped together (Table 1), in order to simplify tabulation.

Haemoglobin detection

Preliminary tests were carried out with Haemostix to determine the levels of haemoglobin that could be recovered from surfaces. Small volumes (1, 0·1, and 0·01 μl) of sheep blood in anticoagulant were spotted onto tiles and allowed to dry. After 2 h the surface was swabbed and tested as described. A positive (1+) result was obtained from 0·01 μl blood dried on to and then recovered from the surface. All environmental samples found to be positive were neutralized and retested

In the first survey 1·5% of surfaces sampled were found to be positive, and in the second 2·4% were found to be positive. Positive results were found most frequently on taps (27·3, first survey and 31·6%), on the surfaces of washbasins (54·5 and 26·3%) particularly behind taps, and on the surfaces of the toilet (18·2 and 31·6%). Positive results were also recorded from the rusty hinges of toilet seats, which could be false positives due to the reaction of iron from the metal hinges with the test strips and were excluded from the calculation of % positive.

Amylase detection

A series of preliminary tests were carried out with the amylase assay to check whether common household products could cause false positive or negative results. These tests were carried out by analysing amylase standards in the presence of hypochlorite bleach (100, 250, 400, 800, and 1000 ppm) and a general purpose cleaner (1:50, 1:100 and 1:500). No false positive or negative results were obtained with any dilution of product tested.

The levels of amylase found on the surfaces varied widely from 0–322 U l^{-1}. Surfaces where amylase levels were found to be high in both studies were: the baby change mat, door handles, washbasins (particularly inside the bowl), and the telephone mouthpiece. In study 1, high levels were also found on cot rails and the upper surfaces of the toilet lid. The highest levels were found on a telephone mouthpiece (322 M l^{-1}), a toilet bowl above the flush (228 M l^{-1}) and inside a washbasin (228 U l^{-1}). Within each sample group the highest frequency of occurrence of amylase (> 20 U l^{-1}) (Table 2) on surfaces was found to be as follows: telephone (26·5, 35%), baby items (27·3, 29·2%), and washbasin (13·8, 17·5%). If the samples were analysed in terms of the possible routes of contamination (Table 3), i.e. those handled frequently, e.g. taps, door handles, telephones, etc., those where aerosol contamination may occur and surfaces where faecal contamination may occur. The frequency of occurrence of amylase values over 20 U l^{-1} is as follows: handled 14·1 and 14·8%, aerosol contamination 10·9 and 12·0%, and possible faecal contamination 7·0 and 10·0% for survey 1 and 2 respectively.

Protein detection

A summary of the protein results (included as a general indicator of hygiene) for both surveys is

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Table 2. *Frequency of occurrence of levels of amylase in both studies*

	Frequency (%) of samples in concentration range (U l^{-1})*										Total no. of samples	
	0·0–4·9		5·0–9·9		10·0–14·9		15·0–19·9		> 20·0			
	1	2	1	2	1	2	1	2	1	2	1	2
Toilet	77·2	72·3	6·2	9·3	7·6	5·7	0·7	3·7	8·3	9·0	289	300
Washbasin	66·1	59·0	9·0	7·5	7·9	13·5	3·2	2·5	13·8	17·5	189	200
Bath	82·4	83·6	6·1	6·4	6·1	6·4	1·5	0·0	3·8	3·6	131	140
Telephone	55·9	47·5	0·0	10·0	17·7	2·5	0·0	5·0	26·5	35·0	34	40
Baby	59·1	41·7	4·5	12·5	9·1	12·5	0·0	4·2	27·3	29·2	22	24
Kitchen	66·7	70·5	9·1	5·1	4·6	8·9	0·0	3·9	19·7	11·5	66	78

* 1, Study 1 (summer); 2, Study 2 (winter).

Table 3. *Frequency* (%) *of detection of amylase* (*at levels > 20 U* L^{-1}) *by potential route of contamination*

Route of contamination	Study 1	Study 2
Items handled	14·1	14·8
Aerosols	10·9	12·0
Faecal contamination	5·0	10·0

shown in Table 4. Only 2·2% of all 1519 samples tested contained no protein. Levels above 10 μg ml^{-1} were found on a variety of surfaces. The highest levels were 4800 μg ml^{-1} on a toilet bowl below the flush and 1127 μg ml^{-1} on a surface near the toilet. If the results are analysed in terms of possible routes of contamination, the frequency of occurrence of levels of protein (> 10 μg ml^{-1}) were as follows: handled 10·6 and 10·1%, aerosol contamination 13·6 and 16·5%, and possible faecal contamination 9·7 and 12·2% for survey 1 and 2 respectively.

Isolation of viruses

Recovery of viruses by the procedure outlined in the methods was evaluated using surfaces artificially contaminated with poliovirus, coxsackie B4 virus and echovirus 11. Virus was pipetted on to the surface, and air dried. The surfaces were swabbed and the VTM left at 4 °C overnight. Samples were diluted in maintenance medium and inoculate onto confluent monolayers of vero cells. The titre of the recovered virus was calculated using the Karber formula [12], the virus recovered from the surfaces was found to be 4 $\log_{10}$ $TCID_{50}$ ml^{-1} lower than that put on to the surface, the lowest level detected was 0·5 $\log_{10}$ $TCID_{50}$ ml^{-1}.

None of the samples from the two studies produced any cytopathic effects (cpe) in cell culture which could be clearly attributed to a particular virus. Samples which showed any cpe were passaged up to four times and where appropriate tested by RT-PCR for enteroviral RNA. One of the major problems encountered with the cell culture was bacterial (particularly *Pseudomonas* spp.) and fungal contamination of the cultures after the environmental samples were added. The strains encountered appeared to be resistant to the type and level of antibiotics (2·5 μg ml^{-1} amphotericin B and 0·05 mg ml^{-1} gentamicin) used in the maintenance media. Bacterial contamination resulted in the discard of a number of culture plates before samples could be passaged, thus reducing the number of samples which actually went through the full isolation procedure. No viruses were isolated from samples using the procedure outlined in the methods.

Detection of viral nucleic acid

RT-PCR was performed on 148 samples from the first survey, i.e. those giving indistinct cpe in appropriate cell cultures. Initially bands of approximately the correct molecular weight were found in 18 samples. Of these, two were found to be repeatedly positive in gel and blot. Non-repeatable results were thought to be due to the presence of very low levels of target sequence which degraded on storage. The two positive samples were from the cold tap handle of a washbasin and the upper surface of a telephone handpiece, both of which are frequently handled. The household where enteroviral RNA was found on a tap handle reported illness 2 weeks prior to sampling.

Three hundred samples from the second survey, which produced an indistinct cpe in cell culture were tested for enteroviral RNA as described. A single positive result was obtained from a sample taken from

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Table 4. *Frequency* (%) *of occurrence of protein in both surveys*

	Frequency (%) of samples in concentration range (μg ml^{-1})*										Total no. of samples	
	0–4·9		5·0–9·9		10·0–14·9		15·0–19·9		> 20			
	1	2	1	2	1	2	1	2	1	2	1	2
Toilet	54·4	43·0	31·4	42·0	6·9	6·7	1·1	2·7	6·3	5·7	287	300
Washbasin	47·6	47·2	31·2	36·7	9·0	4·5	1·1	4·5	10·6	7·0	189	199
Bath	58·0	55·0	25·2	35·7	9·2	4·3	3·8	2·8	3·8	2·1	131	140
Telephone	64·7	45·0	23·5	23·5	0	7·5	5·9	0	5·9	0	34	40
Baby	54·5	45·8	27·3	50·0	9·1	0	4·6	0	4·6	4·2	22	24
Kitchen	52·3	55·1	30·8	33·3	4·6	3·9	4·6	2·8	7·7	6·4	65	78

* 1, Study 1 (summer); 2, Study 2 (winter).

the toilet bowl just below the flush. In this household two out of the three children were ill at the time of sampling. All three PCR reactions from the first RT product resulted in a band of the correct molecular weight by agarose gel electrophoresis. However, there was insufficient product from the re-amplification to enable sequencing of the product which may have allowed identification of the particular picornavirus.

Sixteen samples from the 300 tested n PCR resulted in products of molecular weight bands of 500 bp rather than the specific 415 bp. The higher molecular weight bands were diluted, re-amplified and sequenced using a Perkin–Elmer ABI 7700 Sequence Detection System, the sequence was compared with those on the DNA Star database and found to be from *Pseudomonas* spp., one of the main contaminants identified in the samples.

DISCUSSION

Surveys of the microbiological status of the home have largely concentrated on kitchens, particularly the bacteria which can be found in this environment. This is mainly due to the concern about transmission of bacteria which cause food poisoning [12, 13]. However, it should be noted that viruses play a major role in food related as well as non-food related infections; in fact recently published figures indicate that between 1992 and 1994, 27% of reported outbreaks of gastroenteritis were caused by small round structured viruses, compared with 32% caused by *Salmonella* spp. [14].

The presence of viruses or viral proteins have been demonstrated by a variety of techniques in various clinical settings but not in the domestic environment. Rotavirus has been isolated in day-care centres [3], the virus being found on surfaces during non-outbreak periods. In the clinical setting hepatitis B virus surface antigen has been found on surfaces within a dental surgery [15], and on forms in a clinical laboratory [16]. There are no reports of surveys which look for a wide range of viruses or body fluids which may contain viruses or viral proteins.

The demonstration of viruses or body fluids which may contain viruses would improve our understanding of the risks from surfaces contaminated with viruses and whether particular areas need special attention in terms of hygiene.

Haemoglobin, indicating the presence of blood and therefore possible contamination of surfaces with blood-borne viruses, was found on a range of surfaces; these included taps, washbasins, toilet bowls and seats. The Heamostix detection system is based on the peroxidase-like activity of haemoglobin which catalyses the reaction of cumene hydroperoxide and 3,3′,5,5′-tetramethyl benzidine. Some of these surfaces are frequently handled and could therefore play a part in viral transmission. There are simple and obvious explanations for these findings, e.g. contamination during teeth cleaning, washing cuts and abrasions and menstruation and possibly faeces containing blood. However it does highlight the fact that surfaces may remain 'soiled' for some time, and may not be thoroughly cleaned, and could therefore play a part in the transfer of blood-born viruses to susceptible individuals.

Amylase at levels > 5 U l^{-1} was found on 27·3 and 31·2% of surfaces in study 1 and 2 respectively. Many of the surfaces were handled frequently e.g. taps, door handles, or in contact with urine, e.g. toilet bowl, seat. Almost half (44·1 and 42·5%) of the telephone had levels > 5 U l^{-1}. This is likely to be from contami-

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nation by saliva and sweat, and may indicate areas which are infrequently or inadequately cleaned and may be a potential source of infection particularly from rhinovirus [7].

Failure to isolate any viruses by cell culture could be due to a variety of reasons, the most likely being: (i) at the time of sampling there was no virus on the surface, (ii) virus present on the surface had lost infectivity due to damage by cleaning agents or simply through desiccation, (iii) viruses did not survive in the transport medium, (iv) as suggested by Bryden (1992), the virions were not evenly distributed in the sample and if the titre were low it may be chance if a single dose of inoculum contains enough particles to infect the cell culture [17] or (v) cell culture is not suitable for isolation of viruses in this type of study because of bacterial and fungal contamination and low numbers of virus particles.

It should be remembered that viruses unlike bacteria do not multiply outside their host cells, therefore infectivity levels will start to fall as soon as the virus settles on the surface [3–5]. Removal of viruses by cleaning, or flushing of toilets has not been examined in this type of study and this must play a significant part in virus removal and survival.

The detection of enteroviral RNA from surfaces demonstrates that it is possible to use this technique for environmental monitoring. However the use of PCR for a large scale survey is relatively expensive and time consuming and not without its own problems. However it is very useful as demonstrated in this study for the detection of viral nucleic acid on surfaces, as it overcomes some of the problems of bacterial contamination when cell culture is used and may allow detection of non-culturable viruses such as the small round structured viruses. PCR does have advantages, in that one sample could be amplified, the nucleic acid sequenced and the sequence compared with DNA databases to determine the exact strain of virus.

The use of PCR to demonstrate environmental contamination of viruses represents an alternative to traditional cell culture techniques which may allow more information to be gathered on the risks in the environment. It may also allow us to follow the course of infections through family units and communities, by specifically identifying the causative agent for each infection.

Analysis of the microbiological hazards in the home requires information on the presence of hazards, i.e. type, number, survival, and position; routes of transmission; infective dose/susceptibility. This study along with others [18, 19] has provided some information on the presence of hazards in the home. However further work is required to demonstrate that the presence of these hazards does actually present a risk to health, i.e. actually linking the presence of viruses or bacteria on surfaces to occurrence of illness in the home and whether good hygiene practice can reduce the incidence of ill health in the home.

Currently no study has looked at all of these aspects in the domestic environment.

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REFERENCES

1. Samadi AR, Huq MI, Amed QS. Demonstration of rotavirus in handwashings of attendants of children with diarrhoea. BMJ 1983; **286**: 188.
2. Lew JF, Moe CL, et al. Astroviruses and adenoviruses associated with diarrhoea in children in day-care settings. J Infect Dis 1991; **164**: 673–8.
3. Keswick BH, Pickering LK, DuPont HL, Woodward WE. Survival and detection of rotavirus on environmental surfaces in day-care centres. Appl Env Micro 1983; **46**: 813–6.
4. Nerukar LS, West F, May M, Madden DL, Sever JL. Survival of herpes simplex in water specimens collected from hot tubs in spa facilities and on plastic surfaces. JAMA 1983; **22**: 3081–3.
5. Mbithi JN, Springthorpe S, Sattar SA. Effect of relative humidity and air temperature on the survival of hepatitis A virus on environmental surfaces. Appl Env Micro 1991; **57**: 1394–9.
6. Mahl MC, Sadler C. Virus survival on inanimate surfaces. Can J Microbiol 1975; **21**: 819–23.
7. Pancic F, Carpenter DC, Came PE. Role of infectious secretions in the transmission of rhinoviruses. J Clin Microbiol 1980; **12**: 567–71.
8. Saiki RK, Scharf S, Faloona F, et al. Enzymatic amplification of beta-globulin genomic sequences and restriction site analysis for the diagnosis of sickle-cell anaemia. Science 1985; **230**: 1350–4.
9. Gow JW, Behan WMH, Clements GB, Woodall C, Riding M, Behan PO. Enteroviral RNA sequences detected by polymerase chain reaction in muscle of patients with post-viral fatigue syndrome. BMJ 1991; **302**: 692–6.

10. Maniatis T, Fristch EF, Sambrook J. Molecular cloning: A laboratory manual. Cold Spring Harbour, 1982.
11. The DIG system user's guide for filter hybridisation. Boehringer Mannheim, 1995.
12. Finch JE, Prince J, Hawsworth M. A bacteriological survey of the domestic environment. J Appl Bact 1978; **45**: 357–64.
13. Scott E, Bloomfield SF, Barlow CG. An investigation of the microbial contamination in the home. J Hyg 1982; **89**: 279–93.
14. Djuretic T, Tall PG, Ryan MJ, Evans HS, Adak GK, Cowden JM. General outbreaks of infectious intestinal disease in England and Wales 1992–4. CDR 1996; **6**: R64–8.
15. Piazza M, Guadagnino V, Piciotto L, Borga G, Nappa S, Contamination by hepatitis B surface antigen in dental surgeries. BMJ 1987; **295**: 473–4.
16. Zachoval R, Frosner G, Hass D. Persistence of hepatitis B virus antigen in dried blood. Lancet 1981; i; 778.
17. Bryden AS. Isolation of enteroviruses and adenoviruses in continuous simian cell lines. Med Lab Sci 1992; **49**: 60–5.
18. Buemer RR, Te Giffel MC, Spoorenberg E, Roumbouts FM. *Listeria* species in domestic environment. Epidemiol Infect 1996; **117: 437–42.**
19. Van Reenenhoekstra ES, Samson RA, Verhoeff AP, van Wijen JH. Detection and identification of moulds in Dutch houses and non-industrial working environments. Grana 1991; **30**: 418–23.